

## Product Datasheet

# Cytokeratin 5 Mouse Monoclonal Antibody (orb865591)

<b>Catalog Number</b>	orb865591
<b>Category</b>	Antibodies
<b>Description</b>	Anti-Cytokeratin 5 Antibody (monoclonal, B1J4H0). Tested in Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Human.
<b>Target</b>	Keratin, type II cytoskeletal 5
<b>Clonality</b>	Monoclonal
<b>Species/Host</b>	Mouse
<b>Isotype</b>	Mouse IgG2a
<b>Conjugation</b>	Unconjugated
<b>Reactivity</b>	Human
<b>Form/Appearance</b>	Lyophilized
<b>Concentration</b>	500 µg/ml
<b>Buffer/Preservatives</b>	Each vial contains 4 mg Trehalose, 0.9 mg NaCl and 0.2 mg Na <sub>2</sub> HPO <sub>4</sub> .
<b>Reconstitution</b>	Adding 0.2 ml of distilled water will yield a concentration of 500 µg/ml.
<b>Purification</b>	Immunogen affinity purified.
<b>Immunogen</b>	A synthetic peptide corresponding to a sequence in the middle region of human Cytokeratin 5, different from the related mouse sequence by one amino acid, and identical to the related rat sequence.

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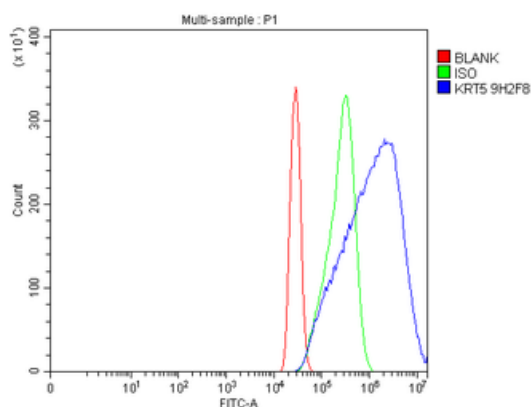
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<b>UniProt ID</b>	<b>P13647</b>
<b>MW</b>	62 kDa
<b>Tested applications</b>	FC, ICC, IF, IHC, WB
<b>Dilution range</b>	Western blot, 0.25-0.5 µg/ml, Human Immunohistochemistry(Paraffin-embedded Section), 2-5 µg/ml, Human Immunocytochemistry/Immunofluorescence, 5 µg/ml, Human Flow Cytometry (Fixed), 1-3 µg/1x <sup>6</sup> cells, Human
<b>Cross Reactivity</b>	No cross-reactivity with other proteins.
<b>Antibody Type</b>	Primary Antibody
<b>Clone Number</b>	B1J4H0
<b>Storage</b>	Maintain refrigerated at 2-8°C for up to 2 weeks. For long term storage store at -20°C in small aliquots to prevent freeze-thaw cycles.
<b>Note</b>	For research use only
<b>Expiration Date</b>	12 months from date of receipt.



Flow Cytometry analysis of A431 cells using anti-Cytokeratin 5 antibody. Overlay histogram showing A431 cells (Blue line). To facilitate intracellular staining, cells were fixed with 4% paraformaldehyde and permeabilized with permeabilization buffer. The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-Cytokeratin 5 Antibody (1 µg/1x10<sup>6</sup> cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (5-10 µg/1x10<sup>6</sup> cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG (1 µg/1x10<sup>6</sup>) used under the same conditions. Unlabelled sample without incubation with primary antibody and secondary antibody (Red line) was used as a blank control.

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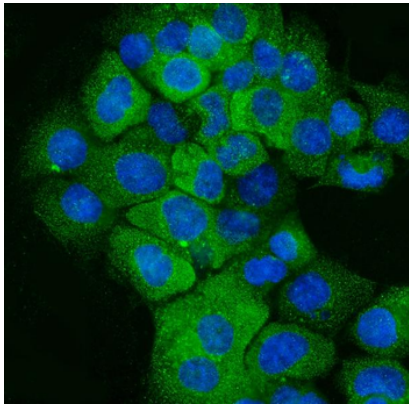
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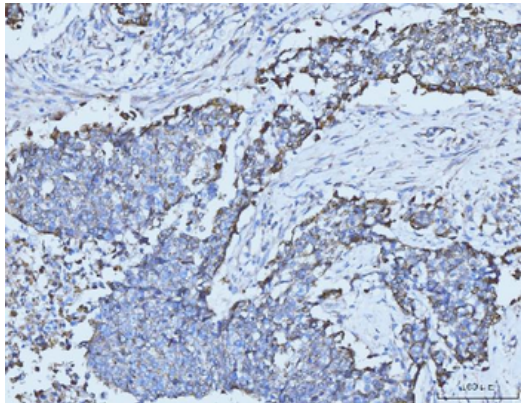
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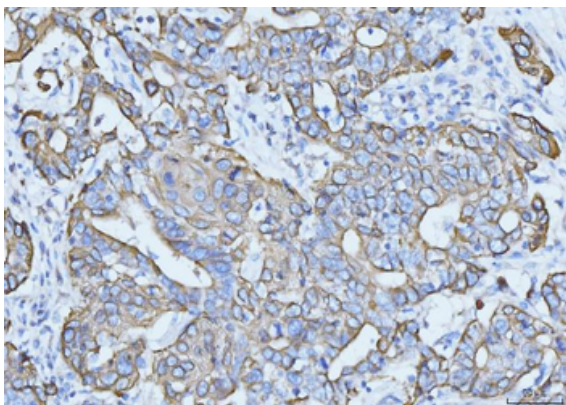
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IF analysis of Cytokeratin 5 using anti-Cytokeratin 5 antibody. Cytokeratin 5 was detected in an immunocytochemical section of A431 cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 5  $\mu\text{g}/\text{mL}$  mouse anti-Cytokeratin 5 Antibody overnight at 4°C. DyLight®488 Conjugated Goat Anti-Mouse IgG was used as secondary antibody at 1:100 dilution and incubated for 30 minutes at 37°C. The section was counterstained with DAPI. Visualize using a fluorescence microscope and filter sets appropriate for the label used.



IHC analysis of Cytokeratin 5 using anti-Cytokeratin 5 antibody. Cytokeratin 5 was detected in a paraffin-embedded section of human lung cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2  $\mu\text{g}/\text{ml}$  mouse anti-Cytokeratin 5 Antibody overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC) with DAB as the chromogen.



IHC analysis of Cytokeratin 5 using anti-Cytokeratin 5 antibody. Cytokeratin 5 was detected in a paraffin-embedded section of human metaplasia of squamous cells of the renal pelvis tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2  $\mu\text{g}/\text{ml}$  mouse anti-Cytokeratin 5 Antibody overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC) with DAB as the chromogen.

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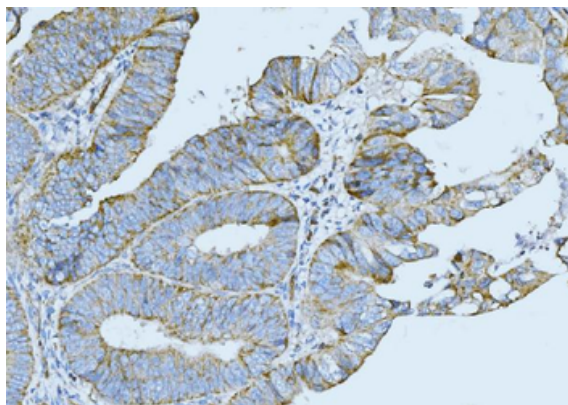
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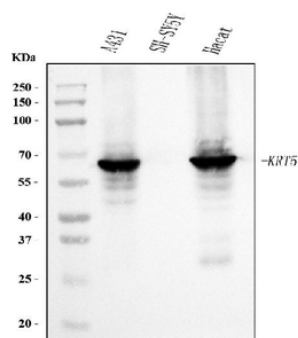
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IHC analysis of Cytokeratin 5 using anti-Cytokeratin 5 antibody. Cytokeratin 5 was detected in a paraffin-embedded section of human rectal moderately differentiated adenocarcinoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-Cytokeratin 5 Antibody overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC) with DAB as the chromogen.



Western blot analysis of Cytokeratin 5 using anti-Cytokeratin 5 antibody. Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 30 µg of sample under reducing conditions. Lane 1: human A431 whole cell lysates, Lane 2: human SH-SY5Y whole cell lysates, Lane 3: human Hacat whole cell lysates. After electrophoresis, proteins were transferred to a nitrocellulose membrane at 150 mA for 50-90 minutes. Blocked the membrane with 5% non-fat milk/TBS for 1.5 hour at RT. The membrane was incubated with mouse anti-Cytokeratin 5 antigen affinity purified monoclonal antibody at 0.5 µg/mL overnight at 4°C, then washed with TBS-0.1% Tween 3 times with 5 minutes each and probed with a goat anti-mouse IgG-HRP secondary antibody at a dilution of 1:10000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit with Tanon 5200 system. A specific band was detected for Cytokeratin 5 at approximately 62 kDa. The expected band size for Cytokeratin 5 is at 62 kDa.

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