abcam

ab300114

# Anti-ADNP antibody [EPR25434-4]

Recombinant | RabMAb | 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** | EPR25434-4 |
| **Purification technique** | Affinity purification Protein A |
| **Concentration** | 0.5445 mg/mL The concentration of this product may be batch-dependent<br>Batch concentration finder → |

## Reactivity data

## ChIP

### Not recommended

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

## WB

### Tested

| | |
|---|---|
| **Species** | Human |
| **Dilution info** | 1/1000 |
| **Notes** | - |

| | |
|---|---|
| **Species** | Mouse |
| **Dilution info** | 1/1000 |
| **Notes** | - |

| | |
|---|---|
| **Species** | Rat |
| **Dilution info** | 1/1000 |
| **Notes** | - |

## IHC-P

### Tested

| | |
|---|---|
| **Species** | Human |
| **Dilution info** | 1/100 |
| **Notes** | Perform heat-mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. |

| | |
|---|---|
| **Species** | Mouse |
| **Dilution info** | 1/100 |
| **Notes** | Perform heat-mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. |

| | |
|---|---|
| **Species** | Rat |
| **Dilution info** | 1/100 |
| **Notes** | Perform heat-mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. |

## IHC-Fr

### Tested

| | |
|---|---|
| **Species** | Mouse |
| **Dilution info** | 1/500 |
| **Notes** | - |

| | |
|---|---|
| **Species** | Rat |
| **Dilution info** | 1/500 |
| **Notes** | - |

### Expected

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

## 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** | - |

| | |
|---|---|
| **Species** | Mouse |
| **Dilution info** | 1/30 |
| **Notes** | - |

### Expected

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

# Target data

See full target information ADNP

| | |
|---|---|
| **Function** | May be involved in transcriptional regulation. May mediate some of the neuroprotective peptide VIP-associated effects involving normal growth and cancer proliferation. Positively modulates WNT-beta-catenin/CTNN1B signaling, acting by regulating phosphorylation of, and thereby stabilizing, CTNNB1. May be required for neural induction and neuronal differentiation. May be involved in erythroid differentiation (By similarity). |

## 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** | The ADNP protein also known as Activity-dependent neuroprotector homeobox is a multifunctional protein with a mass of about 123 kDa. ADNP is highly expressed in the brain but also present in other tissues including heart and kidney. It regulates neural development and synaptic plasticity by interacting with chromatin and |

| | |
|---|---|
| | participating in transcriptional activities. The protein plays a role in cell cycle regulation and is involved in neuroprotection against multiple insults. |
| **Biological function summary** | ADNP plays critical roles in the development of the nervous system and maintenance of neuronal function. As part of a chromatin remodeling complex known as SWI/SNF it modulates the expression of genes necessary for neurogenesis and neuronal survival. ADNP affects microtubule dynamics which are important for neuron structure and transport by regulating activity of associated proteins. These functions make ADNP influential in maintaining a healthy nervous system and protecting against neurodegenerative processes. |
| **Pathways** | ADNP is necessary for proper functioning in neuroprotective pathways and the regulation of gene expression pathways. It closely interacts with the proteins REST and HDAC2 in the regulation of transcription important for neuronal cell survival. The involvement in Wnt signaling pathway an important regulator of cell proliferation and differentiation highlights its importance in neural development and plasticity. ADNP also affects the activity of MAPK/ERK pathway which links it to the control of cell proliferation and survival. |
| **Associated diseases and disorders** | ADNP mutations or dysregulations have been connected to neurodevelopmental disorders such as Helsmoortel-Van der Aa syndrome and Alzheimer's disease. In relation to Helsmoortel-Van der Aa syndrome disruptions in ADNP lead to intellectual disability and developmental delays due to impaired neural growth. In Alzheimer's disease changes in ADNP expression affect the stability and function of tau a protein involved in neurofibrillary tangles. These interactions underline ADNP's significance in neuropsychiatric disorders and potential as a therapeutic target. |

## 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**

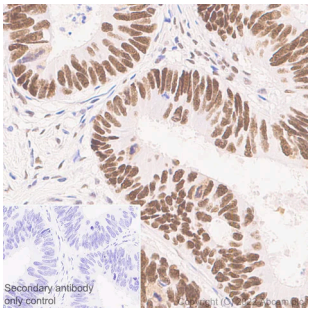
We do not recommend this combination. It is not covered by our product promise.

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.

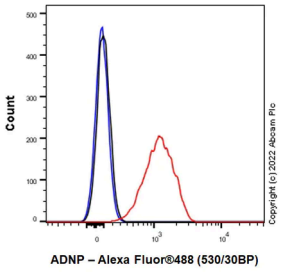
## 18 product images




### Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-ADNP antibody [EPR25434-4] (ab300114)

ADNP Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining of Human colon carcinoma tissue using rabbit Anti-ADNP antibody

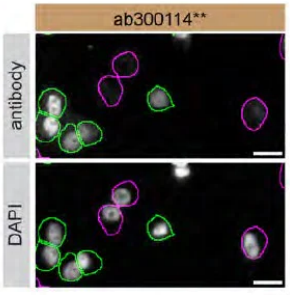
Immunohistochemical analysis of paraffin-embedded Human colon carcinoma tissue labelling ADNP with ab300114 at 1/100 dilution followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Nuclear staining on human colon carcinoma is observed (PMID: 27903678). The section was incubated with ab300114 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin. Secondary antibody only control: PBS was used instead of primary antibody followed by ready to use secondary antibody LeicaDS9800 (Bond™ Polymer Refine Detection). Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins.




### Flow Cytometry (Intracellular) - Anti-ADNP antibody [EPR25434-4] (ab300114)

ADNP Flow Cytometry (Intracellular) staining of HeLa (Human cervix adenocarcinoma epithelial cell) using rabbit Anti-ADNP antibody

Intracellular flow cytometric analysis of 4% paraformaldehyde fixed and 90% methnol permeabilized HeLa (human cervix adenocarcinoma epithelial cell) cells labelling ADNP with ab300114 at 1/500 dilution (Red) compared with a rabbit monoclonal IgG (ab172730) (Black) isotype control. Goat Anti-Rabbit IgG (Alexa Fluor® 488, ab150081) at 1/2000 dilution was used as the secondary antibody.



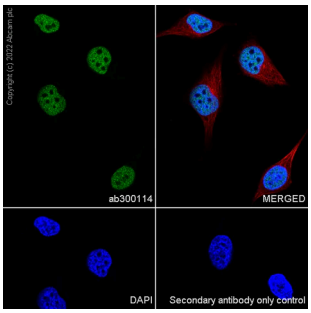

### Immunocytochemistry/ Immunofluorescence - Anti-ADNP antibody [EPR25434-4] (ab300114)

ADNP Immunocytochemistry/ Immunofluorescence staining of DMS 53 cells using rabbit Anti-ADNP antibody

ab300114 was shown to react with ADNP in wild-type DMS 53 cells in immunocytochemistry with loss of signal observed in a ADNP siRNA knockdown cell line. Wild-type and siRNA knockdown cells were mixed and pelleted at a 1:1 ratio on coverslips. The cells were fixed with 4% paraformaldehyde (15 min) then permeabilized with 0.1% Triton X-100 (10min) and then blocked with 1x PBS, 0.01% Triton X-100, 5% BSA, 5% NGS. The cells were then incubated with ab300114 at 1/500 dilution overnight at 4°C followed by a further incubation at room temperature for 1h with a goat anti-rabbit secondary antibody to (Alexa Fluor® 555) at 0.5 ug/ml. Acquisition of the green (wild-type), red (antibody staining) and far-red (siRNA knockdown) channels was performed. Representative grayscale images of the red channel are shown. Wild-type and siRNA knockdown cells are outlined with yellow and magenta dashed line, respectively. Schematic representation of the mosaic strategy used is shown on the bottom-right panel. Image was acquired with a Zeiss(LSM-880).

This data was provided by YCharOS Inc., an open science company with the mission of characterizing commercially available antibody reagents for all human proteins. Abcam and

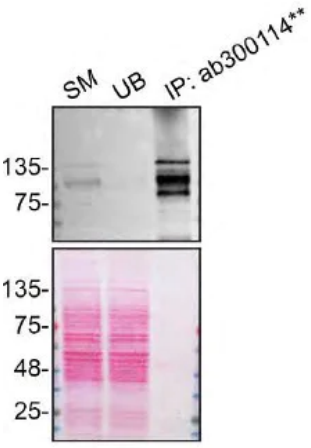
YCharOS are working together to help address the reproducibility crisis by enabling the life science community to better evaluate commercially available antibodies.




## Immunocytochemistry - Anti-ADNP antibody [EPR25434-4] (ab300114)

ADNP Immunocytochemistry staining of HeLa (Human cervix adenocarcinoma epithelial cell) using rabbit Anti-ADNP antibody

Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeilized HeLa (human cervix adenocarcinoma epithelial cell) cells lebelling ADNP with ab300114 at 1/50 dilution, followed by ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed antibody at 1/1000 dilution (Green). Confocal image showing nuclear staining in HeLa cell line. Tubuline was stained with ab195889 Anti-alpha Tubulin mouse monoclonal antibody - Microtubule Marker (Alexa Fluor® 594) at 1/200 dilution (Red). The Nuclear counterstain was DAPI (Blue). Secondary antibody only control: PBS was used instead of ab300114 followed by preabsorbed secondary antibody (ab150081) Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) at 1/1000 dilution.



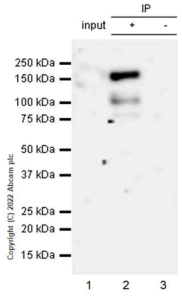

## Immunoprecipitation - Anti-ADNP antibody [EPR25434-4] (ab300114)

Immunoprecipitation of ADNP in DMS 53 cells. Lysates were prepared and immunoprecipitation was performed using 2 µg of ab300114 pre-coupled to Protein A beads. Samples were then washed and processed for western blot.

This data was provided by YCharOS Inc., an open science company with the mission of characterizing commercially available antibody reagents for all human proteins. Abcam and YCharOS are working together to help address the reproducibility crisis by enabling the life science community to better evaluate commercially available antibodies.
All lanes:
Immunoprecipitation - Anti-ADNP antibody [EPR25434-4] (ab300114) at 2 µg

Lanes 1 and 3:
DMS 53 cells

Lane 2:
Unbound fraction: Rabbit monoclonal IgG instead of ab300114 in DMS 53 cells



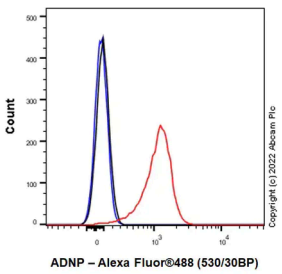

## Immunoprecipitation - Anti-ADNP antibody [EPR25434-4] (ab300114)

ADNP was immunoprecipitated from 0.35 mg HeLa (human cervix adenocarcinoma epithelial cell) whole cell lysate with ab300114 at 1/30 dilution. Western blot was performed on the immunoprecipitate using ab300114 at 1/1000 dilution. VeriBlot for IP secondary antibody(HRP) (ab131366) was used at 1/5000 dilution. Lane 1 (Input): HeLa (human cervix adenocarcinoma epithelial cell) whole cell lysate 10 µg. Lane 2 (+): ab300114 IP in HeLa whole cell lysate. Lane 3 (-): Rabbit monoclonal IgG (ab172730) instead of ab300114 in HeLa whole cell lysate. Blocking and dilution buffer and concentration: 5% NFDM/TBST. Exposure time: 84 seconds
All lanes:
Immunoprecipitation - Anti-ADNP antibody [EPR25434-4] (ab300114) at 1/30 dilution

Lane 1:
HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysate at 10 µg

Lane 2:
ab300114 IP in HeLa whole cell lysate

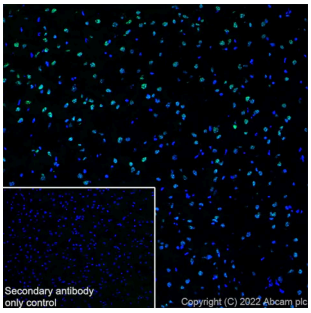
Lane 3:
Rabbit monoclonal IgG (ab172730) instead of ab300114 in HeLa whole cell lysate




## Flow Cytometry (Intracellular) - Anti-ADNP antibody [EPR25434-4] (ab300114)

ADNP Flow Cytometry (Intracellular) staining of NIH/3T3 (Mouse embryonic fibroblast) cells using rabbit Anti-ADNP antibody

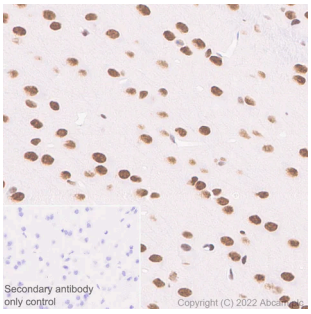
Intracellular flow cytometric analysis of 4% paraformaldehyde fixed 90% methanol permeabilized NIH/3T3 (mouse embryonic fibroblast) cells labelling ADNP with ab300114 at 1/500 dilution (Red) compared with a rabbit monoclonal IgG (ab172730) (Black) isotype control. A Goat Anti-Rabbit IgG (Alexa Fluor® 488, ab150081) at 1/2000 dilution was used as the secondary antibody.




## Immunohistochemistry (Frozen sections) - Anti-ADNP antibody [EPR25434-4] (ab300114)

ADNP Immunohistochemistry (Frozen sections) staining of Mouse cerebrum tissue using rabbit Anti-ADNP antibody

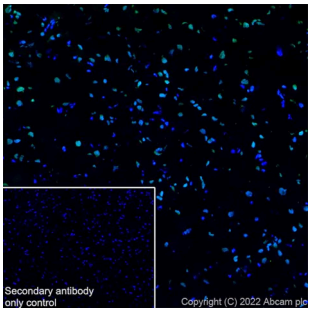
Immunohistochemical analysis of 4% PFA-fixed, 0.2% Triton X-100 permeabilized frozen mouse cerebrum (fresh) tissue labeling ADNP with ab300114 at 1/500 dilution followed by ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 dilution (Green). Positive staining on mouse cerebrum is observed. The nuclear counterstain was DAPI (Blue). Secondary antibody control: PBS was used instead of ab300114 followed by preadsorbed secondary antibody (ab150081) Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) at 1/1000 dilution.




## Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-ADNP antibody [EPR25434-4] (ab300114)

ADNP Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining of Mouse cerebrum tissue using rabbit Anti-ADNP antibody

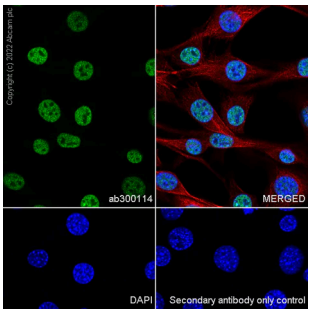
Immunohistochemical analysis of paraffin-embedded Mouse cerebrum tissue labelling ADNP with ab300114 at 1/100 dilution followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection) was used. Nuclear staining on mouse cerebrum is observed. The section was incubated with ab300114 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument Counterstained with Hematoxylin. Secondary antibody only control: PBS was used instead of primary antibody followed by ready to use secondary antibody LeicaDS9800 (Bond™ Polymer Refine Detection). Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins.




## Immunohistochemistry (Frozen sections) - Anti-ADNP antibody [EPR25434-4] (ab300114)

ADNP Immunohistochemistry (Frozen sections) staining of Rat cerebrum tissue using rabbit Anti-ADNP antibody

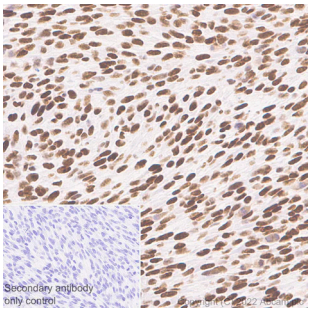
Immunohistochemical analysis of 4% PFA-fixed, 0.2% Triton X-100 permeabilized frozen rat cerebrum (fresh) tissue labeling ADNP with ab300114 at 1/500 dilution followed by ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 dilution (Green). Positive staining on rat cerebrum is observed. The nuclear counterstain was DAPI (Blue). PBS was used instead of ab300114 followed by preabsorbed secondary antibody (ab150081) Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) at 1/1000 dilution.




## Immunocytochemistry - Anti-ADNP antibody [EPR25434-4] (ab300114)

ADNP Immunocytochemistry staining of NIH/3T3 (Mouse embryonic fibroblast) cells using rabbit Anti-ADNP antibody

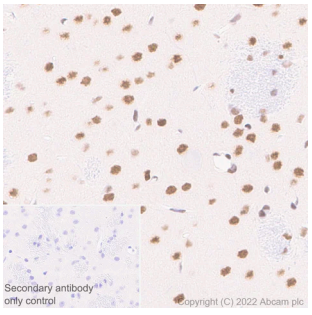
Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeilized NIH/3T3 (mouse embryonic fibroblast) cells lebelling ADNP with ab300114 at 1/50 (11.22 µg/ml) dilution, followed by ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed antibody at 1/1000 (2µg/mL) dilution (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 µg/ml) dilution (Red). The Nuclear counterstain was DAPI (Blue). Secondary antibody control: PBS was used instead of ab300114 followed by preabsorbed secondary antibody (ab150081) Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) at 1/1000 (2 µg/mL) dilution.




## Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-ADNP antibody [EPR25434-4] (ab300114)

ADNP Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining of Mouse glioblastoma tissue using rabbit Anti-ADNP antibody

Immunohistochemical analysis of paraffin-embedded mouse glioblastoma tissue labelling ADNP with ab300114 at 1/100 dilution followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Nuclear staining on mouse glioblastoma is observed. The section was incubated with ab300114 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin. Secondary antibody only control: PBS was used instead of primary antibody followed by ready to use secondary antibody LeicaDS9800 (Bond™ Polymer Refine Detection). Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins.

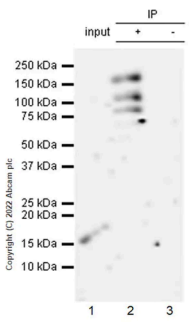



## Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-ADNP antibody [EPR25434-4] (ab300114)

ADNP Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining of Rat cerebrum tissue using rabbit Anti-ADNP antibody

Immunohistochemical analysis of paraffin-embedded rat cerebrum tissue labelling ADNP with ab300114 at 1/100 dilution followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Nuclear staining on rat cerebrum is observed. The section was incubated with ab300114 for 30 mins at room temperature. The immunostaining was performed on a Leica

Biosystems BOND® RX instrument. Counterstained with Hematoxylin. Secondary antibody only control: PBS was used instead of primary antibody followed by ready to use secondary antibody LeicaDS9800 (Bond™ Polymer Refine Detection). Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins.



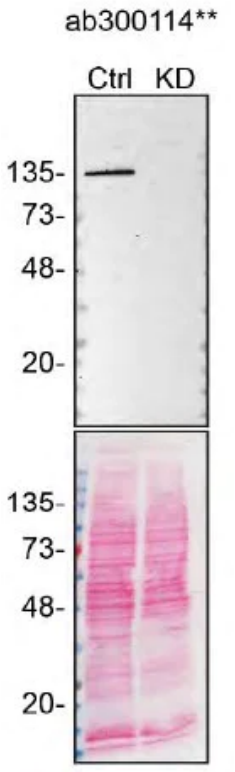

## Immunoprecipitation - Anti-ADNP antibody [EPR25434-4] (ab300114)

ADNP was immunoprecipitated from 0.35 mg NIH/3T3 (mouse embryonic fibroblast) whole cell lysate with ab300114 at 1/30 dilution. Western blot was performed on the immunoprecipitate using ab300114 at 1/1000 dilution. VeriBlot for IP secondary antibody(HRP) (ab131366) was used at 1/5000 dilution. Lane 1 (Input): NIH/3T3 (mouse embryonic fibroblast) whole cell lysate 10 µg. Lane 2 (+): ab300114 IP in NIH/3T3 whole cell lysate. Lane 3 (-): Rabbit monoclonal IgG (ab172730) instead of ab300114 in NIH/3T3 whole cell lysate. Blocking and dilution buffer and concentration: 5% NFDM/TBST. Exposure time: 3 minutes.
All lanes:
Immunoprecipitation - Anti-ADNP antibody [EPR25434-4] (ab300114) at 1/30 dilution

Lane 1:
NIH/3T3 (mouse embryonic fibroblast) whole cell lysate at 10 µg

Lane 2:
ab300114 IP in NIH/3T3 whole cell lysate

Lane 3:
Rabbit monoclonal IgG (ab172730) instead of ab300114 in NIH/3T3 whole cell lysate



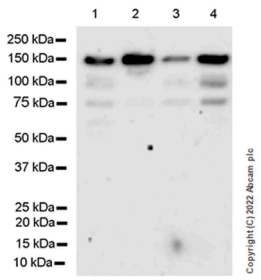

## Western blot - Anti-ADNP antibody [EPR25434-4] (ab300114)

ab300114 was shown to react with ADNP in wild-type DMS 53 cells in Western blot with loss of signal observed in a ADNP siRNA knockdown cell line. Cell lysates from wild-type DMS 53 transfected with either scrambled siRNA or ADNP siRNA were subjected to SDS-PAGE. Membranes were blocked in 5% milk in TBST for 1 hr before incubation with ab300114 overnight at 4 °C at a 1/10000 dilution. Blots were incubated with secondary antibodies at 0.2 µg/mL before imaging.

This data was provided by YCharOS Inc., an open science company with the mission of characterizing commercially available antibody reagents for all human proteins. Abcam and YCharOS are working together to help address the reproducibility crisis by enabling the life science community to better evaluate commercially available antibodies.
All lanes:
Western blot - Anti-ADNP antibody [EPR25434-4] (ab300114) at 1/10000 dilution

Lane 1:
Wild-type DMS 53 transfected with scrambled siRNA control lysate at 30 µg

Lane 2:
DMS 53 transfected with siRNA specifically targeting ADNP cell lysate at 30 µg




## Western blot - Anti-ADNP antibody [EPR25434-4] (ab300114)

ADNP Western blot staining using rabbit Anti-ADNP antibody

Blocking and dilution buffer and concentration: 5% NFDM/TBST. Lysates should be made freshly and used in WB immediately to minimize protein degradation.
All lanes:
Western blot - Anti-ADNP antibody [EPR25434-4] (ab300114) at 1/1000 dilution

Lane 1:
C6 (Rat glial tumor glial cell) whole cell lysate at 20 µg

Lane 2:
U-87 MG (Human glioblastoma-astrocytoma epithelial cell) whole cell lysate at 20 µg

Lane 3:
SH-SY5Y (Human neuroblastoma epithelial cell) whole cell lysate at 20 µg

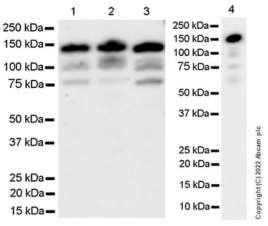
Lane 4:
Neuro-2a (Mouse neuroblastoma neuroblast) whole cell lysate at 20 µg

Secondary

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

Exposure time: 3s

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## Western blot - Anti-ADNP antibody [EPR25434-4] (ab300114)

ADNP Western blot staining using rabbit Anti-ADNP antibody

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

Exposure time:

Lanes 1-3: 114 seconds; Lane 4: 59 seconds.

Lysates should be made freshly and used in WB immediately to minimize protein degradation.

All lanes:
Western blot - Anti-ADNP antibody [EPR25434-4] (ab300114) 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

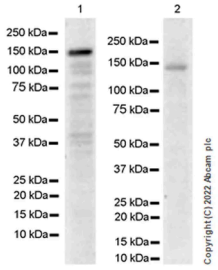
Lane 4:
HeLa (human cervix adenocarcinoma epithelial cell) whole cell lysate at 20 µg

Secondary

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

Predicted band size: 124 kDa

Observed band size: 140 kDa




## Western blot - Anti-ADNP antibody [EPR25434-4] (ab300114)

ADNP Western blot staining using rabbit Anti-ADNP antibody

Blocking and dilution buffer and concentration: 5% NFDM/TBST. Lysates should be made freshly and used in WB immediately to minimize protein degradation. We used fresh mouse brain tissue lysate. This blot was developed using a higher sensitivity ECL substrate.
All lanes:
Western blot - Anti-ADNP antibody [EPR25434-4] (ab300114) at 1/1000 dilution

Lane 1:
Mouse brain tissue lysate at 20 µg

Lane 2:
Rat brain tissue lysate at 20 µg

Secondary

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

Exposure time: 3s

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