Anti-Glucose Transporter GLUT1 Antibody [SA0377] - BSA and Azide free

HA750002

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IF-Cell, IF-Tissue, IHC-P, FC
Molecular Wt:	Predicted band size: 54 kDa
Clone number:	SA0377

Description: Glucose transporter 1 (or GLUT1), also known as solute carrier family 2, facilitated glucose transporter member 1 (SLC2A1), is a uniporter protein that in humans is encoded by the SLC2A1 gene. GLUT1 facilitates the transport of glucose across the plasma membranes of mammalian cells. This gene encodes a major glucose transporter in the mammalian blood-brain barrier. The encoded protein is found primarily in the cell membrane and on the cell surface, where it can also function as a receptor for human T-cell leukemia virus (HTLV) I and II. One good source of GLUT1 is erythrocyte membranes. GLUT1 accounts for 2 percent of the protein in the plasma membrane of erythrocytes. GLUT1, found in the plasma membrane of erythrocytes, is a classic example of a uniporter. After glucose is transported into the erythrocyte, it is rapidly phosphorylated, forming glucose-6-phosphate, which cannot leave the cell. Mutations in this gene can cause GLUT1 deficiency syndrome 1, GLUT1 deficiency syndrome 2, idiopathic generalized epilepsy 12, dystonia 9, and stomatin-deficient cryohydrocytosis.

Immunogen: Synthetic peptide within Human GLUT1 aa 443-492 / 492.

Positive control: HeLa cell lysate, PC-12 cell lysate, HeLa, HT-29 cell lysate, HepG2 cell lysate, NIH/3T3 cell lysate, L-929 cell lysate, mouse brain tissue lysate, rat brain tissue lysate, Jurkat, NIH/3T3, C6, human liver tissue, human placenta tissue, human liver carcinoma tissue, human kidney tissue, mouse liver tissue, mouse kidney tissue, HepG2, human lung cancer tissue, human liver tissue.

Subcellular location: Cell membrane, Melanosome

Database links: SwissProt: P11166 Human | P17809 Mouse | P11167 Rat

Recommended Dilutions:

WB	1:50,000-1:100,000
IF-Cell	1:500-1:1,000
IHC-P	1:5,000-1:10,000
IF-Tissue	1:500-1:1,000
FC	1:500-1:1,000

Storage Buffer: PBS (pH7.4).

Storage Instruction: Store at +4℃ after thawing. Aliquot store at -20℃ or -80℃. Avoid repeated freeze / thaw cycles.

Purity: Protein A affinity purified.

Images

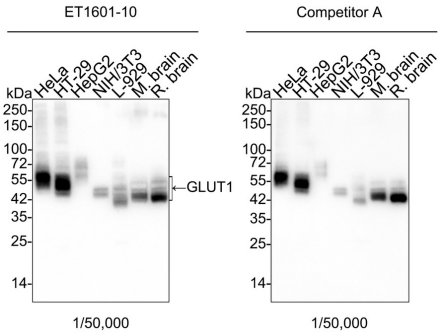

Fig1: Western blot analysis of Glucose Transporter GLUT1 on different lysates with Rabbit anti-Glucose Transporter GLUT1 antibody (HA750002) at 1/50,000 dilution and competitor's antibody at 1/50,000 dilution.

Lane 1: HeLa cell lysate (no heat) (20 µg/Lane)
Lane 2: HT-29 cell lysate (no heat) (20 µg/Lane)
Lane 3: HepG2 cell lysate (no heat) (20 µg/Lane)
Lane 4: NIH/3T3 cell lysate (no heat) (20 µg/Lane)
Lane 5: L-929 cell lysate (no heat) (20 µg/Lane)
Lane 6: Mouse brain tissue lysate (no heat) (20 µg/Lane)
Lane 7: Rat brain tissue lysate (no heat) (20 µg/Lane)

Notice: no heat means the lysate is not boiled.

Predicted band size: 54 kDa
Observed band size: 45-60 kDa
Exposure time: Lane 1-7 (left): 20 seconds; Lane 1-7 (right): 1 minute 30 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 (HA750002) at 1/50,000 dilution and competitor's antibody at 1/50,000 dilution were 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.

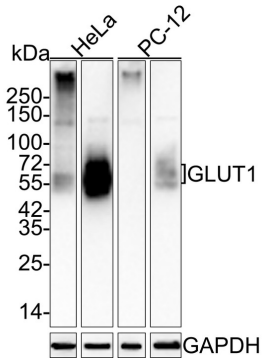

Fig2: Western blot analysis of Glucose Transporter GLUT1 on different lysates with Rabbit anti-Glucose Transporter GLUT1 antibody (HA750002) at 1/50,000 dilution.

Lane 1: HeLa cell lysate (RIPA lysis) (10 µg/Lane)
Lane 2: HeLa cell lysate (hot lysis) (10 µg/Lane)
Lane 3: PC-12 cell lysate (RIPA lysis) (10 µg/Lane)
Lane 4: PC-12 cell lysate (hot lysis) (10 µg/Lane)

Predicted band size: 54 kDa
Observed band size: 54 kDa
Exposure time: 30 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 (HA750002) at 1/50,000 dilution was used in 5% NFDM/TBST at room temperature for 2 hours. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/100,000 dilution was used for 1 hour at room temperature

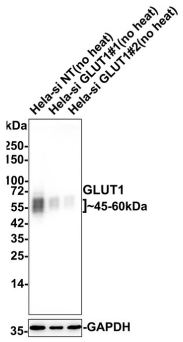

Fig3: Western blot analysis of GLUT1 on different lysates with Rabbit anti-GLUT1 antibody (HA750002) at 1/50,000 dilution.

Lane 1: Hela-si NT cell lysate (no heat)
Lane 2: Hela-si GLUT1#1 cell lysate (no heat)
Lane 3: Hela-si GLUT1#2 cell lysate (no heat)

Notice: no heat means the lysate is not boiled.

Lysates/proteins at 10 µg/Lane.

Predicted band size: 54 kDa
Observed band size: 45-60 kDa

Exposure time: 1minute 50 seconds; ECL: K1801;

4-20% SDS-PAGE gel.

ET1601-10 was shown to specifically react with GLUT1 in Hela-si NT cells. Weakened band was observed when Hela-si GLUT1 sample was tested. Hela-si NT and Hela-si GLUT1 samples were subjected to SDS-PAGE. Proteins were transferred to a PVDF membrane and blocked with 5% NFDM in TBST for 1 hour at room temperature. The primary antibody (ET1601-10, 1/50,000) and Loading control antibody (Rabbit anti-GAPDH, ET1601-4, 1/10,000) were used in 5% BSA at 4℃ overnight. Goat Anti-rabbit IgG-HRP Secondary Antibody (HA1001) at 1:50,000 dilution was used for 1 hour at room temperature.

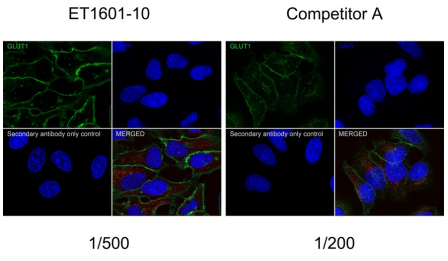

Fig4: Immunocytochemistry analysis of HeLa cells labeling Glucose Transporter GLUT1 with Rabbit anti-Glucose Transporter GLUT1 antibody (HA750002) at 1/500 dilution and competitor's antibody at 1/200 dilution.

Cells were fixed in 100% precooled methanol for 5 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-Glucose Transporter GLUT1 antibody (HA750002) at 1/500 dilution and competitor's antibody at 1/200 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 (M1305-2, 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.

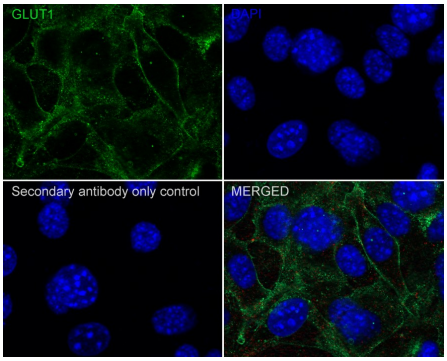

Fig5: Immunocytochemistry analysis of NIH/3T3 cells labeling Glucose Transporter GLUT1 with Rabbit anti-Glucose Transporter GLUT1 antibody (HA750002) at 1/500 dilution.

Cells were fixed in 100% precooled methanol for 5 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-Glucose Transporter GLUT1 antibody (HA750002) at 1/500 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 (M1305-2, 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.

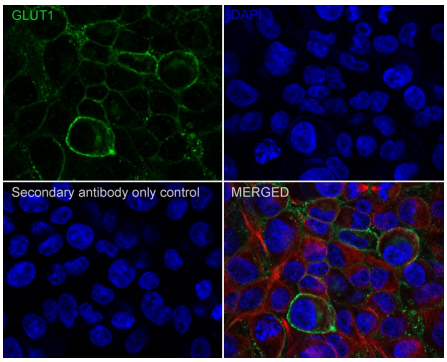

Fig6: Immunocytochemistry analysis of C6 cells labeling Glucose Transporter GLUT1 with Rabbit anti-Glucose Transporter GLUT1 antibody (HA750002) at 1/500 dilution.

Cells were fixed in 100% precooled methanol for 5 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-Glucose Transporter GLUT1 antibody (HA750002) at 1/500 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 (M1305-2, 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.

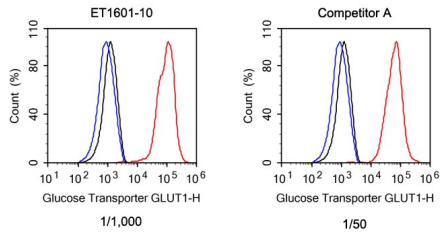

Fig7: Flow cytometric analysis of Jurkat cells labeling Glucose Transporter GLUT1.

Cells were fixed and permeabilized. Then stained with the primary antibody (HA750002, red) at 1/1,000 dilution and competitor's antibody (red) at 1/50 dilution, compared with Rabbit IgG Isotype Control (blue). After incubation of the primary antibody at +4℃ for an hour, the cells were stained with a iFluor™ 488 conjugate-Goat anti-Rabbit IgG Secondary antibody (HA1121) at 1/1,000 dilution for 30 minutes at +4℃. Unlabelled sample was used as a control (cells without incubation with primary antibody; black).

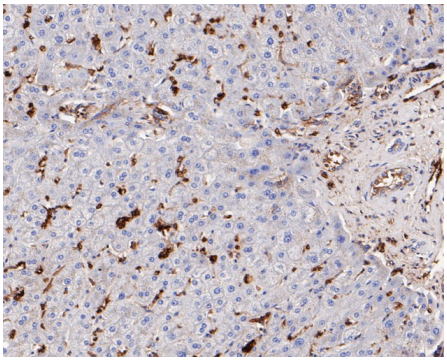

Fig8: Immunohistochemical analysis of paraffin-embedded human liver tissue with Rabbit anti-Glucose Transporter GLUT1 antibody (HA750002) at 1/5,000 dilution.

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 (HA750002) at 1/5,000 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.

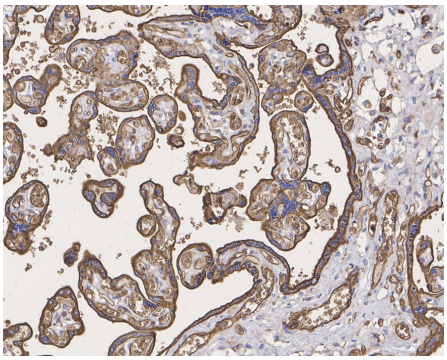

Fig9: Immunohistochemical analysis of paraffin-embedded human placenta tissue with Rabbit anti-Glucose Transporter GLUT1 antibody (HA750002) at 1/5,000 dilution.

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 (HA750002) at 1/5,000 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.

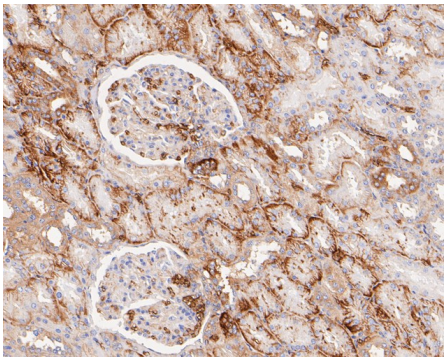

Fig10: Immunohistochemical analysis of paraffin-embedded human kidney tissue with Rabbit anti-Glucose Transporter GLUT1 antibody (HA750002) at 1/10,000 dilution.

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 (HA750002) 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.

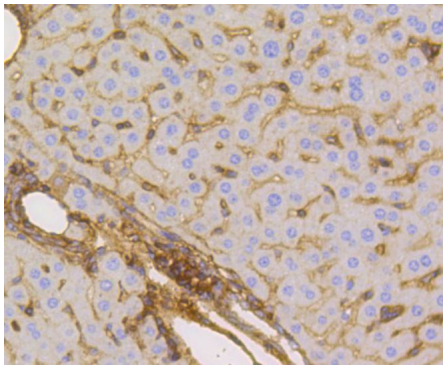

Fig11: Immunohistochemical analysis of paraffin-embedded mouse liver tissue with Rabbit anti-Glucose Transporter GLUT1 antibody (HA750002) at 1/5,000 dilution.

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 (HA750002) at 1/5,000 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.

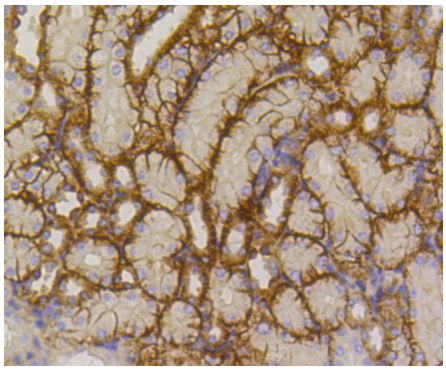

Fig12: Immunohistochemical analysis of paraffin-embedded mouse kidney tissue with Rabbit anti-Glucose Transporter GLUT1 antibody (HA750002) at 1/5,000 dilution.

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 (HA750002) at 1/5,000 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.

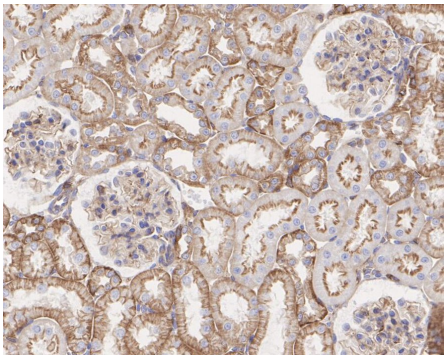

Fig13: Immunohistochemical analysis of paraffin-embedded rat kidney tissue with Rabbit anti-Glucose Transporter GLUT1 antibody (HA750002) at 1/5,000 dilution.

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 (HA750002) 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.

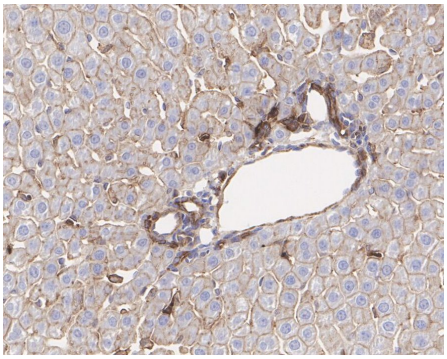

Fig14: Immunohistochemical analysis of paraffin-embedded rat liver tissue with Rabbit anti-Glucose Transporter GLUT1 antibody (HA750002) at 1/5,000 dilution.

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 (HA750002) 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.

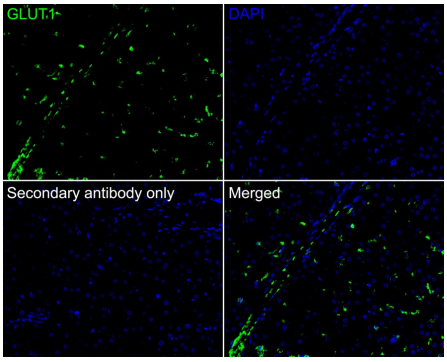

Fig15: Application: IF-tissue

Species: Human

Site: Liver

Sample: Paraffin-embedded section

Antibody concentration: 1/500

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).

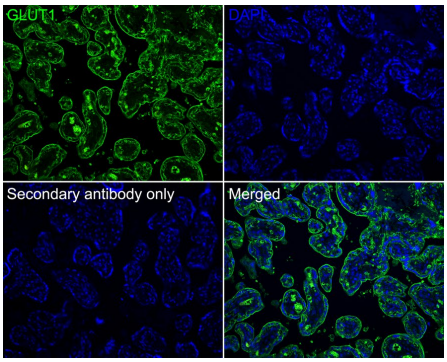

Fig16: Application: IF-tissue

Species: Human

Site: Placenta

Sample: Paraffin-embedded section

Antibody concentration: 1/500

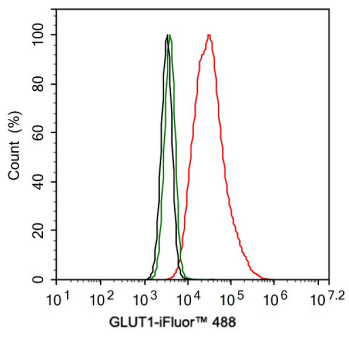

Fig17: Flow cytometric analysis of HepG2 cells labeling Glucose Transporter GLUT1.

Cells were fixed and permeabilized. Then stained with the primary antibody (HA750002, 1ug/ml) (red) compared with Rabbit IgG Isotype Control (green). After incubation of the primary antibody at +4℃ for an hour, the cells were stained with a iFluor™ 488 conjugate-Goat anti-Rabbit IgG Secondary antibody (HA1121) at 1/1,000 dilution for 30 minutes at +4℃. Unlabelled sample was used as a control (cells without incubation with primary antibody; black).

Background References

1. Boyer-Di Ponio J et al. Instruction of circulating endothelial progenitors in vitro towards specialized blood-brain barrier and arterial phenotypes. PLoS One 9:e84179 (2014).
2. Saucillo DC et al. Leptin metabolically licenses T cells for activation to link nutrition and immunity. J Immunol 192:136-44 (2014).