Anti-Cytokeratin 20 Antibody [SA35-03] - BSA and Azide free

HA750026

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Rat
Applications:	WB, IF-Cell, IF-Tissue, IHC-P, FC, IP
Molecular Wt:	Predicted band size: 48 kDa
Clone number:	SA35-03

Description: Keratin 20, often abbreviated CK20, is a protein that in humans is encoded by the KRT20 gene. Keratin 20 is a type I cytokeratin. It is a major cellular protein of mature enterocytes and goblet cells and is specifically found in the gastric and intestinal mucosa. In immunohistochemistry, antibodies to CK20 can be used to identify a range of adenocarcinoma arising from epithelia that normally contain the CK20 protein. For example, the protein is commonly found in colorectal cancer, transitional cell carcinomas and in Merkel cell carcinoma, but is absent in lung cancer, prostate cancer, and non-mucinous ovarian cancer. It is often used in combination with antibodies to CK7 to distinguish different types of glandular tumour.

Immunogen: Synthetic peptide within Human Cytokeratin 20 aa 375-424 / 424.

Positive control: HT-29 cell lysate, LoVo cell lysate, CRC cell lysate, CRC, human colon carcinoma tissue, human small intestine tissue, rat small intestine tissue.

Subcellular location: Cytoplasm

Database links: SwissProt: P35900 Human | P25030 Rat

Recommended Dilutions:

WB	1:2,000-1:5,000
IF-Cell	1:50
IF-Tissue	1:200
IHC-P	1:50-1:500
FC	1:50
IP	1-2μg/sample

Storage Buffer: 1*PBS (pH7.4).

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

Purity: Protein A affinity purified.

Images

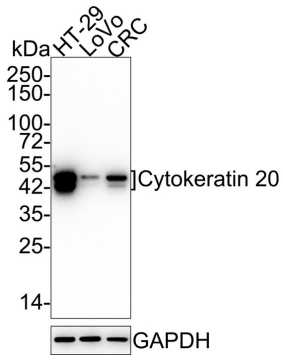

Fig1: Western blot analysis of Cytokeratin 20 on different lysates with Rabbit anti-Cytokeratin 20 antibody (HA750026) at 1/5,000 dilution.

Lane 1: HT-29 cell lysate
Lane 2: LoVo cell lysate
Lane 2: CRC cell lysate

Lysates/proteins at 15 µg/Lane.

Predicted band size: 48 kDa
Observed band size: 48/50 kDa

Exposure time: 1 minute;

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 (HA750026) 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.

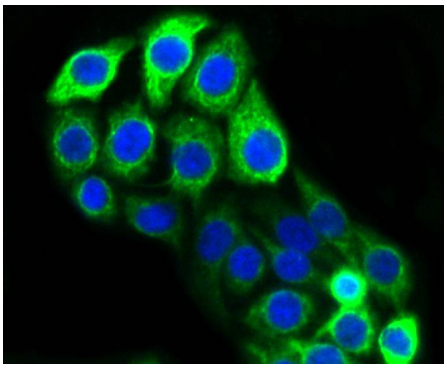

Fig2: ICC staining of Cytokeratin 20 in CRC cells (green). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 1% Blocker BSA for 15 minutes at room temperature. Cells were probed with the primary antibody (HA750026, 1/50) for 1 hour 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).

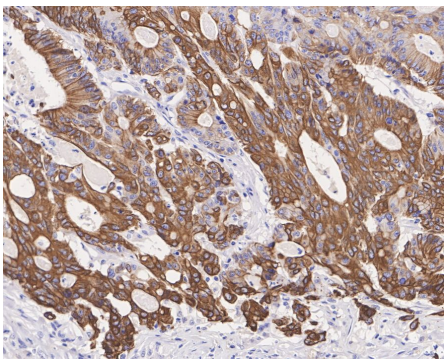

Fig3: Immunohistochemical analysis of paraffin-embedded human colon carcinoma tissue with Rabbit anti-Cytokeratin 20 antibody (HA750026) at 1/500 dilution.

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 (HA750026) at 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.

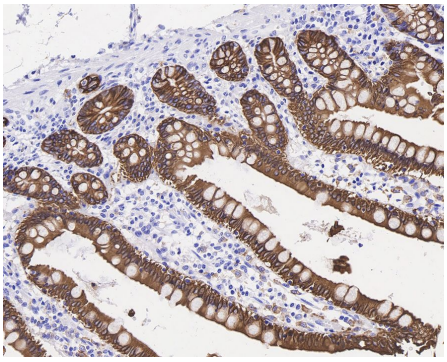

Fig4: Immunohistochemical analysis of paraffin-embedded human small intestine tissue with Rabbit anti-Cytokeratin 20 antibody (HA750026) at 1/200 dilution.

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 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA750026) at 1/200 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.

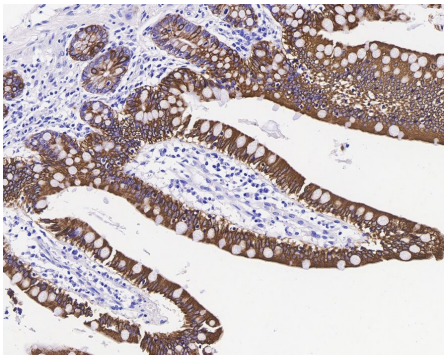

Fig5: Immunohistochemical analysis of paraffin-embedded human small intestine tissue with Rabbit anti-Cytokeratin 20 antibody (HA750026) at 1/200 dilution.

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 (HA750026) at 1/200 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.

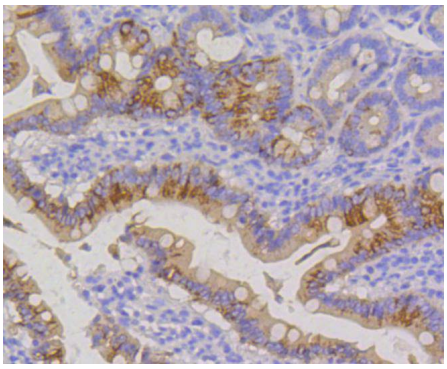

Fig6: Immunohistochemical analysis of paraffin-embedded rat small intestine tissue using anti-Cytokeratin 20 antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes.The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA750026, 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.

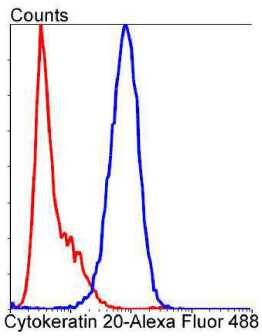

Fig7: Flow cytometric analysis of Cytokeratin 20 was done on CRC cells. The cells were fixed, permeabilized and stained with the primary antibody (HA750026, 1/50) (blue). After incubation of the primary antibody at room temperature for an hour, the cells were stained with a Alexa Fluor 488-conjugated Goat anti-Rabbit IgG Secondary antibody at 1/1,000 dilution for 30 minutes.Unlabelled sample was used as a control (cells without incubation with primary antibody; red).

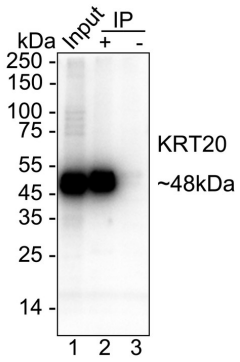

Fig8: Cytokeratin 20 was immunoprecipitated from 0.2 mg HT-29 cell lysate with HA750026 at 2 µg/10 µl beads. Western blot was performed from the immunoprecipitate using HA750026 at 1/1,000 dilution. HRP Conjugated Anti-Rabbit IgG for IP Nano-secondary antibody at 1/5,000 dilution was used for 1 hour at room temperature.

Lane 1: HT-29 cell lysate (input)
Lane 2: HA750026 IP in HT-29 cell lysate
Lane 3: Rabbit IgG instead of HA750026 in HT-29 cell lysate

Blocking/Dilution buffer: 5% NFDM/TBST
Exposure time: 3 seconds; ECL: K1801

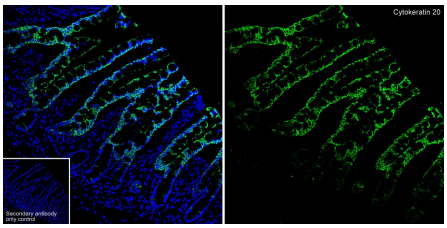

Fig9: Application: IF-Tissue

Species: Human

Site: small intestine

Sample: Paraffin-embedded section

Antibody concentration: 1/200

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

Background References

1. Strand DW et al. Deficiency in metabolic regulators PPAR and PTEN cooperates to drive keratinizing squamous metaplasia in novel models of human tissue regeneration. Am J Pathol 182:449-59 (2013).
2. Volkmer JP et al. Three differentiation states risk-stratify bladder cancer into distinct subtypes. Proc Natl Acad Sci U S A 109:2078-83 (2012).