abcam

Product datasheet

Anti-Glucose Transporter GLUT1 antibody [EPR3915] ab115730





RabMAb

*** 19 Abreviews 235 References 26 Images

Overview

Product name Anti-Glucose Transporter GLUT1 antibody [EPR3915]

Description Rabbit monoclonal [EPR3915] to Glucose Transporter GLUT1

Host species Rabbit

Specificity We recommend not to boil the samples after lysis to get desired WB bands.

Tested applications Suitable for: Flow Cyt (Intra), ICC/IF, WB, IHC-P

Species reactivity Reacts with: Mouse, Rat, Human

Immunogen Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

Positive control WB: NIH/3T3, HepG2, HT-29, SW480, 3T3-L1 and PC-12 whole cell lysates. IHC-P: Rat kidney

> tissue; mouse liver tissue; human lung carcinoma, cervical carcinoma, colon carcinoma, liver, colon, kidney carcinoma, skeletal muscle, urinary bladder, heart and breast tissue. ICC/IF: HepG2 cells and A549 (SLC2A1 knockout A549 cells used as a negative control) cells. Flow Cyt (intra):

HeLa and Jurkat cells.

General notes 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

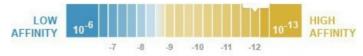
Form Liquid

Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C. Storage instructions

Avoid freeze / thaw cycle.

 $K_D = 7.70 \times 10^{-12} M$ Dissociation constant (K_D)

10⁻¹²



Learn more about K_D

Storage buffer pH: 7.20

Preservative: 0.01% Sodium azide

Constituents: 49% PBS, 50% Glycerol (glycerin, glycerine), 0.05% BSA

Purity Protein A purified

ClonalityMonoclonalClone numberEPR3915

Isotype IgG

Applications

The Abpromise guarantee Our <u>Abpromise guarantee</u> covers the use of ab115730 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	
Flow Cyt (Intra)		1/40. For unpurified, use 1/100 - 1/1000. <u>ab172730</u> - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody.	
ICC/IF	★★★★★ (2)	Use a concentration of 1 μ g/ml. This product gave a positive signal in A549 (SLC2A1 knockout A549 cells used as a negative control) fixed with 100% methanol (5 min).	
WB	★★★★ (11)	1/100000. Detects a band of approximately 40-60 kDa (predicted molecular weight: 54 kDa). We would not recommend boiling due to the possible irreversible aggregation of glycose transporters. If samples are boiled it can prevent some of the protein from entering the gel or produce multimers which are often mistaken for background. Samples should be solubilized in standard SDS Laemmli buffer and	
IHC-P	★★★☆☆(4)	1/250 - 1/500. Perform heat mediated antigen retrieval before commencing with IHC staining protocol. See IHC antigen retrieval protocols.	

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Function Facilitative glucose transporter. This isoform may be responsible for constitutive or basal glucose

uptake. Has a very broad substrate specificity; can transport a wide range of aldoses including

both pentoses and hexoses.

Tissue specificity Expressed at variable levels in many human tissues.

Involvement in diseaseDefects in SLC2A1 are the cause of glucose transporter type 1 deficiency syndrome (GLUT1DS)

[MIM:606777]; also known as blood-brain barrier glucose transport defect. This disease causes a defect in glucose transport across the blood-brain barrier. It is characterized by infantile seizures, delayed development, and acquired microcephaly.

Defects in SLC2A1 are the cause of dystonia type 18 (DYT18) [MIM:612126]. DYT18 is an exercise-induced paroxysmal dystonia/dyskinesia. Dystonia is defined by the presence of sustained involuntary muscle contraction, often leading to abnormal postures. DYT18 is characterized by attacks of involuntary movements triggered by certain stimuli such as sudden movement or prolonged exercise. In some patients involuntary exertion-induced dystonic, choreoathetotic, and ballistic movements may be associated with macrocytic hemolytic anemia.

Sequence similarities

Belongs to the major facilitator superfamily. Sugar transporter (TC 2.A.1.1) family. Glucose transporter subfamily.

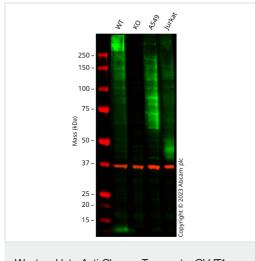
Post-translational modifications

Phosphorylated upon DNA damage, probably by ATM or ATR.

Cellular localization

Cell membrane. Melanosome. Localizes primarily at the cell surface (By similarity). Identified by mass spectrometry in melanosome fractions from stage I to stage IV.

Images



Western blot - Anti-Glucose Transporter GLUT1 antibody [EPR3915] (ab115730)

All lanes : Anti-Glucose Transporter GLUT1 antibody [EPR3915] (ab115730) at 1/100000 dilution

Lane 1: Wild-type HepG2 cell lysate

Lane 2: LC2A1 knockout HepG2 cell lysate

Lane 3 : A549 cell lysate
Lane 4 : Jurkat cell lysate

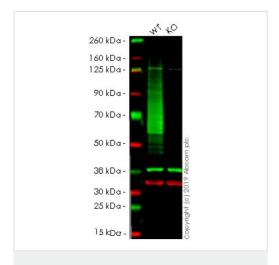
Lysates/proteins at 20 mg/ml per lane.

Performed under reducing conditions.

Predicted band size: 54 kDa **Observed band size:** 50-300 kDa

Western blot: Anti-Glucose Transporter GLUT1 antibody [EPR3915] staining at 1/100000 dilution, shown in green; Mouse anti-GAPDH antibody [6C5] (ab8245) loading control staining at 1/20000 dilution, shown in red. In Western blot, ab115730 was shown to bind specifically to Glucose Transporter GLUT1. A band was observed at 50-300 kDa in wild-type HepG2 cell lysates with no signal observed at this size in SLC2A1 knockout cell line ab280797 (knockout cell lysate ab284224). To generate this image, wild-type and SLC2A1 knockout HepG2 cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a

nitrocellulose membrane. Membranes were blocked in 3% milk in TBS-0.1% Tween[®] 20 (TBS-T) before incubation with primary antibodies overnight at 4°C. Blots were washed four times in TBS-T, incubated with secondary antibodies for 1 h at room temperature, washed again four times then imaged. Secondary antibodies used were Goat anti-Rabbit lgG H&L 800CW and Goat anti-Mouse lgG H&L 680RD at 1/20000 dilution.



Western blot - Anti-Glucose Transporter GLUT1 antibody [EPR3915] (ab115730)

All lanes : Anti-Glucose Transporter GLUT1 antibody [EPR3915] (ab115730) at 1 μg/ml

Lane 1: Wild-type A549 whole cell lysate

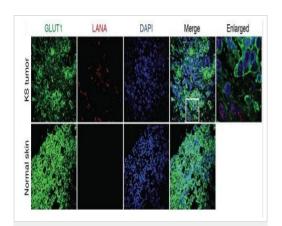
Lane 2: Human SLC2A1 (Glucose Transporter GLUT1) knockout A549 cell line (ab261869)

Lysates/proteins at 20 µg per lane.

Predicted band size: 54 kDa

Lanes 1 - 2: Merged signal (red and green). Green - <u>ab196357</u> observed at 54 kDa. Red - loading control, <u>ab8245</u>, observed at 37 kDa.

<u>ab196357</u> was shown to recognize in wild-type A549 cells as signal was lost at the expected MW in SLC2A1 knockout cells. Additional cross-reactive bands were observed in the wild-type and knockout cells. Wild-type and SLC2A1 knockout samples were subjected to SDS-PAGE. Ab196357 and <u>ab8245</u> (Mouse anti-GAPDH loading control) were incubated overnight at 4°C at 1 μg/ml and 1/20000 dilution respectively. Blots were developed with Goat anti-Rabbit lgG H&L (IRDye® 800CW) preabsorbed <u>ab216773</u> and Goat anti-Mouse lgG H&L (IRDye® 680RD) preabsorbed <u>ab216776</u> secondary antibodies at 1/20000 dilution for 1 hour at room temperature before imaging.



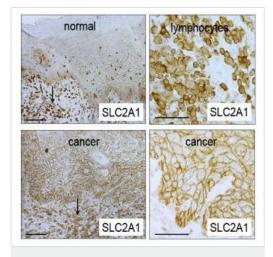
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Glucose Transporter GLUT1 antibody [EPR3915] (ab115730)

Zhu, Y. et al PLoS Pathog. 2016 May 17;12(5):e1005648. doi: 10.1371/journal.ppat.1005648. eCollection 2016 May Reproduced under the Creative Commons license

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GLUT1 and GLUT3 are downregulated in KSHV-infected cells in human KS tumors

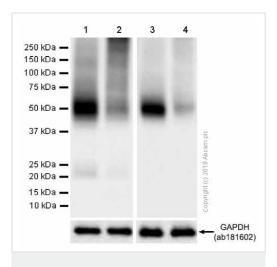
Representative illustration of dual immunofluorescence detection of LANA and GLUT1 or in a normal human skin section and a Karposi Sarcoma (KS) tumor section. Tissues were fixed with paraformaldehyhde and paraffin-embedded.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Glucose Transporter
GLUT1 antibody [EPR3915] (ab115730)

Khaom, R. et al PLoS One. 2016 Aug 11;11(8):e0161163. doi: 10.1371/journal.pone.0161163. eCollection 2016 Reproduced under the Creative Commons license http://creativecommons.org/licenses/by/4.0/ Immunohistochemical expression of Glut1 in normal tongue epithelium and tongue cancer. Expression was greatest in lymphocytes (arrows in left upper and lower panels). In the normal oral epithelium, Glut1 was weakly expressed in the basal and spinous cells (left upper panel). In OSCC, Glut1 was upregulated, showing a level of expression comparable with lymphocytes (left and right lower panels). Scale bar, 100 μ m.

Note: Glut1 = SLC2A (alternative names for the same target).



Western blot - Anti-Glucose Transporter GLUT1 antibody [EPR3915] (ab115730)

All lanes : Anti-Glucose Transporter GLUT1 antibody [EPR3915] (ab115730) at 1/50000 dilution

Lane 1: HT-29 (Human colorectal adenocarcinoma epithelial cell) whole cell lysates unboiled with 5% NFDM/TBST

Lane 2: HT-29 (Human colorectal adenocarcinoma epithelial cell) whole cell lysates boiled with 5% NFDM/TBST

Lane 3: 3T3-L1 (Mouse embryonic fibroblast) whole cell lysates unboiled with 5% NFDM/TBST

Lane 4: 3T3-L1 (Mouse embryonic fibroblast) whole cell lysates boiled with 5% NFDM/TBST

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) (<u>ab97051</u>) at 1/20000 dilution

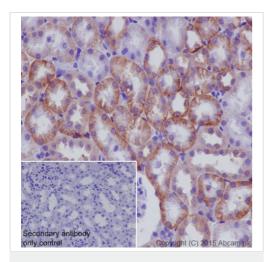
Predicted band size: 54 kDa

Observed band size: 40-60 kDa

Exposure time

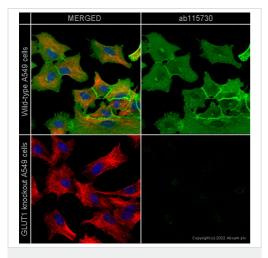
Lane 1 to 2: 10 seconds
Lane 3 to 4: 30 seconds

We recommend not to boil the samples after lysis to get desired WB bands.



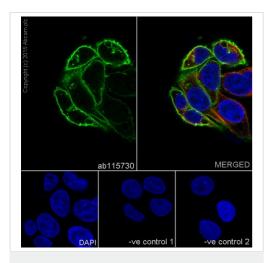
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Glucose Transporter
GLUT1 antibody [EPR3915] (ab115730)

Immunohistochemical staining of paraffin embedded rat kidney with purified ab115730 at a working dilution of 1/500. The secondary antibody used is <u>ab97051</u>, a goat anti-rabbit IgG (H&L) at a dilution of 1/500. The sample is counter-stained with hematoxylin. Antigen retrieval was performed using Tris-EDTA buffer, pH 9.0. PBS was used instead of the primary antibody as the negative control, and is shown in the inset.

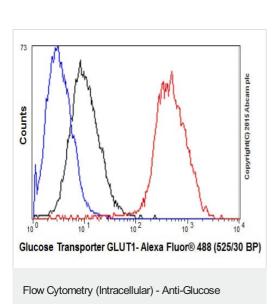


Immunocytochemistry/ Immunofluorescence - Anti-Glucose Transporter GLUT1 antibody [EPR3915] (ab115730)

ab115730 staining SLC2A1 in wild-type A549 cells, with negative expression in SLC2A1 knockout A549 cells. The cells were fixed with 100% methanol (5 min), permeabilised with 0.1% Triton x-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated overnight at +4°C with ab115730 at 1 µg/ml and ab7291, Mouse monoclonal [DM1A] to alpha Tubulin at 0.5 µg/ml. Cells were then incubated with ab150081, Goat polyclonal Secondary Antibody to Rabbit IgG - H&L (Alexa Fluor® 488), pre-adsorbed at 1/1000 dilution (shown in green) and ab150119, Goat polyclonal Secondary Antibody to Mouse IgG - H&L (Alexa Fluor® 647), pre-adsorbed at 1/1000 dilution (shown in red). Nuclear DNA was labelled with DAPI (shown in blue).Image was acquired with a confocal microscope (Leica-Microsystems TCS SP8) and a single confocal section is shown.



Immunocytochemistry/ Immunofluorescence - Anti-Glucose Transporter GLUT1 antibody [EPR3915] (ab115730)



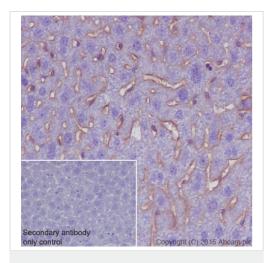
Transporter GLUT1 antibody [EPR3915] (ab115730)

Immunofluorescence staining of HepG2 cells with purified ab115730 at a working dilution of 1/100, counter-stained with DAPI. The secondary antibody was Alexa Fluor[®] 488 goat anti-rabbit (ab150077), used at a dilution of 1/1000. ab7291, a mouse antitubulin antibody (1/1000), was used to stain tubulin along with ab150120 (Alexa Fluor[®] 594 goat anti-mouse, 1/1000), shown in the top right hand panel. The cells were fixed in 4% PFA and permeabilized using 0.1% Triton X 100. The negative controls are shown in bottom middle and right hand panels - for negative control 1, purified ab115730 was used at a dilution of 1/500 followed by an Alexa Fluor[®] 594 goat anti-mouse antibody (ab150120) at a dilution of 1/500. For negative control 2, ab7291 (mouse antitubulin) was used at a dilution of 1/500 followed by an Alexa Fluor[®] 488 goat anti-rabbit antibody (ab150077) at a dilution of 1/400. Alexa Fluor[®] 488 (ab195359) and Alexa Fluor[®] 647 (ab195020)

Alexa Fluor[®] 488 (<u>ab195359</u>) and Alexa Fluor[®] 647 (<u>ab195020</u>) conjugated versions are available for this clone.

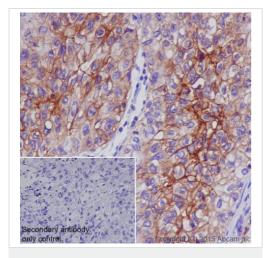
Overlay histogram showing Jurkat cells fixed in 4% PFA and stained with purified ab115730 at a dilution of 1/40 (red line). The secondary antibody used was Alexa Fluorr[®] 488 goat anti-rabbit at a dilution of 1/500. Rabbit monoclonal lgG was used as an isotype control (black line) and cells incubated in the absence of both primary and secondary antibody were used as a negative control (blue line).

Alexa Fluorr[®]488 (<u>ab195359</u>) and Alexa Fluorr[®]647 (<u>ab195020</u>) conjugated versions are available for this clone.



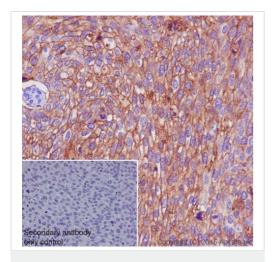
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Glucose Transporter
GLUT1 antibody [EPR3915] (ab115730)

Immunohistochemical staining of paraffin embedded mouse liver with purified ab115730 at a working dilution of 1/500. The secondary antibody used is **ab97051**, a goat anti-rabbit lgG (H&L) at a dilution of 1/500. The sample is counter-stained with hematoxylin. Antigen retrieval was perfomed using Tris-EDTA buffer, pH 9.0. PBS was used instead of the primary antibody as the negative control, and is shown in the inset.



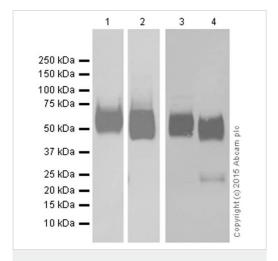
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Glucose Transporter
GLUT1 antibody [EPR3915] (ab115730)

Immunohistochemical staining of paraffin embedded human lung carcinoma with purified ab115730 at a working dilution of 1/500. The secondary antibody used is <u>ab97051</u>, a goat anti-rabbit lgG (H&L) at a dilution of 1/500. The sample is counter-stained with hematoxylin. Antigen retrieval was performed using Tris-EDTA buffer, pH 9.0. PBS was used instead of the primary antibody as the negative control, and is shown in the inset.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Glucose Transporter
GLUT1 antibody [EPR3915] (ab115730)

Immunohistochemical staining of paraffin embedded human cervical carcinoma with purified ab115730 at a working dilution of 1/500. The secondary antibody used is **ab97051**, a goat anti-rabbit lgG (H&L) at a dilution of 1/500. The sample is counter-stained with hematoxylin. Antigen retrieval was performed using Tris-EDTA buffer, pH 9.0. PBS was used instead of the primary antibody as the negative control, and is shown in the inset.



Western blot - Anti-Glucose Transporter GLUT1 antibody [EPR3915] (ab115730)

All lanes : Anti-Glucose Transporter GLUT1 antibody [EPR3915] (ab115730) at 1/1000000 dilution (purified)

Lane 1 : HepG2 whole cell lysate
Lane 2 : Human fetal liver lysate
Lane 3 : HT-29 whole cell lysate
Lane 4 : SW480 whole cell lysate

Lysates/proteins at 10 µg per lane.

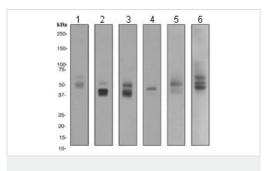
Secondary

All lanes : Anti-rabbit lgG (HRP), specific to the non-reduced form of lgG at 1/1000 dilution

Predicted band size: 54 kDa

Observed band size: 40-60 kDa

Blocking buffer: 5% NFDM/TBST Dilution buffer: 5% NFDM/TBST



Western blot - Anti-Glucose Transporter GLUT1 antibody [EPR3915] (ab115730)

All lanes : Anti-Glucose Transporter GLUT1 antibody [EPR3915] (ab115730) at 1/1000 dilution (Unpurified)

Lane 1: Jurkat lysate

Lane 2: Mouse brain lysate

Lane 3: Human fetal brain lysate

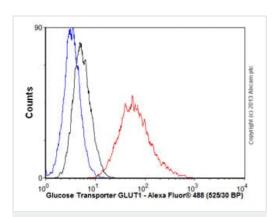
Lane 4: 3T3L1 lysate

Lane 5: Human fetal liver lysate

Lane 6: HepG2 lysate

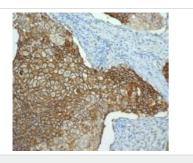
Lysates/proteins at 10 µg per lane.

Predicted band size: 54 kDa



Flow Cytometry (Intracellular) - Anti-Glucose Transporter GLUT1 antibody [EPR3915] (ab115730)

Overlay histogram showing HeLa cells stained with unpurified ab115730 (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab115730, 1/1000 dilution) for 30 min at 22°C. The secondary antibody used was Alexa Fluor® 488 goat anti-rabbit IgG (H+L) (ab150077) at 1/2000 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit IgG (monoclonal) (1µg/1x10⁶ cells) used under the same conditions. Unlabelled sample (blue line) was also used as a control. Acquisition of >5,000 events were collected using a 20mW Argon ion laser (488nm) and 525/30 bandpass filter. This antibody gave a positive signal in HeLa cells fixed with 4% paraformaldehyde (10 min)/permeabilized with 0.1% PBS-Tween for 20 min used under the same conditions.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Glucose Transporter
GLUT1 antibody [EPR3915] (ab115730)

Unpurified ab115730 at 1/250 dilution staining Glucose Transporter GLUT1 in Paraffin-embedded human cervical carcinoma tissue by Immunohistochemistry.

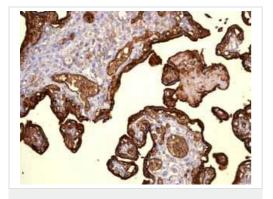
Perform heat mediated antigen retrieval before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Glucose Transporter
GLUT1 antibody [EPR3915] (ab115730)

Unpurified ab115730 at 1/250 dilution staining Glucose Transporter GLUT1 in Paraffin-embedded human colonic adenocarcinoma tissue by Immunohistochemistry.

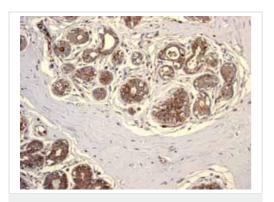
Perform heat mediated antigen retrieval before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Glucose Transporter
GLUT1 antibody [EPR3915] (ab115730)

Unpurified ab115730 showing positive staining in human normal liver tissue.

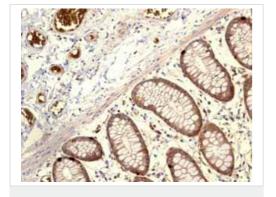
Perform heat mediated antigen retrieval before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Glucose Transporter
GLUT1 antibody [EPR3915] (ab115730)

Unpurified ab115730 showing positive staining in human normal breast tissue.

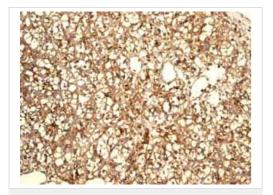
Perform heat mediated antigen retrieval before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Glucose Transporter
GLUT1 antibody [EPR3915] (ab115730)

Unpurified ab115730 showing positive staining in human normal colon tissue.

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Glucose Transporter
GLUT1 antibody [EPR3915] (ab115730)

Unpurified ab115730 showing positive staining in human kidney carcinoma tissue.

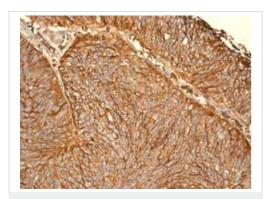
Perform heat mediated antigen retrieval before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Glucose Transporter
GLUT1 antibody [EPR3915] (ab115730)

Unpurified ab115730 showing negative staining in human skeletal muscle tissue.

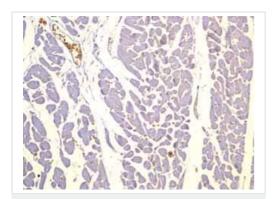
Perform heat mediated antigen retrieval before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Glucose Transporter
GLUT1 antibody [EPR3915] (ab115730)

Unpurified ab115730 showing positive staining in human urinary bladder transitional carcinoma tissue.

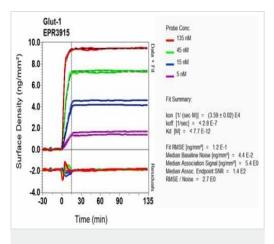
Perform heat mediated antigen retrieval before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Glucose Transporter
GLUT1 antibody [EPR3915] (ab115730)

Unpurified ab115730 showing negative staining in human normal heart tissue.

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.



Ol-RD Scanning - Anti-Glucose Transporter GLUT1 antibody [EPR3915] (ab115730)

Equilibrium disassociation constant (K_D) Learn more about K_D

Click here to learn more about K_D



Anti-Glucose Transporter GLU11 antibody [EPR3915] (ab115730)

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