

# Anti-Cytokeratin 7 Antibody [ST50-05]

ET1609-62



|                            |   |
|----------------------------|---|
| <b>Product Type:</b>       | Recombinant Rabbit monoclonal IgG, primary antibodies |
| <b>Species reactivity:</b> | Human, Mouse  |
| <b>Applications:</b>       | WB, IF-Cell, IF-Tissue, IHC-P, IP, FC, mlHC           |
| <b>Molecular Wt:</b>       | Predicted band size: 51 kDa                           |
| <b>Clone number:</b>       | 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  $\alpha$ -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, A549, human breast tissue, human breast carcinoma tissue, SK-Br-3, human liver tissue, human gastric cancer tissue.

**Subcellular location:** Cytoplasm.

**Database links:** SwissProt: P08729 Human | Q9DCV7 Mouse

**Recommended Dilutions:**

|                  |  |
|------------------|--|
| <b>WB</b>        | 1:5,000                                  |
| <b>IF-Cell</b>   | 1:100                                    |
| <b>IF-Tissue</b> | 1:400                                    |
| <b>IHC-P</b>     | 1:1,000-1:2,000                          |
| <b>FC</b>        | 1:1,000                                  |
| <b>IP</b>        | Use at an assay dependent concentration. |
| <b>mlHC</b>      | 1:1,000                                  |

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

**Storage Instruction:** Shipped at 4°C. Store at +4°C short term (1-2 weeks). It is recommended to aliquot into single-use upon delivery. Store at -20°C long term.

**Purity:** Protein A affinity purified.

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Orders:0086-571-88062880

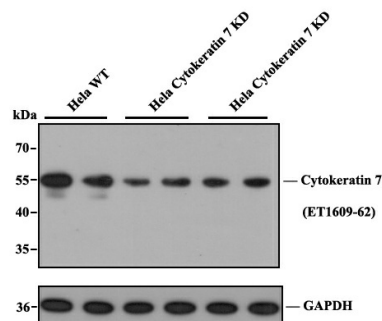
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Applications:WB=Western blot IHC-P=Immunohistochemistry (paraffin) IF-Cell=Immunofluorescence (Cell) IF-Tissue=Immunofluorescence (Tissue) FC=Flow cytometry IP=Immunoprecipitation

## Images



**Fig1:** 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).

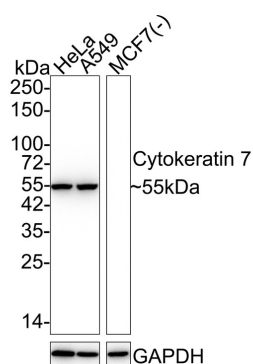
ET1609-62 was shown to specifically react with Cytokeratin 7 in wild-type HeLa cells. Weakened bands were observed when Cytokeratin 7 knockdown samples were tested. Wild-type and Cytokeratin 7 knockdown samples were subjected to SDS-PAGE. Proteins were transferred to a PVDF membrane and blocked with 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°C overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

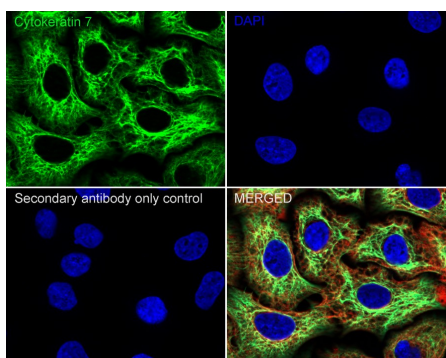
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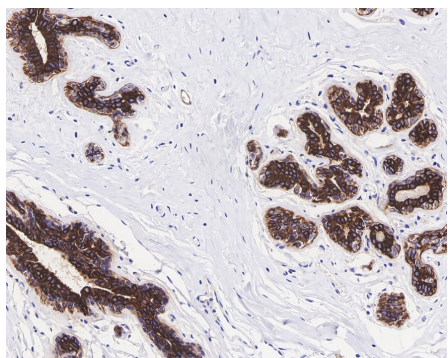
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**Fig3:** Immunocytochemistry analysis of A549 cells labeling Cytokeratin 7 with Rabbit anti-Cytokeratin 7 antibody (ET1609-62) at 1/100 dilution.

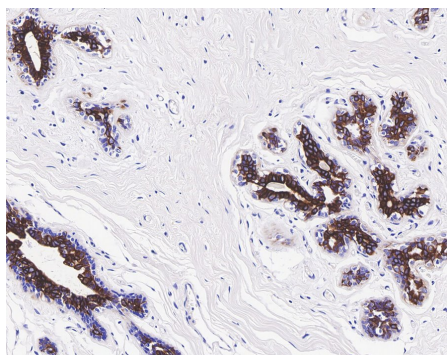
Cells were fixed in 4% paraformaldehyde for 20 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 5 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Rabbit anti-Cytokeratin 7 antibody (ET1609-62) at 1/100 dilution in 1% BSA in PBST overnight at 4 °C. 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°C. Goat Anti-Mouse IgG H&L (iFluor™ 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.



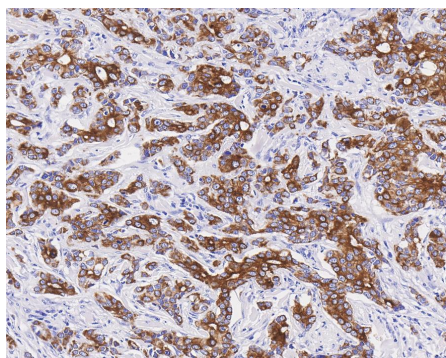
**Fig4:** Immunohistochemical analysis of paraffin-embedded human breast tissue with Rabbit anti-Cytokeratin 7 antibody (ET1609-62) at 1/1,500 dilution.

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<sub>2</sub>O 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.



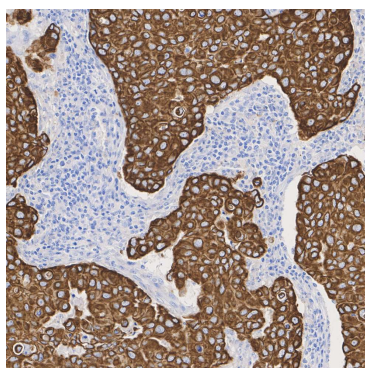
**Fig5:** Immunohistochemical analysis of paraffin-embedded human breast tissue with Rabbit anti-Cytokeratin 7 antibody (ET1609-62) at 1/1,500 dilution.

**The section was not undergone antigen retrieval.** The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH<sub>2</sub>O 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.



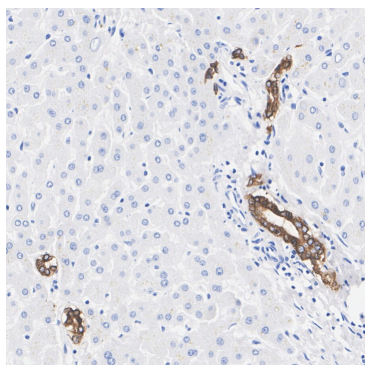
**Fig6:** Immunohistochemical analysis of paraffin-embedded human breast carcinoma tissue with Rabbit anti-Cytokeratin 7 antibody (ET1609-62) at 1/400 dilution.

**The section was not undergone antigen retrieval.** The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH<sub>2</sub>O 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.



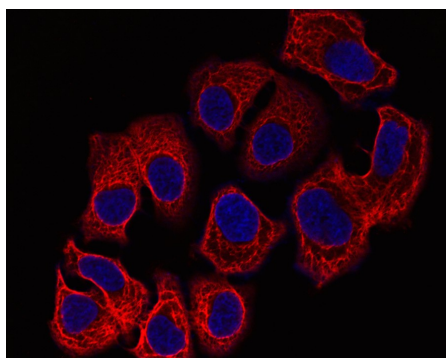
**Fig7:** Immunohistochemical analysis of paraffin-embedded human breast carcinoma tissue with Rabbit anti-Cytokeratin 7 antibody (ET1609-62) at 1/1,500 dilution.

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<sub>2</sub>O 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.



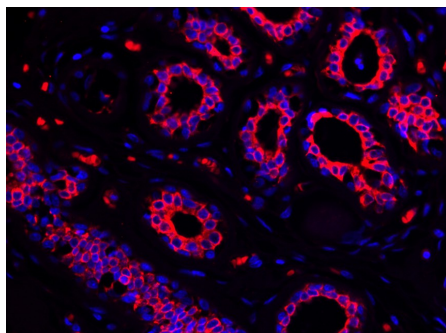
**Fig8:** Immunohistochemical analysis of paraffin-embedded human liver tissue with Rabbit anti-Cytokeratin 7 antibody (ET1609-62) at 1/2,000 dilution.

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 ddH<sub>2</sub>O and PBS, and then probed with the primary antibody (ET1609-62) 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.



**Fig9:** Immunocytochemistry analysis of SK-Br-3 cells labeling Cytokeratin 7.

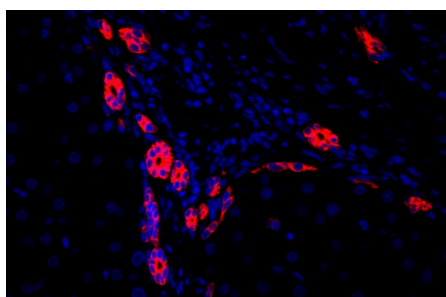
Cells were fixed in 4% paraformaldehyde and permeabilized with 0.1% Triton X-100 in PBS for 10 minutes and blocked with 1% BSA for 15 minutes at room temperature. Cells were probed with the primary antibodies Cytokeratin 7 (ET1609-62, red) at 1/100 dilution for overnight at 4 °C. Goat anti rabbit IgG (iFluor™ 594) (HA1122) was used as the secondary antibody at 1/1,000 dilution. DAPI was used as nuclear counterstain.



**Fig10:** 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°C overnight, washed with PBS.

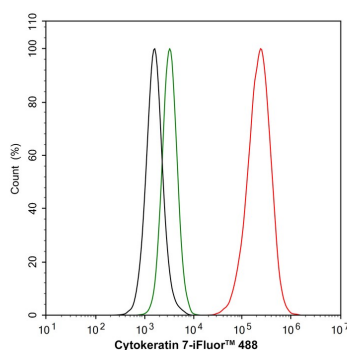
Goat Anti-Rabbit IgG H&L (iFluor™ 594, HA1122) was used as the secondary antibodies at 1/1,000 dilution. Nuclei were counterstained with DAPI (blue).



**Fig11:** Immunofluorescence analysis of paraffin-embedded human liver tissue labeling Cytokeratin 7 (ET1609-62).

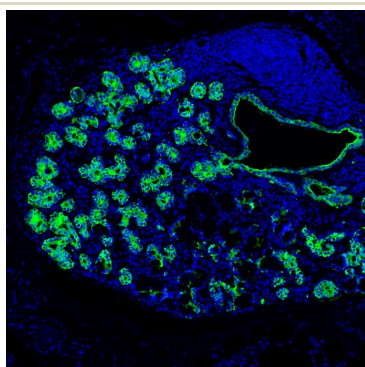
The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 10% negative goat serum for 1 hour at room temperature, washed with PBS. And then probed with the primary antibodies Cytokeratin 7 (ET1609-62, red) at 1/400 dilution at +4°C 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).



**Fig12:** 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°C 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°C. Unlabelled sample was used as a control (cells without incubation with primary antibody; black).



**Fig13:** mIHC analysis of human gastric cancer tissue (Formalin/PFA-fixed paraffin-embedded sections) with Rabbit anti-Cytokeratin 7 antibody (ET1609-62) at 1/1,000 dilution. The immunostaining was performed with the IRISKit® HyperView mTSA Kit (MH900206). Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 30 mins at 95°C. DAPI (blue) was used as a nuclear counter stain. Image acquisition was performed with Olympus VS200 Slide Scanner.

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

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### 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

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