Anti-xCT/SLC7A11 Antibody [A7C6]

HA600098

Product Type:	Mouse monoclonal IgG1, primary antibodies
Species reactivity:	Human, Mouse
Applications:	WB, IF-Cell, IHC-P, FC
Molecular Wt:	Predicted band size: 55 kDa
Clone number:	A7C6

Description: This gene encodes a member of a heteromeric, sodium-independent, anionic amino acid transport system that is highly specific for cysteine and glutamate. In this system, designated Xc(-), the anionic form of cysteine is transported in exchange for glutamate. This protein has been identified as the predominant mediator of Kaposi sarcoma-associated herpesvirus fusion and entry permissiveness into cells. Also, increased expression of this gene in primary gliomas (compared to normal brain tissue) was associated with increased glutamate secretion via the XCT channels, resulting in neuronal cell death.

Immunogen: 293 cell line overexpressing SLC7A11.

Positive control: HT-29 cell lysates, HeLa, A549, HT-29, human colon tissue, human pancreas tissue.

Subcellular location: Cell membrane, Cell projection, microvillus membrane.

Database links: SwissProt: Q9UPY5 Human | Q9WTR6 Mouse

Recommended Dilutions:

WB	1:1,000
IF-Cell	1:100-1:250
IHC-P	1:200-1:600
FC	1:1,000

Storage Buffer: PBS (pH7.4), 0.05% 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

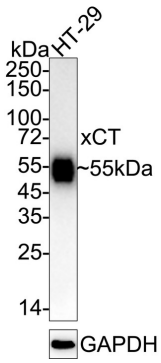

Fig1: Western blot analysis of xCT/SLC7A11 on HT-29 cell lysates with Mouse anti-xCT/SLC7A11 antibody (HA600098) at 1/1,000 dilution.

Lysates/proteins at 10 µg/Lane.

Predicted band size: 55 kDa
Observed band size: 55 kDa

Exposure time: 2 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 (HA600098) at 1/1,000 dilution was used in 5% NFDM/TBST at 4℃ overnight. Goat Anti-Mouse IgG - HRP Secondary Antibody (HA1006) at 1/50,000 dilution was used for 1 hour at room temperature.

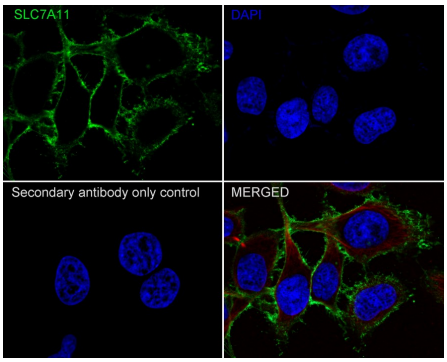

Fig2: Immunocytochemistry analysis of HeLa cells labeling xCT/SLC7A11 with Mouse anti-xCT/SLC7A11 antibody (HA600098) at 1/250 dilution.

Cells were fixed in 4% paraformaldehyde for 20 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS 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 Mouse anti-xCT/SLC7A11 antibody (HA600098) at 1/250 dilution in 1% BSA in PBST overnight at 4 ℃. Goat Anti-Mouse IgG H&L (iFluor™ 488, HA1125) 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 (ET1602-4, red) was stained at 1/100 dilution overnight at +4℃. Goat Anti-Rabbit IgG H&L (iFluor™ 594, HA1122) was used as the secondary antibody at 1/1,000 dilution.

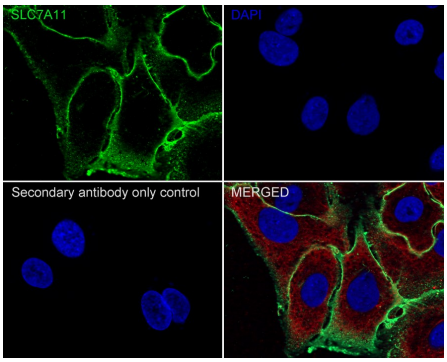

Fig3: Immunocytochemistry analysis of A549 cells labeling xCT/SLC7A11 with Mouse anti-xCT/SLC7A11 antibody (HA600098) at 1/250 dilution.

Cells were fixed in 4% paraformaldehyde for 20 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS 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 Mouse anti-xCT/SLC7A11 antibody (HA600098) at 1/250 dilution in 1% BSA in PBST overnight at 4 ℃. Goat Anti-Mouse IgG H&L (iFluor™ 488, HA1125) 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 (ET1602-4, red) was stained at 1/100 dilution overnight at +4℃. Goat Anti-Rabbit IgG H&L (iFluor™ 594, HA1122) was used as the secondary antibody at 1/1,000 dilution.

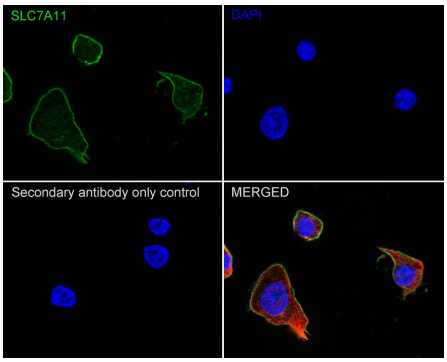

Fig4: Immunocytochemistry analysis of HT-29 cells labeling xCT/SLC7A11 with Mouse anti-xCT/SLC7A11 antibody (HA600098) at 1/100 dilution.

Cells were fixed in 4% paraformaldehyde for 20 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS 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 Mouse anti-xCT/SLC7A11 antibody (HA600098) at 1/100 dilution in 1% BSA in PBST overnight at 4 ℃. Goat Anti-Mouse IgG H&L (iFluor™ 488, HA1125) 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 (ET1602-4, red) was stained at 1/100 dilution overnight at +4℃. Goat Anti-Rabbit IgG H&L (iFluor™ 594, HA1122) was used as the secondary antibody at 1/1,000 dilution.

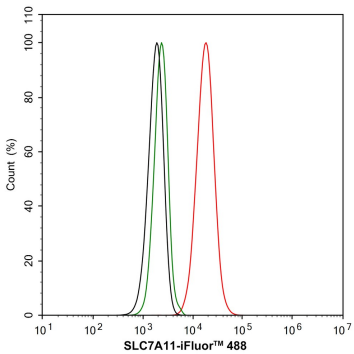

Fig5: Flow cytometric analysis of HT-29 cells labeling xCT/SLC7A11.

Cells were washed twice with cold PBS and resuspend. Then stained with the primary antibody (HA600098, 1/1,000) (red) compared with Mouse IgG1 Isotype Control (green). After incubation of the primary antibody at +4℃ for 30 minutes, the cells were stained with a iFluor™ 488 conjugate-Goat anti-Mouse IgG Secondary antibody (HA1125) at 1/1,000 dilution for 30 minutes at +4℃. Unlabelled sample was used as a control (cells without incubation with primary antibody; black).

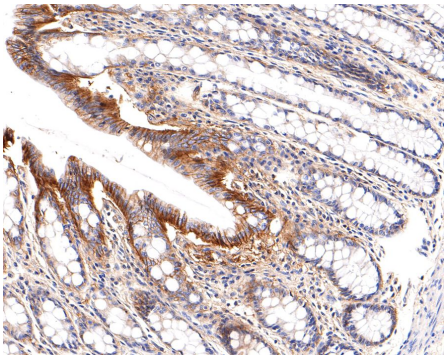

Fig6: Immunohistochemical analysis of paraffin-embedded human colon tissue with Mouse anti-xCT/SLC7A11 antibody (HA600098) at 1/600 dilution.

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 (HA600098) at 1/600 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.

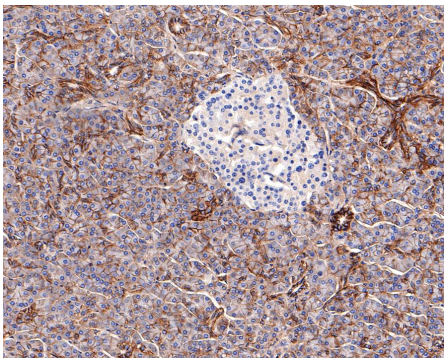

Fig7: Immunohistochemical analysis of paraffin-embedded human pancreas tissue with Mouse anti-xCT/SLC7A11 antibody (HA600098) at 1/200 dilution.

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 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA600098) at 1/200 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.

Note: All products are "FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE".

Background References

1. Koppula P. et. al. Cystine transporter SLC7A11/xCT in cancer: ferroptosis, nutrient dependency, and cancer therapy. Protein Cell. 2021 Aug
2. Lin W. et. al. SLC7A11/xCT in cancer: biological functions and therapeutic implications. Am J Cancer Res. 2020 Oct