abcam

ab259969

Anti-KCC2 antibody [EPR24203-85]

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	EPR24203-85
Purification technique	Affinity purification Protein A
Concentration	0.512 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

ICC/IF

Tested

Species	Mouse
Dilution info	1/200
Notes	-

Species	Rat

Dilution info	1/200
Notes	-

Expected

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

IP

Tested

Species	Mouse
Dilution info	1/30
Notes	-

Species	Rat
Dilution info	1/30
Notes	-

Expected

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

WB

Tested

Species	Mouse
Dilution info	1/1000
Notes	-

Species	Rat
Dilution info	1/1000

Notes	-

Species	Human
Dilution info	1/1000
Notes	-

IHC-Fr

Tested

Species	Mouse
Dilution info	1/100
Notes	-

Species	Rat
Dilution info	1/100
Notes	-

Expected

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

Flow Cyt (Intra)

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	-

IHC-P

Tested

Species	Mouse
Dilution info	1/5000
Notes	-

Species	Rat
Dilution info	1/5000
Notes	-

Species	Human
Dilution info	1/5000
Notes	-

Target data

See full target information SLC12A5

Function	Mediates electroneutral potassium-chloride cotransport in mature neurons and is required for neuronal Cl(-) homeostasis (PubMed:12106695). As major extruder of intracellular chloride, it establishes the low neuronal Cl(-) levels required for chloride influx after binding of GABA-A and glycine to their receptors, with subsequent hyperpolarization and neuronal inhibition (By similarity). Involved in the regulation of dendritic spine formation and maturation (PubMed:24668262).

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	KCC2 also known as the potassium-chloride cotransporter 2 or SLC12A5 is an important neuronal protein. This transporter weighs approximately 125 kDa and is predominantly found in the central nervous system especially in neurons. KCC2 plays a central role in maintaining the intracellular chloride ion concentration by extruding chloride ions out in exchange for potassium ions. Through this action it aids in establishing the inhibitory action of neurotransmitters like GABA and glycine.
Biological function summary	KCC2 contributes significantly to the regulation of neuronal excitability and synaptic transmission. It does not appear as part of a larger protein complex but functions as a homodimer. KCC2's activity helps determine the strength and efficacy of inhibitory synapses influencing processes such as synaptic plasticity and neural development. By managing chloride ion gradients KCC2 ensures proper inhibitory neurotransmission which is essential for balanced neural activity.
Pathways	KCC2 is important for the GABAergic neurotransmission pathway which is significant for inhibitory signaling in the nervous system. This transporter interacts closely with proteins like NKCC1 another cation-chloride cotransporter which has an opposing function in chloride ion movement. The GABAergic pathway relies on KCC2 for hyperpolarizing inhibition contributing to the regulation of neuronal networks and stabilizing membrane potential.

Associated diseases and disorders	KCC2 malfunctions associate with epilepsy and neuropathic pain. Altered expression or dysfunction of KCC2 disrupts inhibitory control leading to abnormal hyperexcitability in neurons which is a characteristic of epilepsy. Moreover in neuropathic pain downregulation of KCC2 impairs glycinergic inhibition contributing to the pathophysiology of chronic pain states. In these conditions KCC2's interaction with proteins such as NKCC1 highlights its role in maintaining inhibitory balance and neural homeostasis.

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

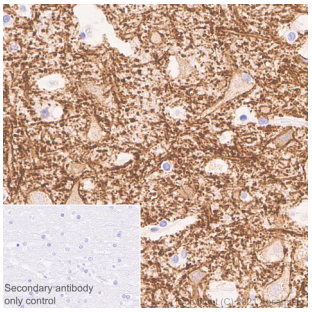
We do not recommend this combination. It is not covered by our product promise.

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



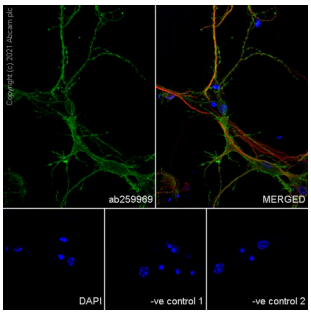
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-KCC2 antibody [EPR24203-85] (ab259969)

KCC2 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-KCC2 antibody

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



Immunocytochemistry/ Immunofluorescence - Anti-KCC2 antibody [EPR24203-85] (ab259969)

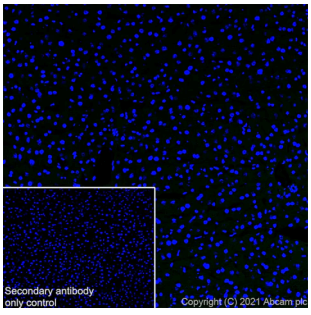
KCC2 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-KCC2 antibody

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).

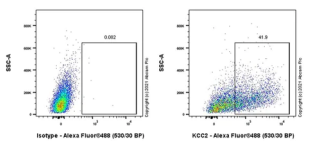


Immunohistochemistry (Frozen sections) - Anti-KCC2 antibody [EPR24203-85] (ab259969)

KCC2 Immunohistochemistry (Frozen sections) staining using rabbit Anti-KCC2 antibody

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.

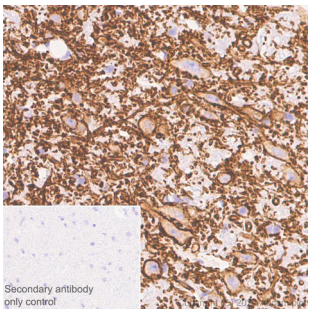
Secondary antibody control: Secondary antibody is ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 dilution (2 µg/ml).



Flow Cytometry (Intracellular) - Anti-KCC2 antibody [EPR24203-85] (ab259969)

KCC2 Flow Cytometry (Intracellular) staining using rabbit Anti-KCC2 antibody

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.



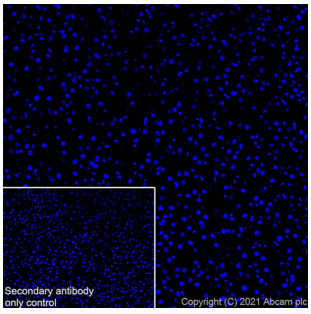
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-KCC2 antibody [EPR24203-85] (ab259969)

KCC2 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-KCC2 antibody

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

---

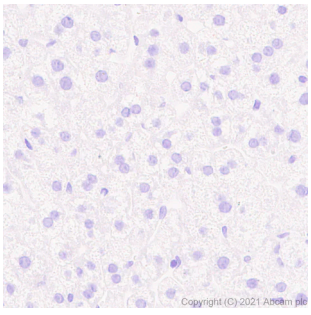


Immunohistochemistry (Frozen sections) - Anti-KCC2 antibody [EPR24203-85] (ab259969)

KCC2 Immunohistochemistry (Frozen sections) staining using rabbit Anti-KCC2 antibody

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).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-KCC2 antibody [EPR24203-85] (ab259969)

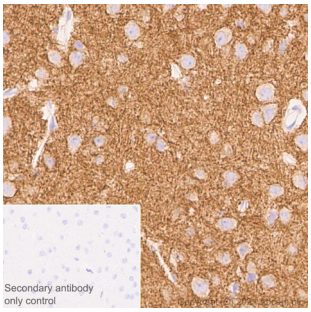
KCC2 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-KCC2 antibody

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

---

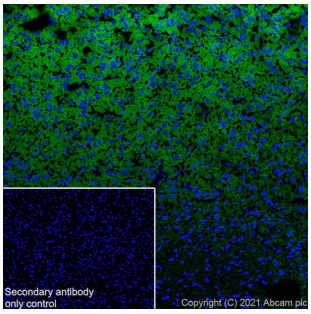


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-KCC2 antibody [EPR24203-85] (ab259969)

KCC2 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-KCC2 antibody

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).

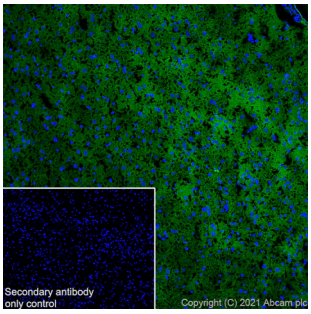
Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins



Immunohistochemistry (Frozen sections) - Anti-KCC2 antibody [EPR24203-85] (ab259969)

KCC2 Immunohistochemistry (Frozen sections) staining using rabbit Anti-KCC2 antibody

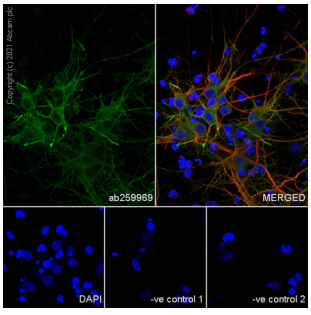
Immunohistochemical analysis of 4% PFA-fixed, 0.2% Triton X-100 permeabilized frozen mouse cerebrum (fresh) tissue labeling KCC2 with ab259969 at 1/100 dilution (5.55 µg/ml) followed by ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 dilution (2 µg/ml) (Green). Positive staining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

KCC2 Immunohistochemistry (Frozen sections) staining using rabbit Anti-KCC2 antibody

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).



Immunocytochemistry/ Immunofluorescence - Anti-KCC2 antibody [EPR24203-85] (ab259969)

KCC2 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-KCC2 antibody

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)

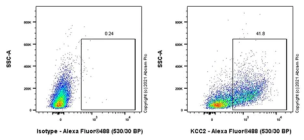
followed by ab150120 Goat Anti-Mouse IgG H&L (Alexa Fluor® 594) antibody at 1/1000 dilution (2 µg/ml) was used to counterstain tubulin (Red). The Nuclear counterstain was DAPI (Blue).

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).

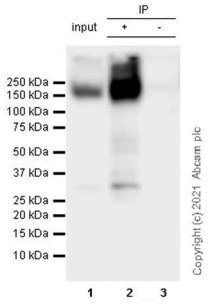
Negative control 2: ab11267 Anti-MAP2 mouse monoclonal antibody was used to counterstain tubulin at 1/500 dilution (4 µg/ml).



Flow Cytometry (Intracellular) - Anti-KCC2 antibody [EPR24203-85] (ab259969)

KCC2 Flow Cytometry (Intracellular) staining using rabbit Anti-KCC2 antibody

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] (ab259969)

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% NFDM/TBST.

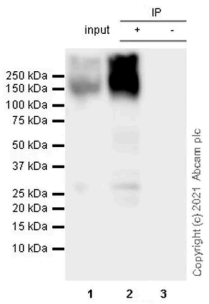
Exposure time: 3 seconds

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).

All lanes:
Immunoprecipitation - Anti-KCC2 antibody [EPR24203-85] (ab259969)

Predicted band size: 126 kDa

Observed band size: 140 kDa



Immunoprecipitation - Anti-KCC2 antibody [EPR24203-85] (ab259969)

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.

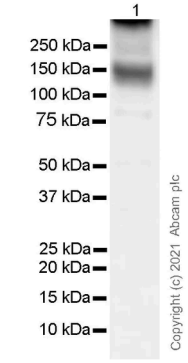
Exposure time: 3 seconds

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).

All lanes:
Immunoprecipitation - Anti-KCC2 antibody [EPR24203-85] (ab259969)

Predicted band size: 126 kDa

Observed band size: 140 kDa



Western blot - Anti-KCC2 antibody [EPR24203-85] (ab259969)

KCC2 Western blot staining of Human spinal cord using rabbit Anti-KCC2 antibody

Blocking and diluting buffer and concentration: 5% NFDM/TBST
Samples are non-boiled as boiling may cause protein aggregates.

Exposure time: 15 seconds

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

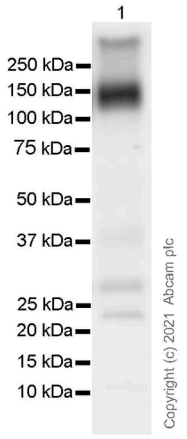
All lanes:
Human spinal cord at 20 µg

Secondary

All lanes:
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] (ab259969)

KCC2 Western blot staining of Mouse spinal cord tissue lysate using rabbit Anti-KCC2 antibody

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

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

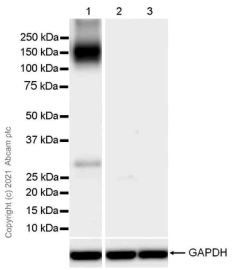
All lanes:
Mouse spinal cord tissue lysate at 20 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Predicted band size: 126 kDa

Observed band size: 140 kDa



Western blot - Anti-KCC2 antibody [EPR24203-85] (ab259969)

KCC2 Western blot staining using rabbit Anti-KCC2 antibody

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

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

Lane 1:
Mouse brain tissue lysate at 20 µg

Lane 2:
Mouse kidney tissue lysate at 20 µg

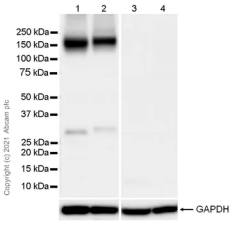
Lane 3:
Mouse liver tissue lysate at 20 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Predicted band size: 126 kDa

Observed band size: 140 kDa



Western blot - Anti-KCC2 antibody [EPR24203-85] (ab259969)

KCC2 Western blot staining using rabbit Anti-KCC2 antibody

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

All lanes:
Western blot - 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:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Predicted band size: 126 kDa

Observed band size: 140 kDa

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