

Anti-KCC2 antibody [EPR24203-85] - BSA and Azide free ab283588

Recombinant RabMAb

19 Images

Overview

Product name	Anti-KCC2 antibody [EPR24203-85] - BSA and Azide free
Description	Rabbit monoclonal [EPR24203-85] to KCC2 - BSA and Azide free
Host species	Rabbit
Tested applications	Suitable for: IHC-Fr, WB, IP, IHC-P, ICC/IF, Flow Cyt (Intra)
Species reactivity	Reacts with: Mouse, Rat, Human
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: Mouse brain, Mouse spinal cord, Rat brain, Rat spinal cord, Human spinal cord lysates. IHC-P: Human cerebrum, Mouse cerebrum and Rat cerebrum tissue. IHC-Fr: Mouse cerebrum and Rat cerebrum tissue. ICC/IF: Mouse primary neural/glia and Rat primary neural/glia cells. IP: Mouse brain and Rat brain tissue lysate. Flow Cyt: Mouse and Rat primary neuron cells.
General notes	ab283588 is the carrier-free version of ab259969 .

Our **carrier-free** antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for increased conjugation efficiency.

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.

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.

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 production

For more information [see here](#).

Our RabMAb[®] technology is a patented hybridoma-based technology for making rabbit

monoclonal antibodies. For details on our patents, please refer to [RabMAb® patents](#).

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C.
Storage buffer	pH: 7.20 Constituent: 100% PBS
Carrier free	Yes
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR24203-85
Isotype	IgG

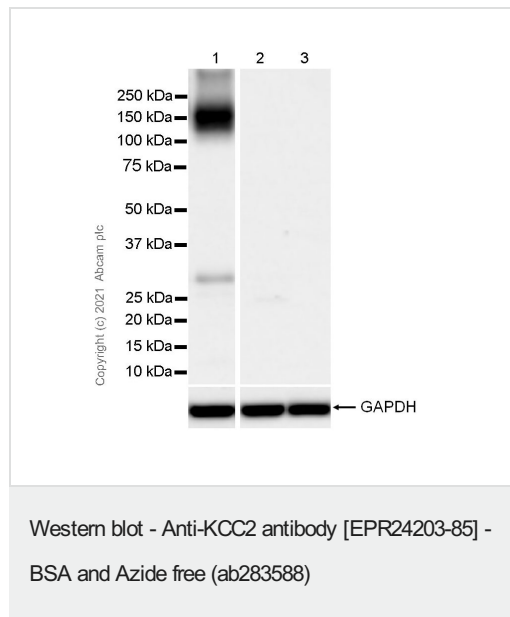
Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab283588 in the following tested applications. The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
IHC-Fr		Use at an assay dependent concentration.
WB		Use at an assay dependent concentration. Predicted molecular weight: 126 kDa.
IP		Use at an assay dependent concentration.
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
ICC/IF		Use at an assay dependent concentration.
Flow Cyt (Intra)		1/500.

Target

Function	Mediates electroneutral potassium-chloride cotransport in mature neurons. Transport occurs under isotonic conditions, but is activated 20-fold by cell swelling. Important for Cl(-) homeostasis in neurons.
Tissue specificity	Brain specific. Detected in neuronal cells.
Sequence similarities	Belongs to the SLC12A transporter family.
Cellular localization	Membrane.



All lanes : Anti-KCC2 antibody [EPR24203-85] ([ab259969](#)) at 1/1000 dilution

Lane 1 : Mouse brain tissue lysate

Lane 2 : Mouse kidney tissue lysate

Lane 3 : Mouse liver tissue lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) ([ab97051](#)) at 1/100000 dilution (Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated)

Predicted band size: 126 kDa

Observed band size: 140 kDa

This data was developed using [ab259969](#), the same antibody clone in a different buffer formulation.

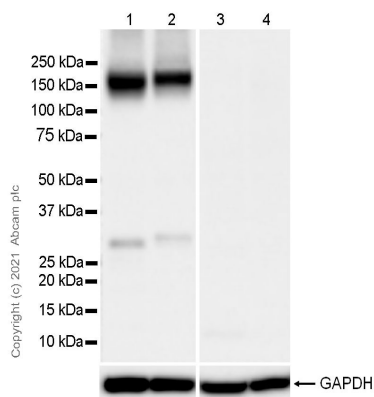
Blocking and diluting buffer and concentration: 5% NFDm/TBST

Samples are non-boiled as boiling may cause protein aggregates.

The observed 30 KD band is like the KCC2 C terminus fragment which has been described in the literature (PMID: 22854961).

Negative control: Kidney, liver (PMID: 10212246).

Exposure time: 6 seconds



Western blot - Anti-KCC2 antibody [EPR24203-85] - BSA and Azide free (ab283588)

All lanes : Anti-KCC2 antibody [EPR24203-85] ([ab259969](#)) at 1/1000 dilution

Lane 1 : Rat brain tissue lysate at 20 µg

Lane 2 : Rat spinal cord tissue lysate at 20 µg

Lane 3 : Rat kidney tissue lysate

Lane 4 : Rat liver tissue lysate

Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) ([ab97051](#)) at 1/100000 dilution (Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated)

Predicted band size: 126 kDa

Observed band size: 140 kDa

This data was developed using [ab259969](#), the same antibody clone in a different buffer formulation.

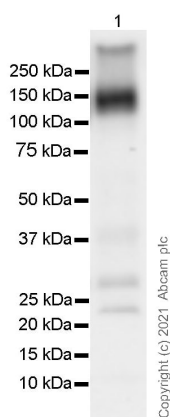
Blocking and diluting buffer and concentration: 5% NFD/MTBST

Samples are non-boiled as boiling may cause protein aggregates.

The observed 30 KD band is like the KCC2 C terminus fragment which has been described in the literature (PMID: 22854961).

Negative control: Kidney, liver (PMID: 10212246).

Exposure time: 6 seconds



Western blot - Anti-KCC2 antibody [EPR24203-85] - BSA and Azide free (ab283588)

Anti-KCC2 antibody [EPR24203-85] ([ab259969](#)) at 1/1000 dilution + Mouse spinal cord tissue lysate at 20 µg

Secondary

Goat Anti-Rabbit IgG H&L (HRP) ([ab97051](#)) at 1/100000 dilution (Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated)

Predicted band size: 126 kDa

Observed band size: 140 kDa

This data was developed using [ab259969](#), the same antibody clone in a different buffer formulation.

Blocking and diluting buffer and concentration: 5% NFD/MTBST

Samples are non-boiled as boiling may cause protein aggregates.

The observed 23, 30 KD bands are like the KCC2 C terminus fragment and the observed 300 KD band is the dimer KCC2, as described in the literature (PMID: 22854961).

Exposure time: 26 seconds

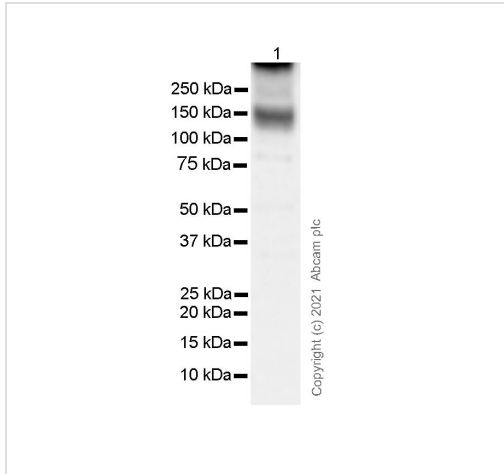
Anti-KCC2 antibody [EPR24203-85] ([ab259969](#)) at 1/1000 dilution + Human spinal cord at 20 µg

Secondary

Goat Anti-Rabbit IgG (HRP) with minimal cross-reactivity with human IgG at 1/5000 dilution

Predicted band size: 126 kDa

Observed band size: 140 kDa



Western blot - Anti-KCC2 antibody [EPR24203-85] - BSA and Azide free ([ab283588](#))

This data was developed using [ab259969](#), the same antibody clone in a different buffer formulation.

Blocking and diluting buffer and concentration: 5% NFDm/TBST

Samples are non-boiled as boiling may cause protein aggregates.

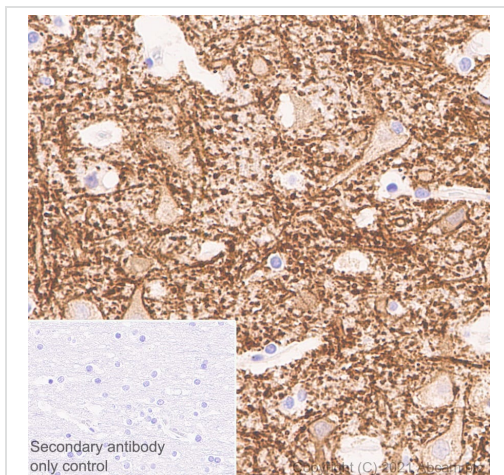
Exposure time: 15 seconds

This data was developed using [ab259969](#), the same antibody clone in a different buffer formulation.

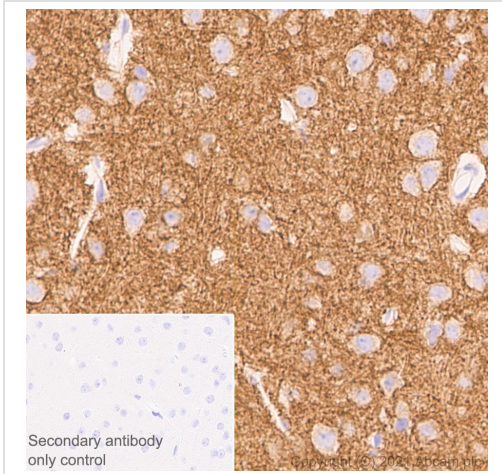
Immunohistochemical analysis of paraffin-embedded human cerebrum tissue labelling KCC2 with [ab259969](#) at 1/5000 dilution (0.111 µg/ml) followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Positive staining on human cerebrum. The section was incubated with [ab259969](#) for 30 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-KCC2 antibody [EPR24203-85] - BSA and Azide free ([ab283588](#))



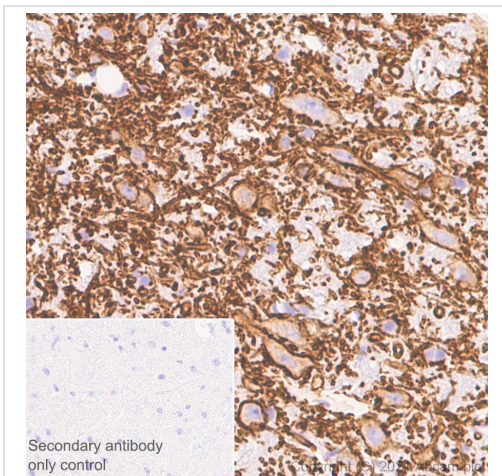
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-KCC2 antibody [EPR24203-85] - BSA and Azide free (ab283588)

This data was developed using [ab259969](#), the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of paraffin-embedded mouse cerebrum tissue labelling KCC2 with [ab259969](#) at 1/5000 dilution (0.111 µg/ml) followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Positive staining on mouse cerebrum (PMID: 17715129). The section was incubated with [ab259969](#) 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



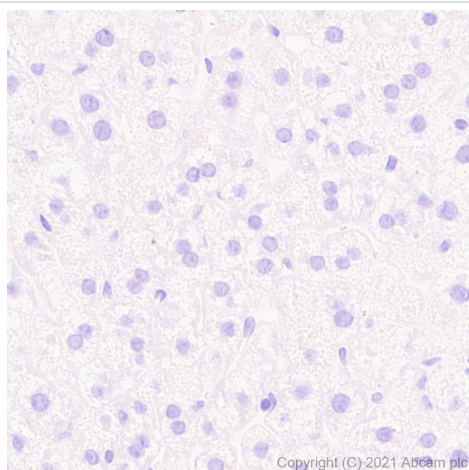
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-KCC2 antibody [EPR24203-85] - BSA and Azide free (ab283588)

This data was developed using [ab259969](#), the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of paraffin-embedded Rat cerebrum tissue labelling KCC2 with [ab259969](#) at 1/5000 dilution (0.111 µg/ml) followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Positive staining on rat cerebrum. The section was incubated with [ab259969](#) 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-KCC2 antibody [EPR24203-85] - BSA and Azide free (ab283588)

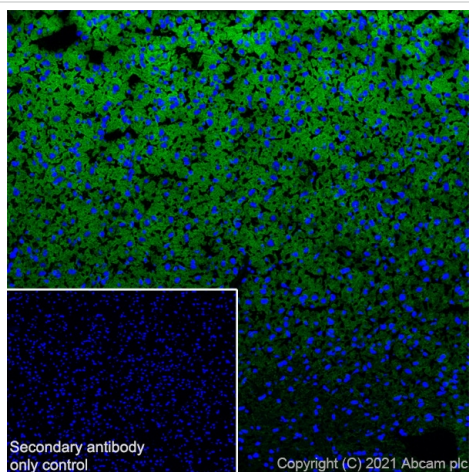
This data was developed using [ab259969](#), the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of paraffin-embedded mouse liver tissue labelling KCC2 with [ab259969](#) at 1/5000 dilution (0.111 µg/ml) followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). The section was incubated with [ab259969](#) for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin.

Negative control: almost no staining on mouse liver.

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

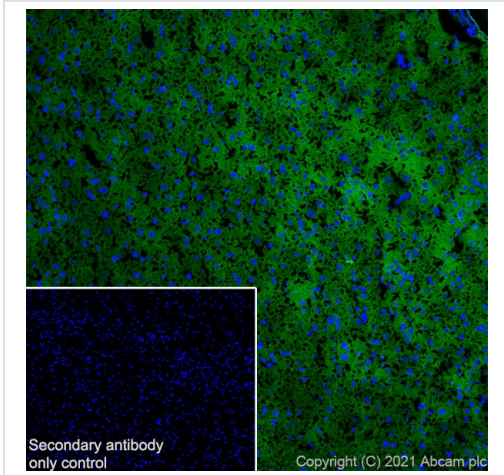


Immunohistochemistry (Frozen sections) - Anti-KCC2 antibody [EPR24203-85] - BSA and Azide free (ab283588)

This data was developed using [ab259969](#), the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of 4% PFA-fixed, 0.2% Triton X-100 permeabilized frozen mouse cerebrum (fresh) tissue labeling KCC2 with [ab259969](#) at 1/100 dilution (5.55 µg/ml) followed by [ab150081](#) Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 dilution (2 µg/ml)(Green). Positive staining on mouse cerebrum is observed. The nuclear counterstain was DAPI (Blue).

Secondary antibody control: Secondary antibody is [ab150081](#) Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 dilution (2 µg/ml).

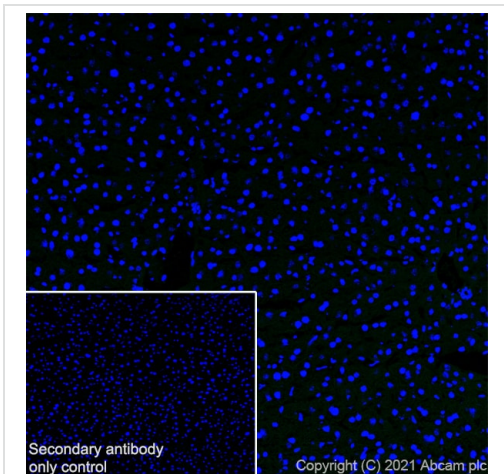


Immunohistochemistry (Frozen sections) - Anti-KCC2 antibody [EPR24203-85] - BSA and Azide free (ab283588)

This data was developed using [ab259969](#), the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of 4% PFA-fixed, 0.2% Triton X-100 permeabilized frozen rat cerebrum (fresh) tissue labeling KCC2 with [ab259969](#) at 1/100 (5.55 µg/ml) dilution followed by [ab150081](#) Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 dilution (2 µg/ml)(Green). Positive staining on rat cerebrum is observed. The nuclear counterstain was DAPI (Blue).

Secondary antibody control: Secondary antibody is [ab150081](#) Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 dilution (2 µg/ml).



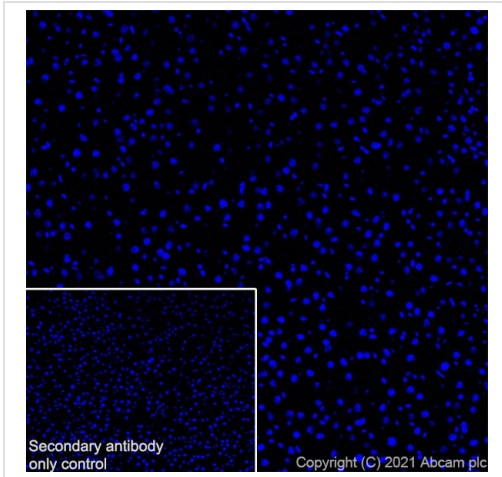
Immunohistochemistry (Frozen sections) - Anti-KCC2 antibody [EPR24203-85] - BSA and Azide free (ab283588)

This data was developed using [ab259969](#), the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of 4% PFA-fixed, 0.2% Triton X-100 permeabilized frozen mouse liver(fresh) tissue labeling KCC2 with [ab259969](#) at 1/100 dilution (5.55 µg/ml) followed by [ab150081](#) Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 dilution (2 µg/ml)(Green). The nuclear counterstain was DAPI (Blue). No staining on mouse liver.

Negative control: liver (PMID:17715129) is observed.

Secondary antibody control: Secondary antibody is [ab150081](#) Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 dilution (2 µg/ml).



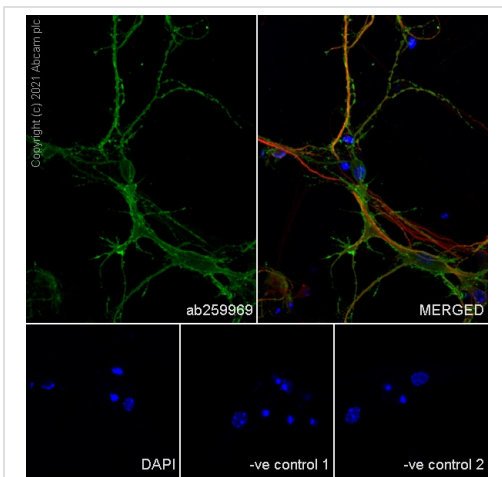
Immunohistochemistry (Frozen sections) - Anti-KCC2 antibody [EPR24203-85] - BSA and Azide free (ab283588)

This data was developed using [ab259969](#), the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of 4% PFA-fixed, 0.2% Triton X-100 permeabilized frozen rat liver (fresh) tissue labeling KCC2 with [ab259969](#) at 1/100 dilution (5.55 µg/ml) followed by [ab150081](#) Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 dilution (2 µg/ml)(Green). The nuclear counterstain was DAPI (Blue). No staining on rat liver.

Negative control: liver (PMID:17715129) is observed.

Secondary antibody control: Secondary antibody is [ab150081](#) Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 dilution (2 µg/ml).



Immunocytochemistry/ Immunofluorescence - Anti-KCC2 antibody [EPR24203-85] - BSA and Azide free (ab283588)

This data was developed using [ab259969](#), the same antibody clone in a different buffer formulation.

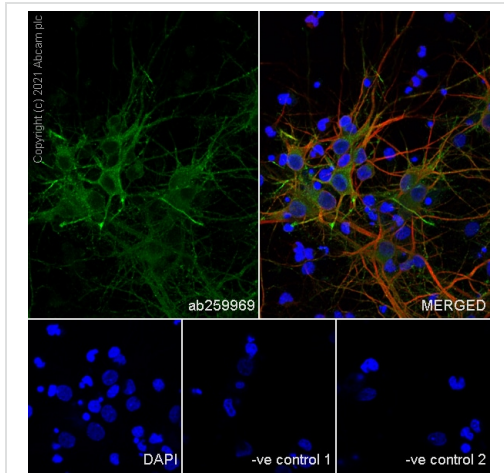
Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeabilized mouse primary neural/glia cells labelling KCC2 with [ab259969](#) at 1/200 dilution (2.775 µg/ml), followed by [ab150081](#) Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed antibody at 1/1000 dilution (2 µg/ml)(Green). [ab11267](#) Anti-MAP2 mouse monoclonal antibody at 1/500 dilution (4 µg/ml) followed by [ab150120](#) Goat Anti-Mouse IgG H&L (Alexa Fluor® 594) antibody at 1/1000 dilution (2 µg/ml) was used to counterstain tubulin (Red). The Nuclear counterstain was DAPI (Blue).

Confocal image showing positive staining in mouse primary neuron. Confocal scanning Z step was set as 0.3 µm followed by image processing with maximum Z projection.

Secondary antibody only control: Secondary antibody is [ab150081](#) Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 dilution (2 µg/ml).

Negative control 1: **ab150120** Goat Anti-Mouse IgG H&L (Alexa Fluor® 594) antibody at 1/1000 dilution (2 µg/ml).

Negative control 2: **ab11267** Anti-MAP2 mouse monoclonal antibody was used to counterstain tubulin at 1/500 dilution (4 µg/ml).



Immunocytochemistry/ Immunofluorescence - Anti-KCC2 antibody [EPR24203-85] - BSA and Azide free (ab283588)

This data was developed using **ab259969**, the same antibody clone in a different buffer formulation.

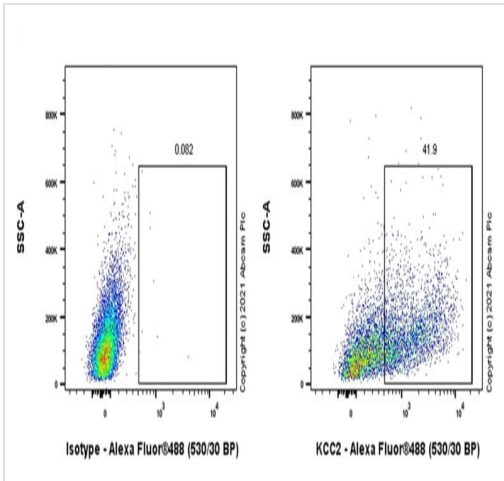
Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeabilized rat primary neural/glia cells labelling KCC2 with **ab259969** at 1/200 dilution (2.775 µg/ml), followed by **ab150081** Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed antibody at 1/1000 dilution (2 µg/ml)(Green). **ab11267** Anti-MAP2 mouse monoclonal antibody at 1/500 dilution (4 µg/ml) followed by **ab150120** Goat Anti-Mouse IgG H&L (Alexa Fluor® 594) antibody at 1/1000 dilution (2 µg/ml) was used to counterstain tubulin (Red). The Nuclear counterstain was DAPI (Blue).

Confocal image showing positive staining in rat primary neuron. Confocal scanning Z step was set as 0.3 µm followed by image processing with maximum Z projection.

Secondary antibody only control: Secondary antibody is **ab150081** Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 dilution (2 µg/ml).

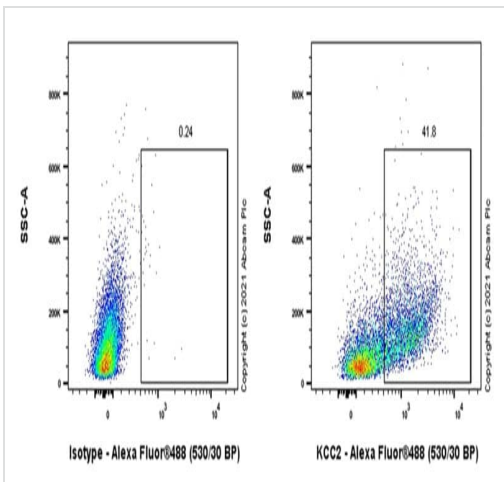
Negative control 1: **ab150120** Goat Anti-Mouse IgG H&L (Alexa Fluor® 594) antibody at 1/1000 dilution (2 µg/ml).

Negative control 2: **ab11267** Anti-MAP2 mouse monoclonal antibody was used to counterstain tubulin at 1/500 dilution (4 µg/ml).



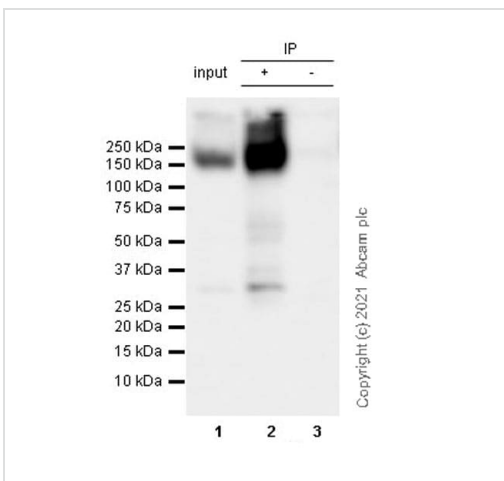
Flow Cytometry (Intracellular) - Anti-KCC2 antibody [EPR24203-85] - BSA and Azide free (ab283588)

Flow cytometric (intracellular) analysis of 4% paraformaldehyde-fixed Mouse primary neuron cells labelling KCC2 with **ab259969** at 1/500 dilution (Right) compared with Isotype control Rabbit monoclonal IgG (**ab172730**) (Left). Goat Anti-Rabbit IgG Alexa Fluor® 488 (**ab150081**) was used as the secondary antibody.



Flow Cytometry (Intracellular) - Anti-KCC2 antibody [EPR24203-85] - BSA and Azide free (ab283588)

Flow cytometric (intracellular) analysis of 4% paraformaldehyde-fixed Rat primary neuron cells labelling KCC2 with **ab259969** at 1/500 dilution (Right) compared with Isotype control Rabbit monoclonal IgG (**ab172730**) (Left). Goat Anti-Rabbit IgG Alexa Fluor® 488 (**ab150081**) at 1/2000 dilution was used as the secondary antibody.



Immunoprecipitation - Anti-KCC2 antibody [EPR24203-85] - BSA and Azide free (ab283588)

This data was developed using **ab259969**, the same antibody clone in a different buffer formulation.

KCC2 was immunoprecipitated from 0.35 mg mouse brain tissue lysate with **ab259969** at 1/30 dilution (2µg in 0.35mg lysates).

Western blot was performed on the immunoprecipitate using **ab259969** at 1/1000 dilution. VeriBlot for IP secondary antibody (HRP) (**ab131366**) was used at 1/5000 dilution.

Lane 1: Mouse brain tissue lysate 10µg

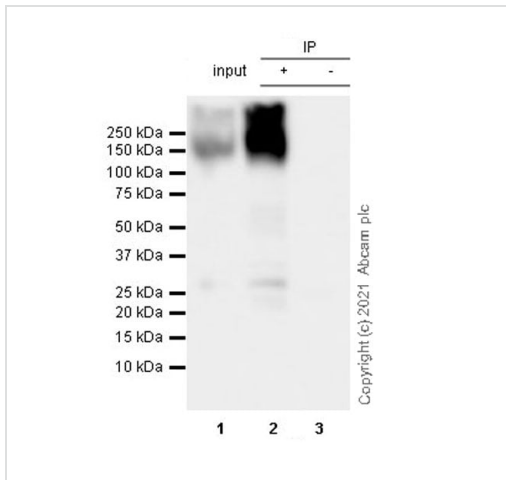
Lane 2: **ab259969** IP in Mouse brain tissue lysate

Lane 3: Rabbit monoclonal IgG (**ab172730**) instead of **ab259969** in mouse brain tissue lysate

Blocking and dilution buffer and concentration: 5% NFD/MTBST.

Exposure time: 3 seconds

The observed 30 KD band is like the KCC2 C terminus fragment and the observed 300 KD band is the dimer KCC2, as described in the literature (PMID: 22854961).



Immunoprecipitation - Anti-KCC2 antibody
[EPR24203-85] - BSA and Azide free (ab283588)

This data was developed using [ab259969](#), the same antibody clone in a different buffer formulation.

KCC2 was immunoprecipitated from 0.35 mg rat brain tissue lysate with [ab259969](#) at 1/30 dilution (2µg in 0.35mg lysates). Western blot was performed on the immunoprecipitate using [ab259969](#) at 1/1000 dilution. VeriBlot for IP secondary antibody (HRP) ([ab131366](#)) was used at 1/5000 dilution.

Lane 1: Rat brain tissue lysate 10µg

Lane 2: [ab259969](#) IP in Rat brain tissue lysate

Lane 3: Rabbit monoclonal IgG ([ab172730](#)) instead of [ab259969](#) in rat brain tissue lysate

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

Exposure time: 3 seconds

The observed 30 KD band is like the KCC2 C terminus fragment and the observed 300 KD band is the dimer KCC2, as described in the literature (PMID: 22854961).

Why choose a recombinant antibody?

- Research with confidence**
Consistent and reproducible results
- Long-term and scalable supply**
Recombinant technology
- Success from the first experiment**
Confirmed specificity
- Ethical standards compliant**
Animal-free production

Anti-KCC2 antibody [EPR24203-85] - BSA and Azide free (ab283588)

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