Anti-Cytokeratin 18 Antibody [6-19]

M0407-19



Product Type: Mouse monodonal IgG2a, primary antibodies

Species reactivity: Human, Mouse, Rat

Applications: WB, IF-Cell, IHC-P, FC

Molecular Wt: Predicted band size: 48 kDa

Clone number: 6-19

Description: Cytokeratin 18 is an acidic keratin which is found primarily in non squamous epithelia and is present in a majority

of adenocarcinomas and ductal carcinomas but not in squamous cell carcinomas. Cytokeratin 18 exists in combination with Cytokeratin 8, a basic keratin. Hepatocellular carcinomas have been reportedly defined by the

use of antibodies that recognize only Cytokeratins 8 and 18.

Immunogen: Synthetic peptide within mouse Cytokeratin 18 aa 374-423 / 423.

Positive control: HeLa cell lysate, K-562 cell lysate, A431 cell lysate, HT-29 cell lysate, HepG2 cell lysate, HCT 116 cell lysate,

Huh7 cell lysate, HeLa, human breast cancer tissue, human liver tissue, mouse liver tissue, rat liver tissue.

Subcellular location: Cytoplasm, Nucleus.

Database links: SwissProt P05783 Human | P05784 Mouse | Q5BJY9 Rat

Recommended Dilutions:

WB 1:10,000
IF-Cell 1:100
IHC-P 1:200
FC 1:1,000

Storage Buffer: 1*PBS (pH7.4), 0.2% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.

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

Purity: Protein A affinity purified.

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Images

Fig1: Western blot analysis of Cytokeratin 18 on different lysates with Mouse anti-Cytokeratin 18 antibody (M0407-19) at 1/2,000 dilution.

Lane 1: HeLa cell lysate Lane 2: K-562 cell lysate Lane 3: A431 cell lysate Lane 4: HT-29 cell lysate Lane 5: HepG2 cell lysate Lane 6: HCT 116 cell lysate Lane 7: Huh7 cell lysate

Lysates/proteins at 20 µg/Lane.

Predicted band size: 48 kDa Observed band size: 48 kDa

Exposure time: 5 seconds;

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 (M0407-19) at 1/2,000 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.

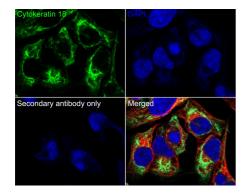


Fig2: Immunocytochemistry analysis of HeLa cells labeling Cytokeratin 18 with Mouse anti-Cytokeratin 18 antibody (M0407-19) at 1/100 dilution.

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-Cytokeratin 18 antibody (M0407-19) at 1/100 dilution in 1% BSA in PBST overnight at 4 °C. 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 $^{\circ}$ C. Goat Anti-Rabbit IgG H&L (iFluor TM 594, HA1122) were used as the secondary antibody at 1/1,000 dilution.

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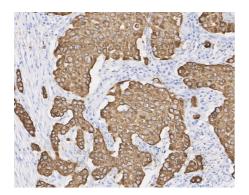


Fig3: Immunohistochemical analysis of paraffin-embedded human breast cancer tissue with Mouse anti-Cytokeratin 18 antibody (M0407-19) at 1/2,000 dilution.

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 ddH₂O and PBS, and then probed with the primary antibody (M0407-19) at 1/2,000 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.

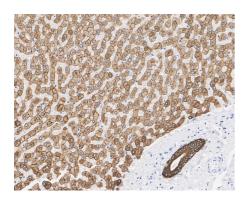


Fig4: Immunohistochemical analysis of paraffin-embedded human liver tissue with Mouse anti-Cytokeratin 18 antibody (M0407-19) at 1/2,000 dilution.

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 ddH₂O and PBS, and then probed with the primary antibody (M0407-19) at 1/2,000 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.

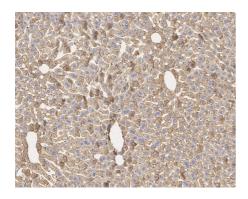


Fig5: Immunohistochemical analysis of paraffin-embedded mouse liver tissue with Mouse anti-Cytokeratin 18 antibody (M0407-19) at 1/2,000 dilution.

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 ddH₂O and PBS, and then probed with the primary antibody (M0407-19) at 1/2,000 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.

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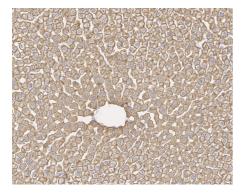


Fig6: Immunohistochemical analysis of paraffin-embedded rat liver tissue with Mouse anti-Cytokeratin 18 antibody (M0407-19) at 1/2,000 dilution.

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 ddH₂O and PBS, and then probed with the primary antibody (M0407-19) at 1/2,000 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.

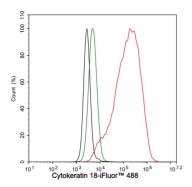


Fig7: Flow cytometric analysis of HeLa cells labeling Cytokeratin 18.

Cells were fixed and permeabilized. Then stained with the primary antibody (M0407-19, 1µg/mL) (red) compared with Mouse IgG1 Isotype Control (green). After incubation of the primary antibody at +4 $^{\circ}$ 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 $^{\circ}$ C. 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. Korver S et al. The application of cytokeratin-18 as a biomarker for drug-induced liver injury. Arch Toxicol. 2021 Nov
- 2. Goralska J et al. Plasma Cytokeratin-18 Fragment Level Reflects the Metabolic Phenotype in Obesity. Biomolecules. 2023 Apr