Anti-Cytokeratin 7 Antibody [ST50-05]

ET1609-62

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse
Applications:	WB, IF-Cell, IF-Tissue, IHC-P, IP, FC
Molecular Wt:	Predicted band size: 51 kDa
Clone number:	ST50-05

Description: Cytokeratins comprise a diverse group of intermediate filament proteins (IFPs) that are expressed as pairs in both keratinized and non-keratinized epithelial tissue, where they constitute up to 85% of mature keratinocytes in the vertebrate epidermis. Cytokeratins play a critical role in differentiation and tissue specialization and function to maintain the overall structural integrity of epithelial cells. The a-helical coiled-coil dimers associate laterally end-to-end to form 10 nm diameter filaments. Cytokeratins are useful markers of tissue differentiation and, in addition, they aid in the characterization of malignant tumors. Cytokeratin 7 (also known as sarcolectin) agglutinates normal and transformed cells with a high affinity for simple sugars. Cytokeratin 7 also inhibits the synthesis of interferon-dependent secondary proteins thus reversing the antiviral effect of interferon induction and restoring cells to their status ad primum. In normal and transformed cells, Cytokeratin 7 localizes to the membrane.

Immunogen: Synthetic peptide within Human Cytokeratin 7 aa 18-67.

Positive control: HeLa cell lysate, A549 cell lysate, Hela, A549, BT-20, human breast tissue, human breast carcinoma, human liver tissue, human placenta tissue, SK-BR-3.

Subcellular location: Cytoplasm.

Database links: SwissProt: P08729 Human | Q9DCV7 Mouse

Recommended Dilutions:

WB	1:5,000
IF-Cell	1:100-1:400
IF-Tissue	1:400
IHC-P	1:50-1:1,500
FC	1:1000
IP	Use at an assay dependent concentration.

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

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

Purity: Protein A affinity purified.

Images

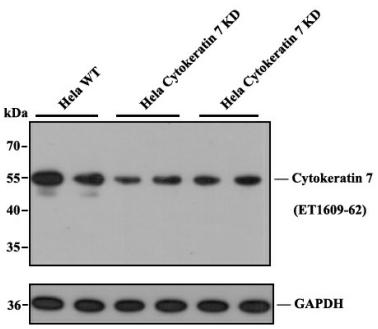

Fig1: All lanes: Western blot analysis of Cytokeratin 7 with anti-Cytokeratin 7 antibody [ST50-05] (ET1609-62) at 1:1,000 dilution.
Lane 1/2: Wild-type Hela whole cell lysate (20 µg).
Lane 3/4: Cytokeratin 7 fragment 1 knockdown Hela whole cell lysate (20 µg).
Lane 5/6: Cytokeratin 7 fragment 2 knockdown Hela whole cell lysate (20 µg).

ET1609-62 was shown to specifically react with Cytokeratin 7 in wild-type Hela cells. Weakened bands were observed when Cytokeratin 7 knockdown samples were tested. Wild-type and Cytokeratin 7 knockdown samples were subjected to SDS-PAGE. Proteins were transferred to a PVDF membrane and blocked with 5% NFDM in TBST for 1 hour at room temperature. The primary antibody (ET1609-62, 1/1,000) and Loading control antibody (Rabbit anti-GAPDH , ET1601-4, 1/10,000) were used in 5% BSA at room temperature for 2 hours. Goat Anti-Rabbit IgG-HRP Secondary Antibody (HA1001) at 1:200,000 dilution was used for 1 hour at room temperature.

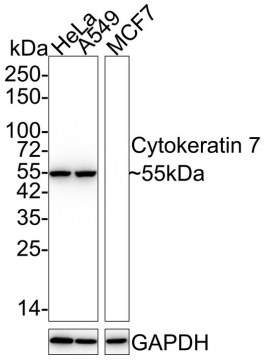

Fig2: Western blot analysis of Cytokeratin 7 on different lysates with Rabbit anti-Cytokeratin 7 antibody (ET1609-62) at 1/5,000 dilution.

Lane 1: HeLa cell lysate
Lane 2: A549 cell lysate
Lane 3: MCF7 cell lysate (negative)

Lysates/proteins at 15 µg/Lane.

Predicted band size: 51 kDa
Observed band size: 55 kDa

Exposure time: 20 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 (ET1609-62) at 1/5,000 dilution was used in 5% NFDM/TBST at 4℃ overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

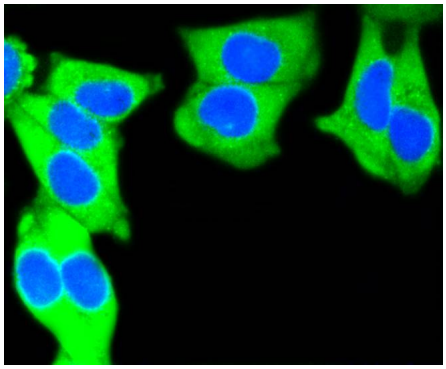

Fig3: Immunocytochemistry staining of Cytokeratin 7 in Hela cells (green). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 1% Blocker BSA for 15 minutes at room temperature. Cells were probed with the primary antibody (ET1609-62, 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).

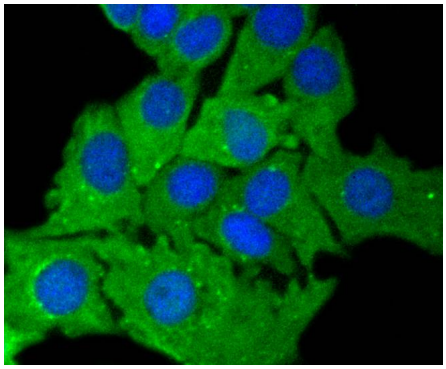

Fig4: Immunocytochemistry staining of Cytokeratin 7 in BT-20 cells (green). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 1% Blocker BSA for 15 minutes at room temperature. Cells were probed with the primary antibody (ET1609-62, 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).

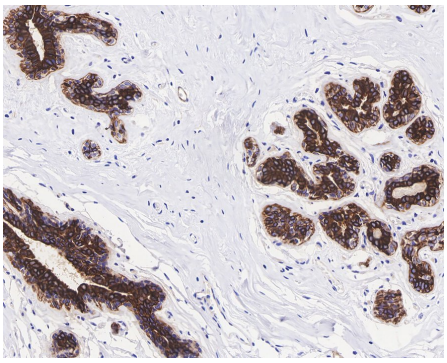

Fig5: Immunohistochemical analysis of paraffin-embedded human breast tissue with Rabbit anti-Cytokeratin 7 antibody (ET1609-62) at 1/1,500 dilution.

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 (ET1609-62) at 1/1,500 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.

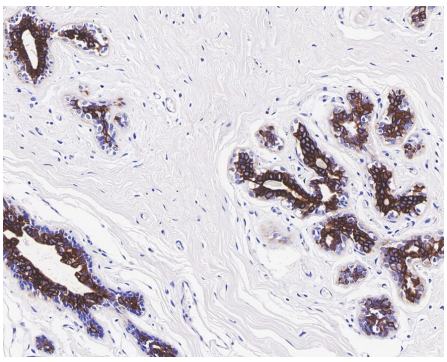

Fig6: Immunohistochemical analysis of paraffin-embedded human breast tissue with Rabbit anti-Cytokeratin 7 antibody (ET1609-62) at 1/1,500 dilution.

The section was not undergone antigen retrieval. 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 (ET1609-62) at 1/1,500 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.

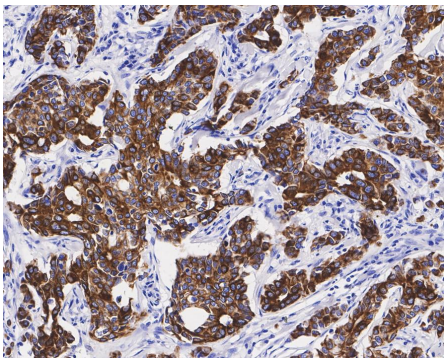

Fig7: Immunohistochemical analysis of paraffin-embedded human breast carcinoma tissue with Rabbit anti-Cytokeratin 7 antibody (ET1609-62) at 1/1,500 dilution.

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 (ET1609-62) at 1/1,500 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.

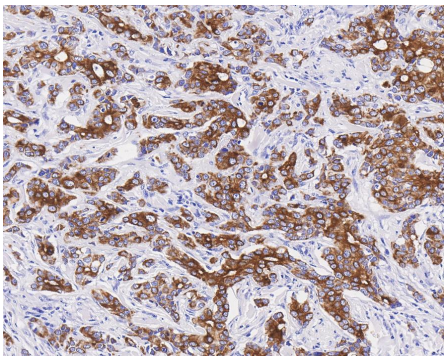

Fig8: Immunohistochemical analysis of paraffin-embedded human breast carcinoma tissue with Rabbit anti-Cytokeratin 7 antibody (ET1609-62) at 1/400 dilution.

The section was not undergone antigen retrieval. 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 (ET1609-62) at 1/400 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.

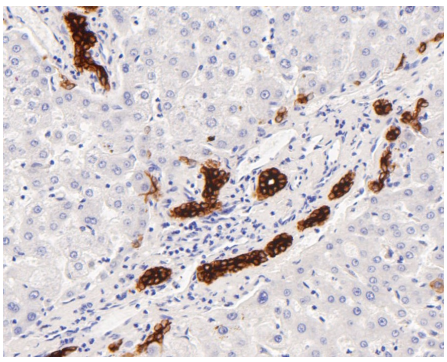

Fig9: Immunohistochemical analysis of paraffin-embedded human liver tissue using anti-Cytokeratin 7 antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes.The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1609-62, 1/200) 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.

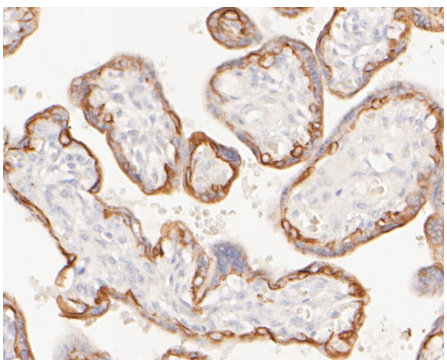

Fig10: Immunohistochemical analysis of paraffin-embedded human placenta tissue using anti-Cytokeratin 7 antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes.The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1609-62, 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.

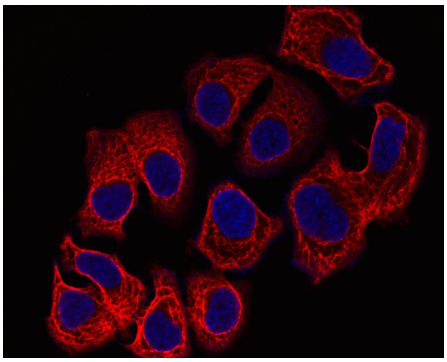

Fig11: Immunocytochemistry analysis of SKBR-3 cells labeling Cytokeratin 7.
Cells were fixed in 4% paraformaldehyde and permeabilized with 0.1% Triton X-100 in PBS for 10 minutes and blocked with 1% BSA for 15 minutes at room temperature. Cells were probed with the primary antibodies Cytokeratin 7 (ET1609-62, red) at 1/100 dilution for overnight at 4 ℃. Goat anti rabbit IgG (iFluorTM 594) (HA1122) was used as the secondary antibody at 1/1,000 dilution dilution. DAPI was used as nuclear counterstain.

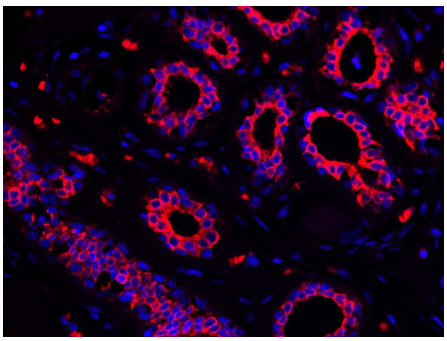

Fig12: Immunofluorescence analysis of paraffin-embedded human breast tissue labeling Cytokeratin 7 (ET1609-62).

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 10% negative goat serum for 1 hour at room temperature, washed with PBS. And then probed with the primary antibodies Cytokeratin 7 (ET1609-62, red) at 1/400 dilution at +4℃ overnight, washed with PBS.

Goat Anti-Rabbit IgG H&L (iFluor™ 594, HA1122) was used as the secondary antibodies at 1/1,000 dilution. Nuclei were counterstained with DAPI (blue).

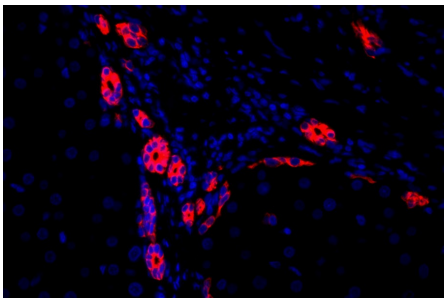

Fig13: Immunofluorescence analysis of paraffin-embedded human liver tissue labeling Cytokeratin 7 (ET1609-62).

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 10% negative goat serum for 1 hour at room temperature, washed with PBS. And then probed with the primary antibodies Cytokeratin 7 (ET1609-62, red) at 1/400 dilution at +4℃ overnight, washed with PBS.

Goat Anti-Rabbit IgG H&L (iFluor™ 594, HA1122) was used as the secondary antibodies at 1/1,000 dilution. Nuclei were counterstained with DAPI (blue).

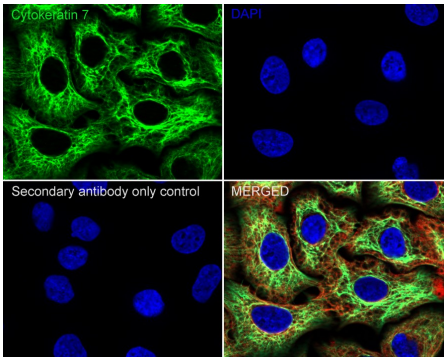

Fig14: Immunocytochemistry analysis of A549 cells labeling Cytokeratin 7 with Rabbit anti-Cytokeratin 7 antibody (ET1609-62) at 1/100 dilution.

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 Rabbit anti-Cytokeratin 7 antibody (ET1609-62) at 1/100 dilution in 1% BSA in PBST overnight at 4 ℃. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) 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 (M1305-2, red) was stained at 1/100 dilution overnight at +4℃. Goat Anti-Mouse IgG H&L (iFluor™ 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

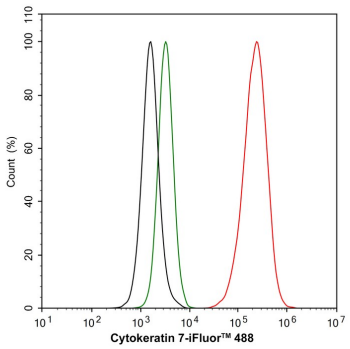

Fig15: Flow cytometric analysis of A549 cells labeling Cytokeratin 7.

Cells were fixed and permeabilized. Then stained with the primary antibody (ET1609-62, 1μg/mL) (red) compared with Rabbit IgG Isotype Control (green). After incubation of the primary antibody at +4℃ for an hour, the cells were stained with a iFluor™ 488 conjugate-Goat anti-Rabbit IgG Secondary antibody (HA1121) at 1/1,000 dilution for 30 minutes at +4℃. 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. Hrudka J. et al. Cytokeratin 7 expression as a predictor of an unfavorable prognosis in colorectal carcinoma. Sci Rep. 2021 Sep
2. Statz E. et al. Cytokeratin 7, GATA3, and SOX-10 is a Comprehensive Panel in Diagnosing Triple Negative Breast Cancer Brain Metastases. Int J Surg Pathol. 2021 Aug