Anti-Aquaporin 4 Antibody [PSH06-38]

HA722672

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IHC-P, IF-Tissue, IHC-Fr, IP
Molecular Wt:	Predicted band size: 35 kDa
Clone number:	PSH06-38

Description: Aquaporin-4, also known as AQP-4, is a water channel protein encoded by the AQP4 gene in humans. AQP-4 belongs to the aquaporin family of integral membrane proteins that conduct water through the cell membrane. A limited number of aquaporins are found within the central nervous system (CNS): AQP1, 3, 4, 5, 8, 9, and 11, but more exclusive representation of AQP1, 4, and 9 are found in the brain and spinal cord. AQP4 shows the largest presence in the cerebellum and spinal cord grey matter. In the CNS, AQP4 is the most prevalent aquaporin channel, specifically located at the perimicrovessel astrocyte foot processes, glia limitans, and ependyma. In addition, this channel is commonly found facilitating water movement near cerebrospinal fluid and vasculature. Aquaporin-4 was first identified in 1986. It was the first evidence of the existence of water transport channels. The method that was used to discover the existence of the transport channels was through knockout experiments. With this technique they were able to show the significant role of AQP4 in CNS injuries and brain water imbalances. In 1994 the channel was successfully cloned and initially named Mercury-Insensitive Water Channel.

Immunogen:	Recombinant protein within human 224-323.
Positive control:	Mouse brain tissue lysate, Rat brain tissue lysate, mouse brain tissue, rat brain tissue, human kidney tissue, human brain tissue.
Subcellular location:	Cell membrane, Basolateral cell membrane, Endosome membrane, Cell membrane, sarcolemma, Cell projection.
Database links:	SwissProt: P55087 Human | P55088 Mouse | P47863 Rat

Recommended Dilutions:

WB	1:2,000
IHC-P	1:2,000-1:10,000
IF-Tissue	1:200
IHC-Fr	1:200
IP	1-2μg/sample

Storage Buffer:	PBS (pH7.4), 0.1% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.
Storage Instruction:	Store at +4℃ after thawing. Aliquot store at -20℃. Avoid repeated freeze / thaw cycles.
Purity:	Protein A affinity purified.

Images

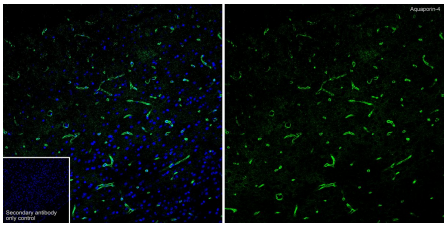

Fig1: Immunofluorescence analysis of frozen mouse brain tissue with Rabbit anti-Aquaporin 4 antibody (HA722672) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for about 2 minutes in microwave oven. The tissues were blocked in 10% negative goat serum for 1 hour at room temperature, washed with PBS, and then probed with the primary antibody (HA722672, green) at 1/200 dilution overnight at 4 ℃, washed with PBS. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. Nuclei were counterstained with DAPI (blue).

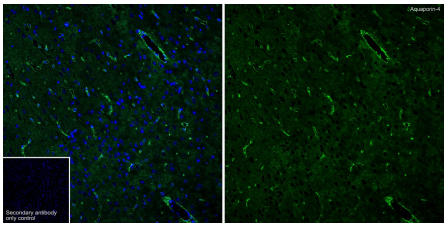

Fig2: Immunofluorescence analysis of frozen rat brain tissue with Rabbit anti-Aquaporin 4 antibody (HA722672) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for about 2 minutes in microwave oven. The tissues were blocked in 10% negative goat serum for 1 hour at room temperature, washed with PBS, and then probed with the primary antibody (HA722672, green) at 1/200 dilution overnight at 4 ℃, washed with PBS. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. Nuclei were counterstained with DAPI (blue).

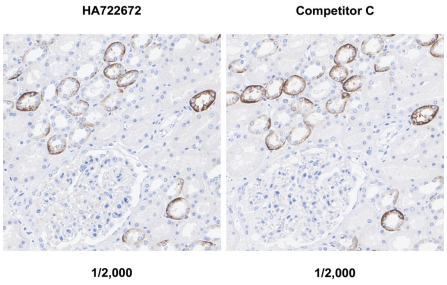

Fig3: Immunohistochemical analysis of paraffin-embedded human kidney tissue with Rabbit anti-Aquaporin 4 antibody (HA722672) at 1/2,000 dilution and competitor's antibody at 1/2,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA722672) at 1/2,000 dilution and competitor's antibody at 1/2,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

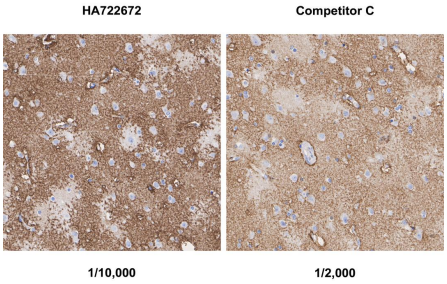

Fig4: Immunohistochemical analysis of paraffin-embedded human brain tissue with Rabbit anti-Aquaporin 4 antibody (HA722672) at 1/10,000 dilution and competitor's antibody at 1/2,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA722672) at 1/10,000 dilution and competitor's antibody at 1/2,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

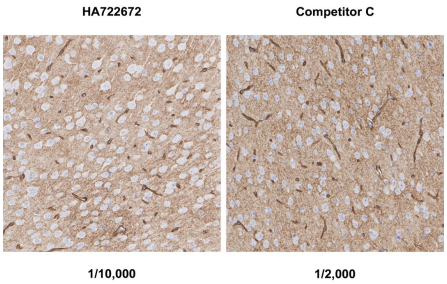

Fig5: Immunohistochemical analysis of paraffin-embedded mouse brain tissue with Rabbit anti-Aquaporin 4 antibody (HA722672) at 1/10,000 dilution and competitor's antibody at 1/2,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA722672) at 1/10,000 dilution and competitor's antibody at 1/2,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

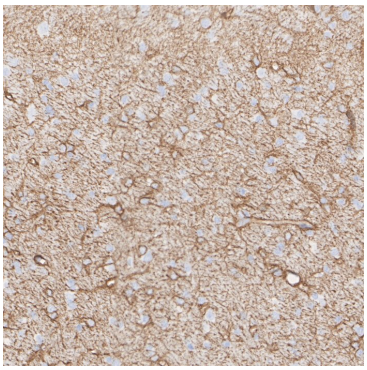

Fig6: Immunohistochemical analysis of paraffin-embedded rat brain tissue with Rabbit anti-Aquaporin 4 antibody (HA722672) at 1/10,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA722672) at 1/10,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

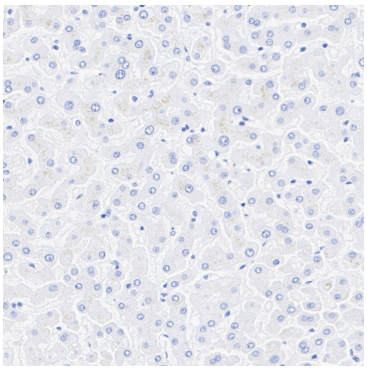

Fig7: Immunohistochemical analysis of paraffin-embedded human liver tissue (negative) with Rabbit anti-Aquaporin 4 antibody (HA722672) at 1/10,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA722672) at 1/10,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

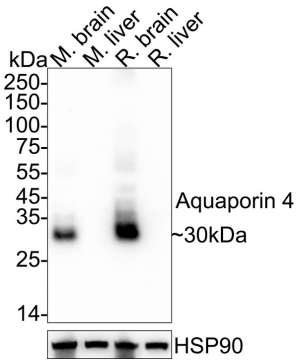

Fig8: Western blot analysis of Aquaporin 4 on different lysates with Rabbit anti-Aquaporin 4 antibody (HA722672) at 1/2,000 dilution.

Lane 1: Mouse brain tissue lysate (no heat) (40 µg/Lane)
Lane 2: Mouse liver tissue lysate (negative) (40 µg/Lane)
Lane 3: Rat brain tissue lysate (no heat) (40 µg/Lane)
Lane 4: Rat liver tissue lysate (negative) (40 µg/Lane)

Predicted band size: 35 kDa
Observed band size: 30 kDa

Exposure time: 16 seconds; ECL: K1801;
4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (HA722672) at 1/2,000 dilution was used in 5% NFDM/TBST at 4℃ overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

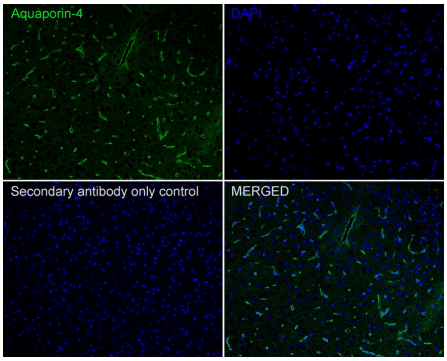

Fig9: Immunofluorescence analysis of paraffin-embedded mouse brain tissue labeling Aquaporin 4 with Rabbit anti-Aquaporin 4 antibody (HA722672) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 10% negative goat serum for 1 hour at room temperature, washed with PBS, and then probed with the primary antibody (HA722672, green) at 1/200 dilution overnight at 4 ℃, washed with PBS. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. Nuclei were counterstained with DAPI (blue).

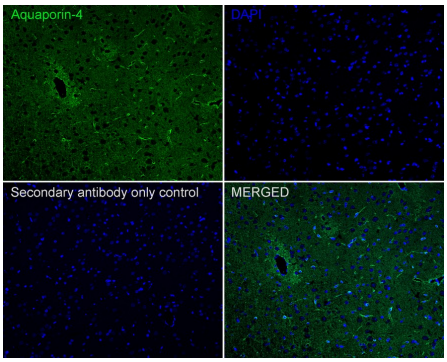

Fig10: Immunofluorescence analysis of paraffin-embedded rat brain tissue labeling Aquaporin 4 with Rabbit anti-Aquaporin 4 antibody (HA722672) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 10% negative goat serum for 1 hour at room temperature, washed with PBS, and then probed with the primary antibody (HA722672, green) at 1/200 dilution overnight at 4 ℃, washed with PBS. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. Nuclei were counterstained with DAPI (blue).

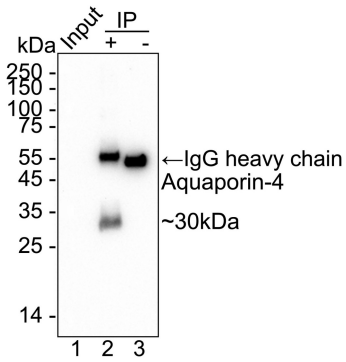

Fig11: Aquaporin 4 was immunoprecipitated from 0.2 mg mouse brain tissue lysate with HA722672 at 2 µg/25 µl agarose. Western blot was performed from the immunoprecipitate using HA722672 at 1/1,000 dilution. Mouse anti Rabbit IgG heavy chain (Fc) secondary antibody (M1003-7) at 1/100,000 dilution was used for 1 hour at room temperature.

Lane 1: Mouse brain tissue lysate (input)
Lane 2: HA722672 IP in mouse brain tissue lysate
Lane 3: Rabbit IgG instead of HA722672 in mouse brain tissue lysate

Blocking/Dilution buffer: 5% NFDM/TBST
Exposure time: 1 minute; ECL: K1801

Background References

1. Pittock SJ et al. Ravulizumab in Aquaporin-4-Positive Neuromyelitis Optica Spectrum Disorder. Ann Neurol. 2023 Jun
2. Kitchen P et al. Targeting Aquaporin-4 Subcellular Localization to Treat Central Nervous System Edema. Cell. 2020 May