Anti-Glucosidase 2 subunit beta Antibody [A6H9] HA600054

Product Type:	Mouse monoclonal IgG1, primary antibodies
Species reactivity:	Human
Applications:	WB, IF-Cell, IHC-P, FC
Molecular Wt:	Predicted band size: 59 kDa
Clone number:	A6H9
Description:	Regulatory subunit of glucosidase II that cleaves sequentially the 2 innermost alpha-1,3- linked glucose residues from the Glc(2)Man(9)GlcNAc(2) oligosaccharide precursor of immature glycoproteins. Required for efficient PKD1/Polycystin-1 biogenesis and trafficking to the plasma membrane of the primary cilia (By similarity). This gene encodes the beta- subunit of glucosidase II, an N-linked glycan-processing enzyme in the endoplasmic reticulum. The encoded protein is an acidic phosphoprotein known to be a substrate for protein kinase C. Mutations in this gene have been associated with the autosomal dominant polycystic liver disease. Alternative splicing results in multiple transcript variants.
lmmunogen:	Recombinant protein within human Glucosidase 2 subunit beta aa 51-250.
Positive control:	HeLa cell lysate, 293T cell lysate, Daudi cell lysate, Jurkat cell lysate, A431 cell lysate, Hela, human placenta tissue.
Subcellular location:	Endoplasmic reticulum.
Database links:	SwissProt: P14314 Human
Recommended Dilutions: WB IF-Cell IHC-P FC	1:500-1:2,000 1:100 1:200 1:500-1:1,000
Storage Buffer:	PBS (pH7.4), 0.05% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.
Storage Instruction:	Shipped at 4 $^\circ\!\!{\rm C}$. Store at +4 $^\circ\!\!{\rm C}$ short term (1-2 weeks). It is recommended to aliquot into single-use upon delivery. Store at -20 $^\circ\!\!{\rm C}$ long term.
Purity:	Protein G affinity purified.

Hangzhou Huaan Biotechnology Co., Ltd.

Orders:0086-571-88062880

Technical:0086-571-89986345

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Applications:WB=Western blot IHC-P=Immunohistochemistry (paraffin) IF-Cell=Immunofluorescence (Cell) IF-Tissue=Immunofluorescence (Tissue) FC=Flow cytometry IP=Immunoprecipitation

Images



HAP1

Glucosidase 2

GAPD

nit hot

kDa WT KD

250 150 100

75

55 45

35

35

Fig1: Western blot analysis of Glucosidase 2 subunit beta on different lysates with Mouse anti-Glucosidase 2 subunit beta antibody (HA600054) at 1/1,000 dilution.

Lane 1: HeLa cell lysate Lane 2: 293T cell lysate Lane 3: Daudi cell lysate Lane 4: Jurkat cell lysate Lane 5: A431 cell lysate

Lysates/proteins at 20 µg/Lane.

Predicted band size: 59 kDa Observed band size: 80 kDa

Exposure time: 5 minutes;

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 (HA600054) at 1/1,000 dilution was used in 5% NFDM/TBST at 4° C overnight. Goat Anti-Mouse IgG - HRP Secondary Antibody (HA1006) at 1/50,000 dilution was used for 1 hour at room temperature.

Fig2: Western blot analysis of Glucosidase 2 subunit beta on different lysates with Mouse anti-Glucosidase 2 subunit beta antibody (HA600054) at 1/2,000 dilution.

Lane 1: HAP1-parental cell lysate Lane 2: HAP1-Glucosidase 2 subunit beta KD cell lysate

Lysates/proteins at 10 µg/Lane.

Predicted band size: 59 kDa Observed band size: 70 kDa

Exposure time: 9 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 (HA600054) at 1/2,000 dilution was used in primary antibody dilution (K1803) at 4° C overnight. Goat Anti-Mouse IgG - HRP Secondary Antibody (HA1006) at 1/50,000 dilution was used for 1 hour at room temperature.

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Fig3: Immunocytochemistry analysis of Hela cells labeling Glucosidase 2 subunit beta with Mouse anti-Glucosidase 2 subunit beta antibody (HA600054) at 1/100 dilution.

Cells were fixed in 4% paraformaldehyde for 15 minutes, permeabilized with 0.1% Triton X-100 in PBS for 15 minutes, and then blocked with 1% BSA for 30 minutes at room temperature. Cells were then incubated with Mouse anti-Glucosidase 2 subunit beta antibody (HA600054) at 1/100 dilution in 1% BSA overnight at 4 $^{\circ}$ C. Goat Anti-Mouse IgG H&L (iFluor M 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.



Fig4: Immunohistochemical analysis of paraffin-embedded human placenta tissue with Mouse anti-Glucosidase 2 subunit beta antibody (HA600054) 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 ddH₂O and PBS, and then probed with the primary antibody (HA600054) 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.



Fig5: Flow cytometric analysis of Hela cells labeling Glucosidase 2 subunit beta.

Cells were fixed and permeabilized. Then stained with the primary antibody (HA600054, 1ug/ml) (red) compared with Mouse IgG1 Isotype Control (green). After incubation of the primary antibody at +4 °C for an hour, the cells were stained with a iFluor TM 488 conjugate-Goat anti-Mouse IgG Secondary antibody (HA1125) at 1/1,000 dilution for 30 minutes at +4 °C. Unlabelled sample was used as a control (cells without incubation with primary antibody; black).

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

Background References

- 1. Huang R. et. al. PRKCSH Alternative Splicing Involves in Silica-Induced Expression of Epithelial-Mesenchymal Transition Markers and Cell Proliferation. Dose Response. 2020 May
- Shin GC. et. al. PRKCSH contributes to tumorigenesis by selective boosting of IRE1 signaling pathway. Nat Commun. 2019 Jul

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