abcam

Product datasheet

Anti-Cytokeratin 20 antibody [EPR1622Y] - Cytoskeleton Marker ab76126

יעלאעבע RabMAb

★★★★★ 17 Abreviews 46 References 画像数 14

製品の概要

製品名 Anti-Cytokeratin 20 antibody [EPR1622Y] - Cytoskeleton Marker

製品の詳細 Rabbit monoclonal [EPR1622Y] to Cytokeratin 20 - Cytoskeleton Marker

由来種 Rabbit

特異性 The immunogen of this antibody is 73% homolog with Mouse-Cytokeratin 20. This antibody gives

positive results for mouse samples in Western Blot only. Therefore we do not recommend this

antibody for mouse samples and do not cover mouse with our Abpromise guarantee.

アプリケーション 適用あり: Flow Cyt (Intra), ICC/IF, WB, IHC-P

種交差性 交差種: Human

交差が予測される動物種: Rat, Goat, Pig, Common marmoset 4

免疫原 Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

ポジティブ・コントロール WB: HT-29, DLD-1 and Human small intestine cell lysates. IHC-P: Human colon adenocarcinoma

and urinary bladder transitional carcinoma tissue. ICC: HT-29 cells. Flow Cyt (intra): LoVo cells.

特記事項 This product is a recombinant monoclonal antibody, which offers several advantages including:

- High batch-to-batch consistency and reproducibility

- Improved sensitivity and specificity

- Long-term security of supply

- Animal-free production

For more information see here.

Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb**® **patents**.

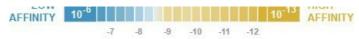
製品の特性

製品の状態 Liquid

保存方法 Shipped at 4°C. Upon delivery aliquot and store at -20°C. Avoid freeze / thaw cycles.

 $K_D = 3.10 \times 10^{-11} M$ 解離定数(KD値)





Learn more about K_D

バッファー pH: 7.20

Preservative: 0.01% Sodium azide

Constituents: 9% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA, 50% Tissue culture

supernatant

精製度 Protein A purified

ポリ/モノ モノクローナル **クローン名** EPR1622Y

アイソタイプ IgG

アプリケーション

The Abpromise guarantee <u>Abpromise保証は、</u>次のテスト済みアプリケーションにおけるab76126の使用に適用されますアプリケーションノートには、推奨の開始希釈率がありますが、適切な希釈率につきましてはご検討ください。

アプリケーション	Abreviews	特記事項
Flow Cyt (Intra)		1/1000. ab172730 - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody.
ICC/IF	★★★★ (3)	1/100 - 1/500.
WB		1/10000 - 1/50000. Predicted molecular weight: 48 kDa.
IHC-P	*** * <u>(12)</u>	1/100 - 1/250. Perform heat mediated antigen retrieval before commencing with IHC staining protocol.

ターゲット情報

機能 Plays a significant role in maintaining keratin filament organization in intestinal epithelia. When

phosphorylated, plays a role in the secretion of mucin in the small intestine.

組織特異性 Expressed predominantly in the intestinal epithelium. Expressed in luminal cells of colonic

mucosa. Also expressed in the Merkel cells of keratinized oral mucosa; specifically at the tips of some rete ridges of the gingival mucosa, in the basal layer of the palatal mucosa and in the taste

buds of lingual mucosa.

配列類似性 Belongs to the intermediate filament family.

発生段階 First detected at embryonic week 8 in individual 'converted' simple epithelial cells of the

developing intestinal mucosa. In later fetal stages, synthesis extends over most goblet cells and a variable number of villus enterocytes. In the developing gastric and intestinal mucosa, expressed in all enterocytes and goblet cells as well as certain 'low-differentiated' columnar cells, whereas

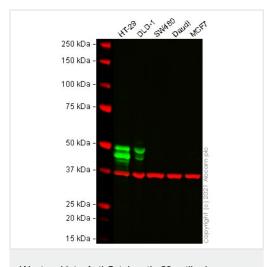
the neuroendocrine and Paneth cells are negative.

翻訳後修飾 Hyperphosphorylation at Ser-13 occurs during the early stages of apoptosis but becomes less

prominent during the later stages. Phosphorylation at Ser-13 also increases in response to stress

brought on by cell injury.

画像



Western blot - Anti-Cytokeratin 20 antibody

[EPR1622Y] - Cytoskeleton Marker (ab76126)

All lanes : Anti-Cytokeratin 20 antibody [EPR1622Y] - Cytoskeleton Marker (ab76126) at 1/10000 dilution

Lane 1: HT-29 cell lysate

Lane 2: DLD-1 cell lysate

Lane 3: SW480 cell lysate

Lane 4 : Daudi cell lysate

Lane 5: MCF7 cell lysate

Lysates/proteins at 20 µg per lane.

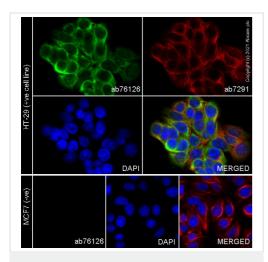
Performed under reducing conditions.

Predicted band size: 48 kDa

Additional bands at: 45 kDa. We are unsure as to the identity of

these extra bands.

False colour image of Western blot: Anti-Cytokeratin 20 antibody [EPR1622Y] - Cytoskeleton Marker staining at 1/10000 dilution, shown in green; Mouse anti-GAPDH antibody [6C5] (ab8245) loading control staining at 1/20000 dilution, shown in red. In Western blot, ab76126 was shown to bind specifically to Cytokeratin 20. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 3 % milk in TBS-0.1 % Tween® 20 (TBS-T) before incubation with primary antibodies overnight at 4°C. Blots were washed four times in TBS-T, incubated with secondary antibodies for 1 h at room temperature, washed again four times then imaged. Secondary antibodies used were Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed (ab216776) at 1/20000 dilution.

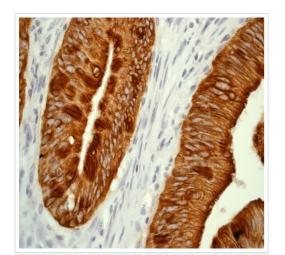


Immunocytochemistry/ Immunofluorescence - Anti-Cytokeratin 20 antibody [EPR1622Y] - Cytoskeleton Marker (ab76126)

ab76126 staining Cytokeratin 20 in HT-29 cells, with negative expression in MCF7 cells. The cells were fixed with 100% methanol (5 min), permeabilised with 0.1% Triton x-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated overnight at +4°C with ab76126 at 0.1 µg/ml and ab7291, Mouse monoclonal [DM1A] to alpha Tubulin at 0.5 µg/ml. Cells were then incubated with ab150081, Goat polyclonal Secondary Antibody to Rabbit lgG - H&L (Alexa Fluor® 488), pre-adsorbed at 1/1000 dilution (shown in green) and ab150119, Goat polyclonal Secondary Antibody to Mouse lgG - H&L (Alexa Fluor® 647), pre-adsorbed at 1/1000 dilution (shown in red). Nuclear DNA was labelled with DAPI (shown in blue).

Image was acquired with a confocal microscope (Leica-Microsystems TCS SP8) and a single confocal section is shown.

This product also work with 4% formaldehyde (10 min) fixation under the same testing conditions.

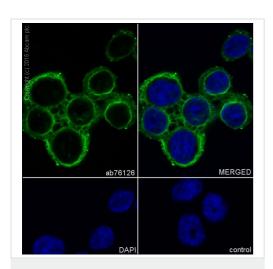


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Cytokeratin 20 antibody

[EPR1622Y] - Cytoskeleton Marker (ab76126)

ab76126 at 1/100 dilution staining Cytokeratin 20 in human colon adenocarcinoma by Immunohistochemistry, Paraffin-embedded tissue.

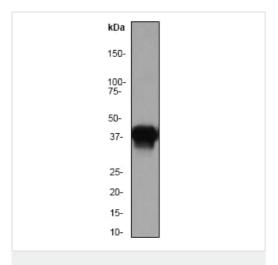
Perform heat mediated antigen retrieval before commencing with IHC staining protocol.



Immunocytochemistry/ Immunofluorescence - Anti-Cytokeratin 20 antibody [EPR1622Y] (ab76126)

Immunocytochemistry/Immunofluorescence analysis of HT-29 (human colorectal adenocarcinoma) cells labelling Cytokeratin 20 with purified ab76126 at 1/500. Cells were fixed with 100% methanol. ab150077, Alexa Fluor[®] 488-conjugated goat anti-rabbit lgG (1/1000) was used as the secondary antibody. Nuclei were counterstained with DAPI (blue).

Secondary Only Control: PBS was used instead of the primary antibody as the negative control.



Western blot - Anti-Cytokeratin 20 antibody [EPR1622Y] (ab76126)

Anti-Cytokeratin 20 antibody [EPR1622Y] - Cytoskeleton Marker (ab76126) at 1/50000 dilution + human small intestine lysate at 10 μg

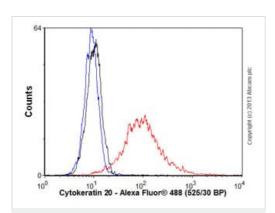
Secondary

goat anti-rabbit-HRP at 1/1000 dilution

Predicted band size: 48 kDa **Observed band size:** 44 kDa

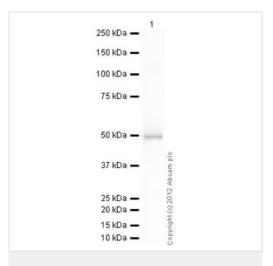
Primary: ab76126, 1/50000 dilution

Sample: human small intestine lysate, 10 ug Secondary: goat anti-rabbit-HRP, 1/1000 dilution



Flow Cytometry (Intracellular) - Anti-Cytokeratin 20 antibody [EPR1622Y] - Cytoskeleton Marker (ab76126)

Overlay histogram showing LoVo (Human colorectal adenocarcinoma cell line) cells stained with ab76126 (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Triton X-100 for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab76126, 1/1000 dilution) for 30 min at 22°C. The secondary antibody used was Alexa Fluor 488 goat anti-rabbit lgG (H&L) (ab150077) at 1/2000 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit lgG (monoclonal) (1µg/1x106 cells) used under the same conditions. Unlabelled sample (blue line) was also used as a control. Acquisition of >5,000 events were collected using a 20mW Argon ion laser (488nm) and 525/30 bandpass filter.



Western blot - Anti-Cytokeratin 20 antibody [EPR1622Y] (ab76126)

Anti-Cytokeratin 20 antibody [EPR1622Y] - Cytoskeleton Marker (ab76126) at 1/50000 dilution + Recombinant Human Cytokeratin 20 protein (ab73640) at 0.01 µg

Secondary

Goat Anti-Rabbit IgG H&L (HRP) preadsorbed (ab97080) at 1/5000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

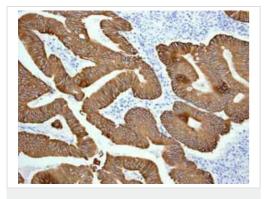
Predicted band size: 48 kDa

Exposure time: 20 seconds

Primary: ab76126, 1/50000 dilution

Sample: <u>ab73640</u>, 0.01 ug

Secondary: ab97080, 1/5000 dilution



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Cytokeratin 20 antibody
[EPR1622Y] - Cytoskeleton Marker (ab76126)

ab76126 showing positive staining in human colonic adenocarcinoma tissue.

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.

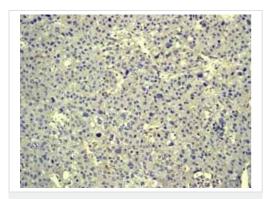


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Cytokeratin 20 antibody

[EPR1622Y] - Cytoskeleton Marker (ab76126)

ab76126 showing positive staining in human urinary bladder transitional carcinoma tissue.

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.

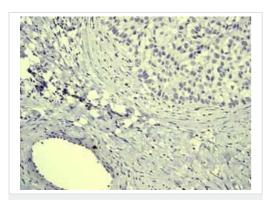


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Cytokeratin 20 antibody

[EPR1622Y] - Cytoskeleton Marker (ab76126)

ab76126 showing **negative staining** in human hepatocellular carcinoma tissue.

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.

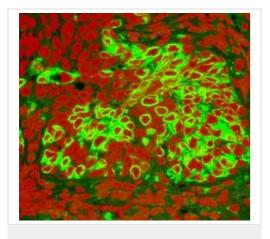


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Cytokeratin 20 antibody

[EPR1622Y] - Cytoskeleton Marker (ab76126)

ab76126 showing **negative staining** in human breast carcinoma tissue.

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Cytokeratin 20 antibody

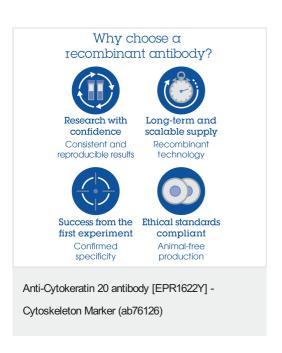
[EPR1622Y] - Cytoskeleton Marker (ab76126)

Fluorescent immunohistochemical analysis of paraffin-embedded human colonic adenocarcinoma tissue using ab76126. Green-CK20 red-PI

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.

OI-RD Scanning - Anti-Cytokeratin 20 antibody [EPR1622Y] - Cytoskeleton Marker (ab76126) Equilibrium disassociation constant (K_D) Learn more about K_D

Click here to learn more about K_D



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