Anti-pan Cytokeratin Antibody [PDH09-10]

HA601138

Product Type:	Recombinant Mouse monoclonal IgG1, primary antibodies
Species reactivity:	Human
Applications:	IHC-P, mIHC, IF-Cell, IF-Tissue, WB, FC
Clone number:	PDH09-10

Description: Cytokeratins comprise a diverse group of intermediate filament proteins (IFPs) that are expressed as pairs in both keratinized and non-keratinized epithelial tissue. Cytokeratins play a critical role in differentiation and tissue specialization and function to maintain the overall structural integrity of epithelial cells. Cytokeratins have been found to be useful markers of tissue differentiation which is directly applicable to the characterization of malignant tumors. For example, cytokeratins 10 and 13 are expressed highly in a subset of squamous cell carcinomas while cytokeratin 18 is expressed in a majority of adenocarcinomas and basal cell carcinomas. Since each epithelium contains at least one acidic and one basic keratin, this antibody is used to observe the distribution of keratin-containing cells in normal epithelia and to identify neoplasms derived from such epithelium. Recognizes humancytokeratins 10, 14, 15, 16 and 19, and cytokeratins 1, 2, 3, 4, 5, 6, 7 and 8.

Immunogen: Full length native protein corresponding to Human pan Cytokeratin.

Positive control: Human non-small cell lung cancer, human gastric cancer, human cervical cancer, human stomach carcinoma tissue, human colon carcinoma tissue, human liver tissue, human small intestine tissue, A431.

Subcellular location: Cytoplasmic

Database links: SwissProt: Q01546 Human | Q7Z794 Human | P48668 Human | P13645 Human

Recommended Dilutions:

IHC-P	1:1,000-1:2,000
mIHC	1:3,000
IF-Cell	1:100
IF-Tissue	1:200
WB	1:2,000
FC	1:1,000

Storage Buffer: PBS (pH7.4), 0.1% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.

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

Purity: Protein A affinity purified.

Images

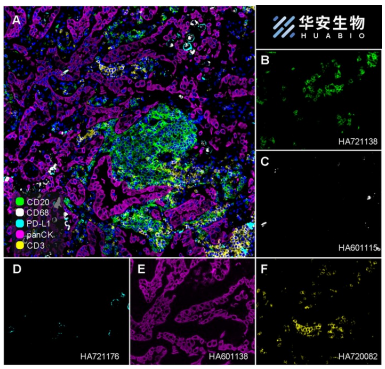

Fig1: Fluorescence multiplex immunohistochemical analysis of the human non-small cell lung cancer (Formalin/PFA-fixed paraffin-embedded sections). Panel A: the merged image of anti-CD20 (HA721138, green), anti-CD68 (HA601115, gray), anti-PD-L1 (HA721176, cyan), anti-panCK (HA601138, magenta) and anti-CD3 (HA720082, yellow) on human non-small cell lung cancer. Panel B: anti- CD20 stained on B cells. Panel C: anti-CD68 stained on macrophage M1 and macrophage M2. Panel D: anti-PD-L1 stained on dendritic cells and macrophages cells. Panel E: anti-panCK stained on cancer cells. Panel F: anti-CD3 stained on T cells. HRP Conjugated UltraPolymer Goat Polyclonal Antibody HA1119/HA1120 was used as a secondary antibody. The immunostaining was performed with the Sequential Immuno-staining Kit (IRISKit™MH010101, www.luminiris.cn). The section was incubated in five rounds of staining: in the order of HA721138 (1/1,500 dilution), HA601115 (1/2,000 dilution), HA721176 (1/1,000 dilution), HA601138 (1/3,000 dilution), and HA720082 (1/500 dilution) for 20 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system. Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 30 mins at 95℃. DAPI (blue) was used as a nuclear counter stain. Image acquisition was performed with Olympus VS200 Slide Scanner.

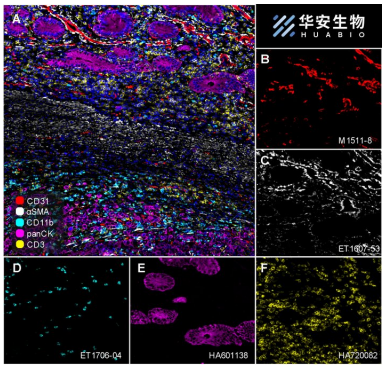

Fig2: Fluorescence multiplex immunohistochemical analysis of the human gastric cancer (Formalin/PFA-fixed paraffin-embedded sections). Panel A: the merged image of anti-CD31 (M1511-8, red), anti-αSMA (ET1607-53, gray), anti-CD11b (ET1706-04, cyan), anti-panCK (HA601138, magenta) and anti-CD3 (HA720082, yellow) on human gastric cancer. Panel B: anti-CD31 stained on the endothelial cells. Panel C: anti-αSMA stained on cancer-associated fibroblasts and smooth muscle cells. Panel D: anti-CD11b stained on myeloid cells. Panel E: anti-panCK stained on cancer cells. Panel F: anti-CD3 stained on T cells. HRP Conjugated UltraPolymer Goat Polyclonal Antibody HA1119/HA1120 was used as a secondary antibody. The immunostaining was performed with the Sequential Immuno-staining Kit (IRISKit™MH010101, www.luminiris.cn). The section was incubated in five rounds of staining: in the order of M1511-8 (1/1,000 dilution), ET1607-53 (1/2,000 dilution), ET1706-04 (1/1,000 dilution), HA601138 (1/3,000 dilution), and HA720082 (1/500 dilution) for 20 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system. Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 30 mins at 95℃. DAPI (blue) was used as a nuclear counter stain. Image acquisition was performed with Olympus VS200 Slide Scanner.

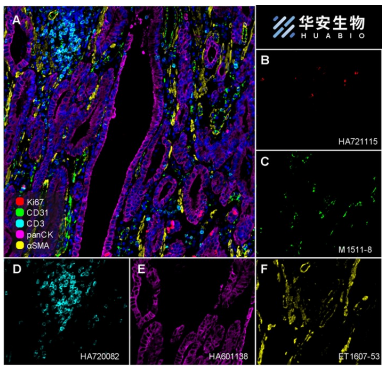

Fig3: Fluorescence multiplex immunohistochemical analysis of the human gastric cancer (Formalin/PFA-fixed paraffin-embedded sections). Panel A: the merged image of anti-Ki67 (HA721115, red), anti-CD31 (M1511-8, green), anti-CD3 (HA720082, cyan), anti-panCK (HA601138, magenta) and anti-αSMA (ET1607-53, yellow) on human gastric cancer. Panel B: anti- Ki67 stained on cells in G1, S, G2 and M phases of cell cycle. Panel C: anti-CD31 stained on the endothelial cells. Panel D: anti-CD3 stained on T cells. Panel E: anti-panCK stained on cancer cells. Panel F: anti-αSMA stained on cancer-associated fibroblasts and smooth muscle cells. HRP Conjugated UltraPolymer Goat Polyclonal Antibody HA1119/HA1120 was used as a secondary antibody. The immunostaining was performed with the Sequential Immuno-staining Kit (IRISKit™MH010101, www.luminiris.cn). The section was incubated in five rounds of staining: in the order of HA721115 (1/2,000 dilution), M1511-8 (1/1,000 dilution), HA720082 (1/500 dilution), HA601138 (1/3,000 dilution), and ET1607-53 (1/2,000 dilution) for 20 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system. Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 30 mins at 95℃. DAPI (blue) was used as a nuclear counter stain. Image acquisition was performed with Olympus VS200 Slide Scanner.

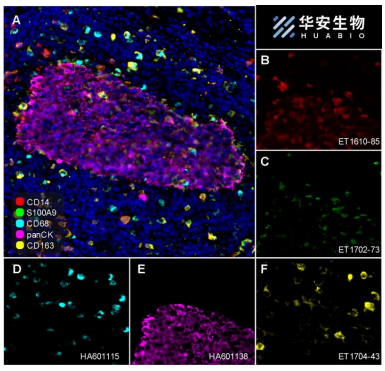

Fig4: Fluorescence multiplex immunohistochemical analysis of the human cervical cancer (Formalin/PFA-fixed paraffin-embedded sections). Panel A: the merged image of anti-CD14 (ET1610-85, red), anti-S100A9 (ET1702-73, green), anti-CD68 (HA601115, cyan), anti-panCK (HA601138, magenta) and anti-CD163 (ET1704-43, yellow) on human cervical cancer. Panel B: anti-CD14 stained on monocyte and MDSCs. Panel C: anti-S100A9 stained on MDSCs. Panel D: anti-CD68 stained on macrophage M1 and macrophage M2. Panel E: anti-panCK stained on tumor cells. Panel F: anti-CD163 stained on macrophage M2. HRP Conjugated UltraPolymer Goat Polyclonal Antibody HA1119/HA1120 was used as a secondary antibody. The immunostaining was performed with the Sequential Immuno-staining Kit (IRISKit™MH010101, www.luminiris.cn). The section was incubated in five rounds of staining: in the order of ET1610-85 (1/1,000 dilution), ET1702-73 (1/1,000 dilution), HA601115 (1/2,000 dilution), HA601138 (1/3,000 dilution), and ET1704-43 (1/2,000 dilution) for 20 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system. Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 30 mins at 95℃. DAPI (blue) was used as a nuclear counter stain. Image acquisition was performed with Olympus VS200 Slide Scanner.

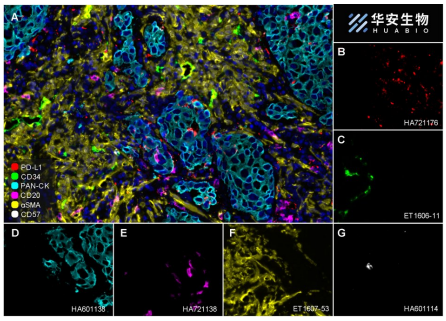

Fig5: Fluorescence multiplex immunohistochemical analysis of Human non-small cell lung cancer (Formalin/PFA-fixed paraffin-embedded sections). Panel A: the merged image of anti-PD-L1 (HA721176, red), anti-CD34 (ET1606-11, green), anti-Pan-CK (HA601138, cyan), anti-CD20 (HA721138, magenta), anti-αSMA (ET1607-53, yellow) and anti-CD57 (HA601114, white) on NSCLC. Panel B: anti-PD-L1 stained on dendritic cells and macrophages cells. Panel C: anti- CD34 stained on endothelial cells. Panel D: anti-Pan-CK stained on cancer cells. Panel E: CD20 stained on B cells. Panel F: anti-αSMA stained on cancer-associated fibroblasts and smooth muscle cells. Panel G: anti-CD57 stained on NK cells and T cells. HRP Conjugated UltraPolymer Goat Polyclonal Antibody HA1119/HA1120 was used as a secondary antibody. The immunostaining was performed with the Sequential Immuno-staining Kit (IRISKit™MH010101, www.luminiris.cn). The section was incubated in six rounds of staining: in the order of HA721176 (1/1,000 dilution), ET1606-11 (1/1,000 dilution), HA601138 (1/3,000 dilution), HA721138 (1/2,000 dilution), ET1607-53 (1/3,000 dilution) and HA601114 (1/1,000 dilution) for 20 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system. Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 30 mins at 95℃. DAPI (blue) was used as a nuclear counter stain. Image acquisition was performed with Olympus VS200 Slide Scanner.

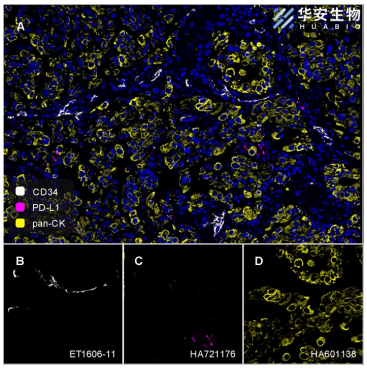

Fig6: Fluorescence multiplex immunohistochemical analysis of human non-small cell lung cancer (Formalin/PFA-fixed paraffin-embedded sections). Panel A: the merged image of anti-CD34 (ET1606-11, White), anti-PD-L1 (HA721176, Violet) and anti-pan Cytokeratin (HA601138, Yellow) on NSCLC. HRP Conjugated UltraPolymer Goat Polyclonal Antibody HA1119/HA1120 was used as a secondary antibody. The immunostaining was performed with the Sequential Immuno-staining Kit (IRISKit™MH010101, www.luminiris.cn). The section was incubated in three rounds of staining: in the order of ET1606-11 (1/2,000 dilution), HA721176 (1/1,000 dilution) and HA601138 (1/3,000 dilution) for 20 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system. Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 30 mins at 95℃. DAPI (blue) was used as a nuclear counter stain. Image acquisition was performed with Zeiss Observer 7 Inverted Fluorescence Microscope.

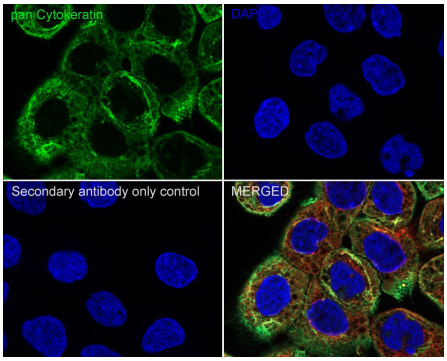

Fig7: Immunocytochemistry analysis of A431 cells labeling pan Cytokeratin with Mouse anti-pan Cytokeratin antibody (HA601138) at 1/100 dilution.

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-pan Cytokeratin antibody (HA601138) at 1/100 dilution in 1% BSA in PBST overnight at 4 ℃. 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℃. Goat Anti-Rabbit IgG H&L (iFluor™ 594, HA1122) were used as the secondary antibody at 1/1,000 dilution.

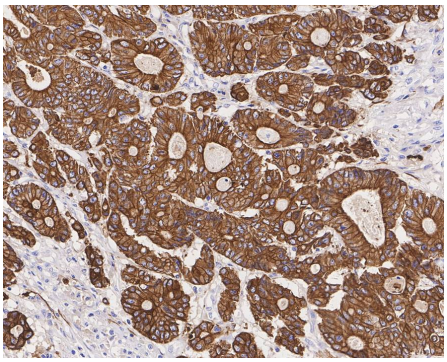

Fig8: Immunohistochemical analysis of paraffin-embedded human colon carcinoma tissue with Mouse anti-pan Cytokeratin antibody (HA601138) at 1/2,000 dilution.

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_2O and PBS, and then probed with the primary antibody (HA601138) 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.

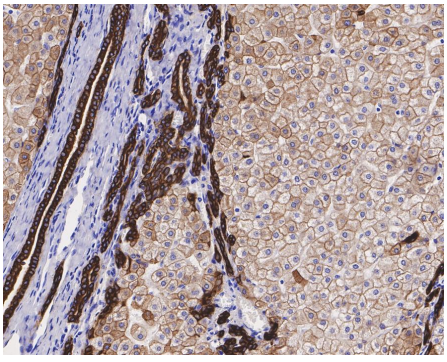

Fig9: Immunohistochemical analysis of paraffin-embedded human liver tissue with Mouse anti-pan Cytokeratin antibody (HA601138) at 1/2,000 dilution.

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_2O and PBS, and then probed with the primary antibody (HA601138) 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.

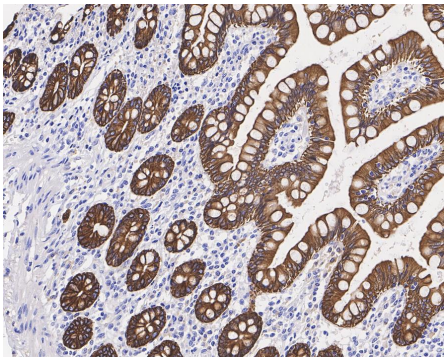

Fig10: Immunohistochemical analysis of paraffin-embedded human small intestine tissue with Mouse anti-pan Cytokeratin antibody (HA601138) at 1/2,000 dilution.

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_2O and PBS, and then probed with the primary antibody (HA601138) 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.

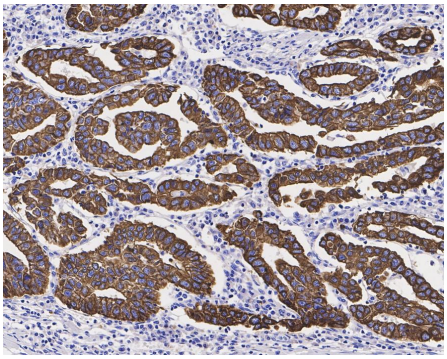

Fig11: Immunohistochemical analysis of paraffin-embedded human stomach carcinoma tissue with Mouse anti-pan Cytokeratin antibody (HA601138) at 1/1,000 dilution.

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_2O and PBS, and then probed with the primary antibody (HA601138) at 1/1,000 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.

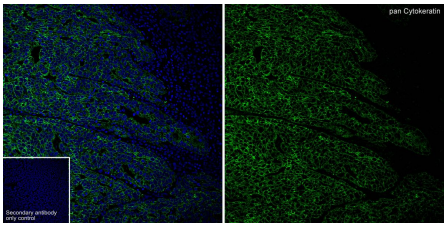

Fig12: Immunofluorescence analysis of paraffin-embedded human stomach cancer tissue labeling pan Cytokeratin with Mouse anti-pan Cytokeratin antibody (HA601138) at 1/200 dilution.

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 10% negative goat serum for 1 hour at room temperature, washed with PBS, and then probed with the primary antibody (HA601138, green) at 1/200 dilution overnight at 4 ℃, washed with PBS. Goat Anti-Mouse IgG H&L (iFluor™ 488, HA1125) was used as the secondary antibody at 1/1,000 dilution. Nuclei were counterstained with DAPI (blue).

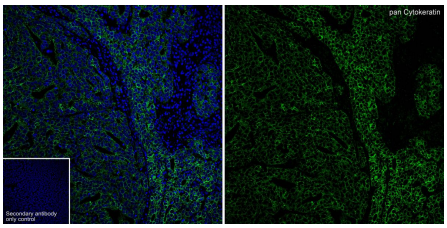

Fig13: Immunofluorescence analysis of paraffin-embedded human stomach cancer tissue labeling pan Cytokeratin with Mouse anti-pan Cytokeratin antibody (HA601138) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) overnight at 60℃ water bath. The tissues were blocked in 10% negative goat serum for 1 hour at room temperature, washed with PBS, and then probed with the primary antibody (HA601138, green) at 1/200 dilution overnight at 4 ℃, washed with PBS. Goat Anti-Mouse IgG H&L (iFluor™ 488, HA1125) was used as the secondary antibody at 1/1,000 dilution. Nuclei were counterstained with DAPI (blue).

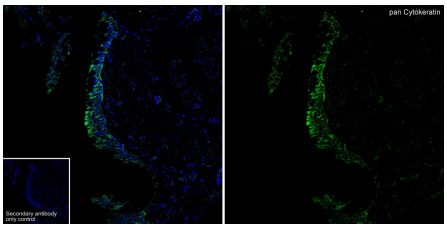

Fig14: Immunofluorescence analysis of paraffin-embedded human trachea tissue labeling pan Cytokeratin with Mouse anti-pan Cytokeratin antibody (HA601138) at 1/200 dilution.

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 10% negative goat serum for 1 hour at room temperature, washed with PBS, and then probed with the primary antibody (HA601138, green) at 1/200 dilution overnight at 4 ℃, washed with PBS. Goat Anti-Mouse IgG H&L (iFluor™ 488, HA1125) was used as the secondary antibody at 1/1,000 dilution. Nuclei were counterstained with DAPI (blue).

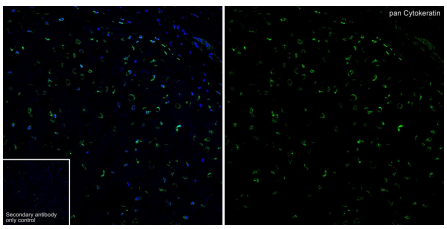

Fig15: Immunofluorescence analysis of paraffin-embedded human tracheal cartilage tissue labeling pan Cytokeratin with Mouse anti-pan Cytokeratin antibody (HA601138) at 1/200 dilution.

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 10% negative goat serum for 1 hour at room temperature, washed with PBS, and then probed with the primary antibody (HA601138, green) at 1/200 dilution overnight at 4 ℃, washed with PBS. Goat Anti-Mouse IgG H&L (iFluor™ 488, HA1125) was used as the secondary antibody at 1/1,000 dilution. Nuclei were counterstained with DAPI (blue).

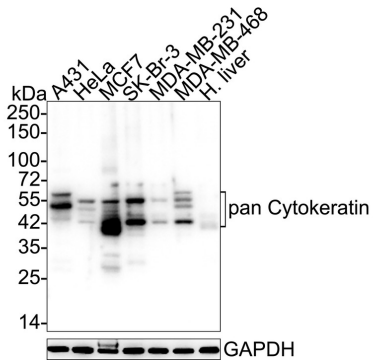

Fig16: Western blot analysis of pan Cytokeratin on different lysates with Mouse anti-pan Cytokeratin antibody (HA601138) at 1/2,000 dilution.

Lane 1: A431 cell lysate (10 µg/Lane)
Lane 2: HeLa cell lysate (10 µg/Lane)
Lane 3: MCF7 cell lysate (10 µg/Lane)
Lane 4: SK-Br-3 cell lysate (10 µg/Lane)
Lane 5: MDA-MB-231 cell lysate (10 µg/Lane)
Lane 6: MDA-MB-468 cell lysate (10 µg/Lane)
Lane 7: Human liver tissue lysate (20 µg/Lane)

Exposure time: 4 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 (HA601138) at 1/2,000 dilution was used in 5% NFDM/TBST at room temperature for 2 hours. Goat Anti-Mouse IgG - HRP Secondary Antibody (HA1006) at 1/50,000 dilution was used for 1 hour at room temperature.

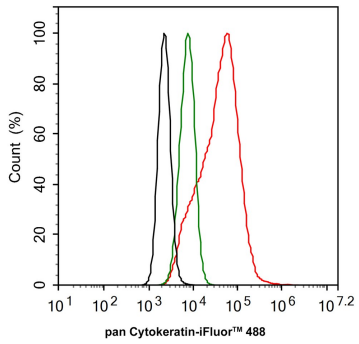

Fig17: Flow cytometric analysis of A431 cells labeling pan Cytokeratin.

Cells were fixed and permeabilized. Then stained with the primary antibody (HA601138, 1µg/mL) (red) compared with Mouse IgG1 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-Mouse IgG Secondary antibody (HA1125) at 1/1,000 dilution for 30 minutes at +4℃. Unlabelled sample was used as a control (cells without incubation with primary antibody; black).

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