Anti-MRP2 Antibody [JA32-01]

ET1704-47



Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Applications:	WB. IF-Cell. IHC-P. FC
Molecular Wt:	Predicted band size: 174 kDa
Clone number:	JA32-01
Description:	Multi-drug resistance protein 2 (MRP2), also known as ABCC2, is an ATP binding cassette (ABC) transporter responsible for biliary excretion of xenobiotics, endobiotics, and their metabolites. Deficiency in ABCC2 results in the clinical disorder Dubin-Johnson syndrome. MRP2 is found to be expressed in a variety of human cancers, and is associated with resistance of tumor cells to various anticancer drugs including cisplatin. The predicted molecular weight of MRP2 is 174 kDa, while mature MRP2 usually has a slower migration around 190-250 kDa due to the glycosylation.
lmmunogen:	Synthetic peptide within Human MRP2 aa 1496-1545 / 1545.
Positive control:	HepG2 cell lysate, A549 cell lysate, HeLa cell lysate, HepG2, 293T, human liver tissue, human kidney tissue, human pancreas tissue, A549.
Subcellular location:	Apical cell membrane.
Database links:	SwissProt: Q92887 Human
Recommended Dilutions:	
WB	1:2,000
IF-Cell	1:50-1:200
IHC-P	1:50-1:800
FC	1:50-1:100
Storage Buffer:	1*TBS (pH7.4), 0.05% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.
Storage Instruction:	Store at +4 $^\circ\!\!C$ after thawing. Aliquot store at -20 $^\circ\!\!C$ or -80 $^\circ\!\!C$. Avoid repeated freeze / thaw cycles.
Purity:	Protein A 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

Fig1: Western blot analysis of MRP2 on different lysates with Rabbit anti-MRP2 antibody (ET1704-47) at 1/2,000 dilution.

Lane 1: HepG2 cell lysate Lane 2: A549 cell lysate Lane 3: HeLa cell lysate (negative)

Lysates/proteins at 15 µg/Lane.

Predicted band size: 174 kDa Observed band size: 300 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 (ET1704-47) at 1/2,000 dilution was used in 5% NFDM/TBST at 4° C 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 MRP2 on different lysates with Rabbit anti-MRP2 antibody (ET1704-47) at 1/2,000 dilution.

Lane 1: A549-si NT cell lysate (no heat) Lane 2: A549-si MRP2 cell lysate (no heat)

Notice: no heat means the lysate is not boiled.

Lysates/proteins at 10 µg/Lane.

Predicted band size: 174 kDa Observed band size: 300 kDa

Exposure time: 3 minutes; 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 (ET1704-47) at 1/2,000 dilution was used in 5% NFDM/TBST at 4° C overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.



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~300kDa MRP2

GAPDH

72 55

42-35-

25

14



Fig3: ICC staining of MRP2 in HepG2 cells (green). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 1% Blocker BSA for 15 minutes at room temperature. Cells were probed with the primary antibody (ET1704-47, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor®488 Goat anti-Rabbit IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).

Fig4: ICC staining of MRP2 in 293T cells (green). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 1% Blocker BSA for 15 minutes at room temperature. Cells were probed with the primary antibody (ET1704-47, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor®488 Goat anti-Rabbit IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).



Fig5: Immunohistochemical analysis of paraffin-embedded human liver tissue with Rabbit anti-MRP2 antibody (ET1704-47) at 1/800 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 (ET1704-47) at 1/800 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 human kidney tissue with Rabbit anti-MRP2 antibody (ET1704-47) at 1/800 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 (ET1704-47) at 1/800 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.

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Fig7: Immunohistochemical analysis of paraffin-embedded human pancreas tissue using anti-MRP2 antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes. The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ET1704-47, 1/50) for 30 minutes 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: Flow cytometric analysis of MRP2 was done on A549 cells. The cells were fixed, permeabilized and stained with the primary antibody (ET1704-47, 1/50) (red). After incubation of the primary antibody at room temperature for an hour, the cells were stained with a Alexa Fluor 488-conjugated Goat anti-Rabbit IgG Secondary antibody at 1/1000 dilution for 30 minutes.Unlabelled sample was used as a control (cells without incubation with primary antibody; black).

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

Background References

- 1. Ulvestad, M. et al. Drug metabolizing enzyme and transporter protein profiles of hepatocytes derived from human embryonic and induced pluripotent stem cells. Biochemical pharmacology 86: 691-702 (2013).
- 2. Tiwari, AK. et al. Overlapping functions of ABC transporters in topotecan disposition as determined in gene knockout mouse models. Mol. Cancer Ther 12: 1343-55 (2013).

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