Anti-SDHB Antibody [JE77-70]

HA722901

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IHC-P, IP, IF-Cell
Molecular Wt:	Predicted band size: 32 kDa
Clone number:	JE77-70

Description: Iron-sulfur protein (IP) subunit of the succinate dehydrogenase complex (mitochondrial respiratory chain complex II), responsible for transferring electrons from succinate to ubiquinone (coenzyme Q). SDH also oxidizes malate to the non-canonical enol form of oxaloacetate, enol-oxaloacetate (By similarity). Enol-oxaloacetate, which is a potent inhibitor of the succinate dehydrogenase activity, is further isomerized into keto-oxaloacetate (By similarity).

Immunogen: Synthetic peptide within human SDHB aa 1-280 / 280.

Positive control: K-562 cell lysate, HEK-293 cell lysate, NIH/3T3 cell lysate, PC-12 cell lysate, Mouse liver tissue lysate, Rat liver tissue lysate, K-562, NIH/3T3, human liver cancer tissue, human kidney tissue, rat kidney tissue, mouse kidney tissue, PC-12.

Subcellular location: Mitochondrion inner membrane.

Database links: SwissProt: P21912 Human | Q9CQA3 Mouse | P21913 Rat

Recommended Dilutions:

WB	1:2,000
IHC-P	1:1,000
IP	1-2μg/sample
IF-Cell	1:250

Storage Buffer: 1*TBS (pH7.4), 0.05% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.

Storage Instruction: Shipped at 4℃. Store at +4℃ short term (1-2 weeks). It is recommended to aliquot into single-use upon delivery. Store at -20℃ long term.

Purity: Protein A affinity purified.

Images

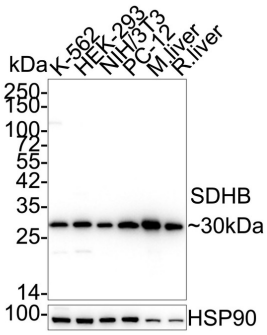

Fig1: Western blot analysis of SDHB on different lysates with Rabbit anti-SDHB antibody (HA722901) at 1/2,000 dilution.

Lane 1: K-562 cell lysate
Lane 2: HEK-293 cell lysate
Lane 3: NIH/3T3 cell lysate
Lane 4: PC-12 cell lysate
Lane 5: Mouse liver tissue lysate
Lane 6: Rat liver tissue lysate

Lysates/proteins at 20 µg/Lane.

Predicted band size: 32 kDa
Observed band size: 30 kDa

Exposure time: 4 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 (HA722901) 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.

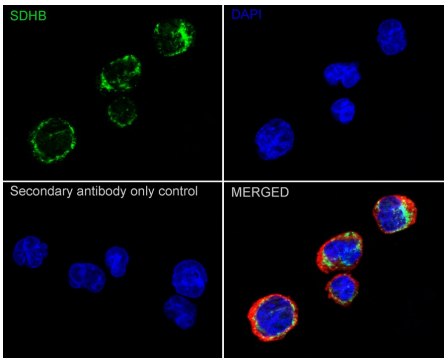

Fig2: Immunocytochemistry analysis of K-562 cells labeling SDHB with Rabbit anti-SDHB antibody (HA722901) at 1/250 dilution.

Cells were fixed in 4% paraformaldehyde for 15 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 15 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Rabbit anti-SDHB antibody (HA722901) at 1/250 dilution in 1% BSA in PBST overnight at 4 ℃. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

Beta tubulin (HA601187, red) was stained at 1/100 dilution overnight at +4℃. Goat Anti-Mouse IgG H&L (iFluor™ 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

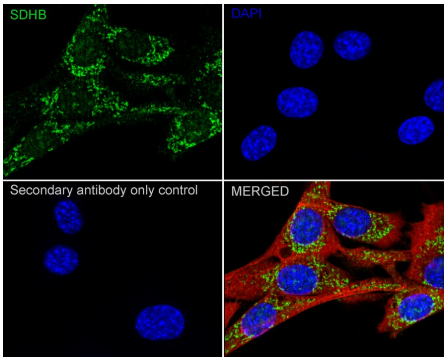

Fig3: Immunocytochemistry analysis of NIH/3T3 cells labeling SDHB with Rabbit anti-SDHB antibody (HA722901) at 1/250 dilution.

Cells were fixed in 4% paraformaldehyde for 15 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 15 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Rabbit anti-SDHB antibody (HA722901) at 1/250 dilution in 1% BSA in PBST overnight at 4 ℃. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

Beta tubulin (HA601187, red) was stained at 1/100 dilution overnight at +4℃. Goat Anti-Mouse IgG H&L (iFluor™ 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

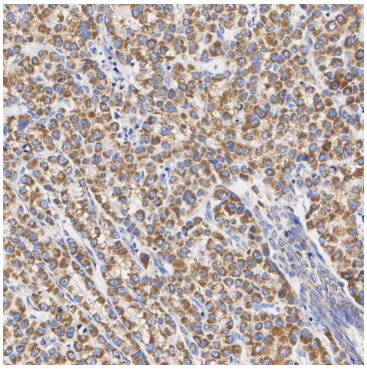

Fig4: Immunohistochemical analysis of paraffin-embedded human liver cancer tissue with Rabbit anti-SDHB antibody (HA722901) at 1/1,000 dilution.

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 (HA722901) at 1/1,000 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.

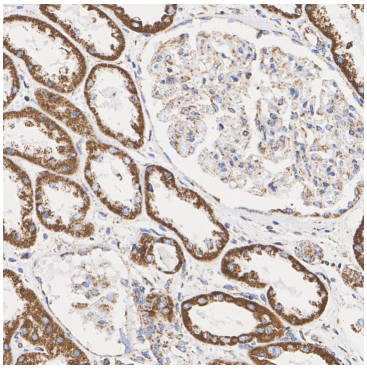

Fig5: Immunohistochemical analysis of paraffin-embedded human kidney tissue with Rabbit anti-SDHB antibody (HA722901) at 1/1,000 dilution.

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 (HA722901) at 1/1,000 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.

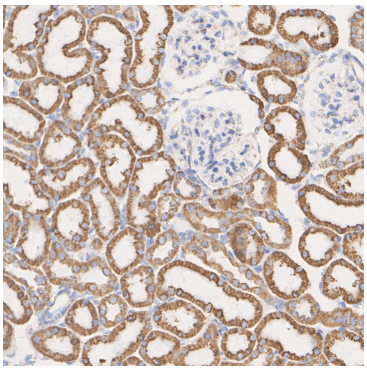

Fig6: Immunohistochemical analysis of paraffin-embedded rat kidney tissue with Rabbit anti-SDHB antibody (HA722901) at 1/1,000 dilution.

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 (HA722901) at 1/1,000 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.

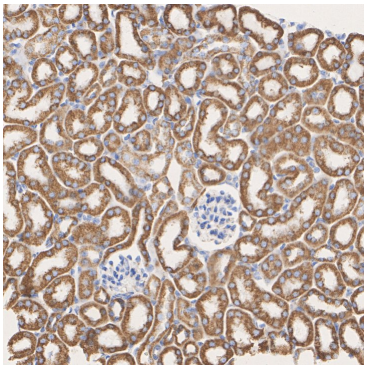

Fig7: Immunohistochemical analysis of paraffin-embedded mouse kidney tissue with Rabbit anti-SDHB antibody (HA722901) at 1/1,000 dilution.

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 (HA722901) at 1/1,000 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.

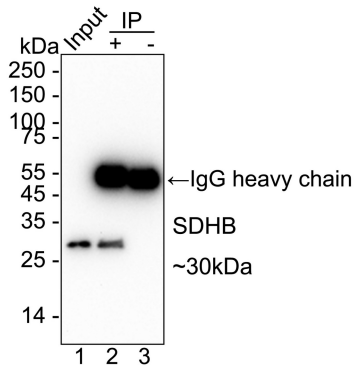

Fig8: SDHB was immunoprecipitated from 0.2 mg HEK-293 cell lysate with HA722901 at 2 µg/10 µl beads. Western blot was performed from the immunoprecipitate using HA722901 at 1/1,000 dilution. Mouse anti Rabbit IgG heavy chain (Fc) secondary antibody (M1003-7) at 1/100,000 dilution was used for 1 hour at room temperature.

Lane 1: HEK-293 cell lysate (input)
Lane 2: HA722901 IP in HEK-293 cell lysate
Lane 3: Rabbit IgG instead of HA722901 in HEK-293 cell lysate

Blocking/Dilution buffer: 5% NFDM/TBST
Exposure time: 40 seconds; ECL: K1801

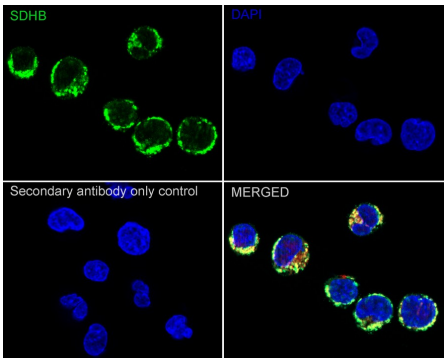

Fig9: Immunocytochemistry analysis of PC-12 cells labeling SDHB with Rabbit anti-SDHB antibody (HA722901) at 1/250 dilution.

Cells were fixed in 4% paraformaldehyde for 15 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 15 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Rabbit anti-SDHB antibody (HA722901) at 1/250 dilution in 1% BSA in PBST overnight at 4 ℃. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Counterstained with Mitotracker. Nuclear DNA was labelled in blue with DAPI.

Background References

1. Taïeb D et al. Management of phaeochromocytoma and paraganglioma in patients with germline SDHB pathogenic variants: an international expert Consensus statement. Nat Rev Endocrinol. 2024 Mar.
2. Fagundes GFC et al. Evidence for a Founder Effect of SDHB Exon 1 Deletion in Brazilian Patients With Paraganglioma. J Clin Endocrinol Metab. 2023 Jul.