abcam

Product datasheet

Anti-Cytokeratin 8 antibody [M20] - Cytoskeleton Marker ab9023

KO VALIDATED

10 Abreviews 34 References 8 Images

Overview

Product name	Anti-Cytokeratin 8 antibody [M20] - Cytoskeleton Marker
Description	Mouse monoclonal [M20] to Cytokeratin 8 - Cytoskeleton Marker
Host species	Mouse
Tested applications	**Suitable for:** Flow Cyt, ICC, ICC/IF, IHC-P, IHC-Fr, WB
Species reactivity	**Reacts with:** Rat, Rabbit, Human
Immunogen	Full length native protein (purified) corresponding to Human Cytokeratin 8. Keratin isolated from the human breast carcinoma cell line MCF-7.
Positive control	WB: HeLa, A431, PC3 and LNCaP cell lysates. ICC/IF: MCF-7, pancreatic cancer and epithelial ovarian cancer cells. IHC-Fr: Human colon tissue.
General notes	Cytokeratins are a subfamily of intermediate filament proteins and are characterized by a remarkable biochemical diversity, represented in epithelial tissues by at least 20 different polypeptides. They range in molecular weight between 40 kDa and 68 kDa and isoelectric pH between 4.9 – 7.8. The individual cytokeratin polypeptides are numbered 1 to 20. The various epithelia in the human body usually express cytokeratins which are not only characteristic of the type of epithelium, but also related to the degree of maturation or differentiation within an epithelium. Cytokeratin subtype expression patterns are used to an increasing extent in the distinction of different types of epithelial malignancies. The cytokeratin antibodies are not only of assistance in the differential diagnosis of tumors using immunohistochemistry on tissue sections, but are also a useful tool in cytopathology and flow cytometric assays. The Life Science industry has been in the grips of a reproducibility crisis for a number of years. Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets your needs before purchasing. If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be found below, along with publications, customer reviews and Q&As

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C.
Storage buffer	pH: 7.3 Preservative: 0.1% Sodium azide Constituents: PBS, 1% Fetal calf serum
Purity	Tissue culture supernatant
Primary antibody notes	Cytokeratins are a subfamily of intermediate filament proteins and are characterized by a remarkable biochemical diversity, represented in epithelial tissues by at least 20 different polypeptides. They range in molecular weight between 40 kDa and 68 kDa and isoelectric pH between 4.9 – 7.8. The individual cytokeratin polypeptides are numbered 1 to 20. The various epithelia in the human body usually express cytokeratins which are not only characteristic of the type of epithelium, but also related to the degree of maturation or differentiation within an epithelium. Cytokeratin subtype expression patterns are used to an increasing extent in the distinction of different types of epithelial malignancies. The cytokeratin antibodies are not only of assistance in the differential diagnosis of tumors using immunohistochemistry on tissue sections, but are also a useful tool in cytopathology and flow cytometric assays.
Clonality	Monoclonal
Clone number	M20
Isotype	IgG1

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab9023 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

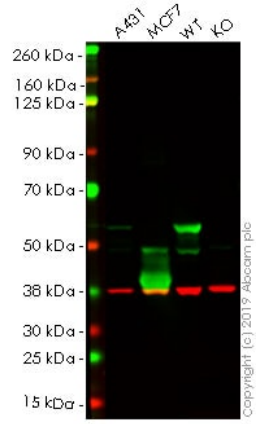
Application	Abreviews	Notes
Flow Cyt		Use 2µg for 10^6 cells. **ab170190** - Mouse monoclonal IgG1, is suitable for use as an isotype control with this antibody.
ICC	★★★★★ (1)	Use at an assay dependent concentration.
ICC/IF	★★★★☆ (4)	Use at an assay dependent concentration.
IHC-P	★★★★☆ (1)	1/5 - 1/25.
IHC-Fr		1/100 - 1/200. For human colon 20min ice cold acetone fixation was carried out with 60min incubation with ab9023 at 37C.
WB	★★★★☆ (4)	1/100 - 1/1000.

Target

Function	Together with KRT19, helps to link the contractile apparatus to dystrophin at the costameres of striated muscle.
Tissue specificity	Observed in muscle fibers accumulating in the costameres of myoplasm at the sarcolemma

	membrane in structures that contain dystrophin and spectrin. Expressed in gingival mucosa and hard palate of the oral cavity.
Involvement in disease	Cirrhosis
Sequence similarities	Belongs to the intermediate filament family.
Post-translational modifications	Phosphorylation on serine residues is enhanced during EGF stimulation and mitosis. Ser-74 phosphorylation plays an important role in keratin filament reorganization. O-glycosylated. O-GlcNAcylation at multiple sites increases solubility, and decreases stability by inducing proteasomal degradation. O-glycosylated (O-GlcNAcylated), in a cell cycle-dependent manner.
Cellular localization	Cytoplasm. Nucleus, nucleoplasm. Nucleus matrix.

Images



Western blot - Anti-Cytokeratin 8 antibody [M20] - Cytoskeleton Marker (ab9023)

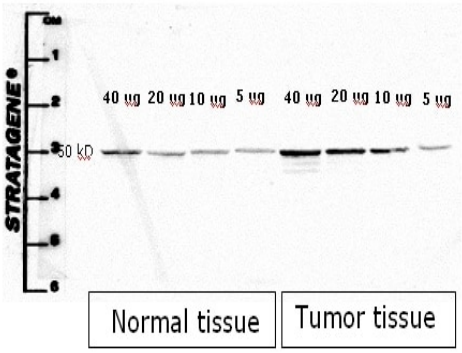
All lanes : Anti-Cytokeratin 8 antibody [M20] - Cytoskeleton Marker (ab9023) at 1/1000 dilution

Lane 1 : A431 cell lysate
Lane 2 : MCF7 cell lysate
Lane 3 : Wild-type HeLa cell lysate
Lane 4 : KRT8 knockout HeLa cell lysate

Lysates/proteins at 20 µg per lane.

Lanes 1 - 4: Merged signal (red and green). Green - ab9023 observed at 55 kDa. Red - loading control, **ab181602** observed at 37 kDa.

ab9023 was shown to react with Cytokeratin 8 in wild-type HeLa cells. Loss of signal was observed when knockout cell line **ab255400** (knockout cell lysate **ab263785**) was used. Wild-type and Cytokeratin 8 knockout samples were subjected to SDS-PAGE. ab9023 and Anti-GAPDH antibody EPR16891] - Loading Control (**ab181602**) were incubated overnight at 4°C at 1 in 1000 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Mouse IgG H&L (IRDye® 800CW) preadsorbed (**ab216772**) and Goat Anti-Rabbit IgG H&L (IRDye® 680RD) preadsorbed (**ab216777**) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



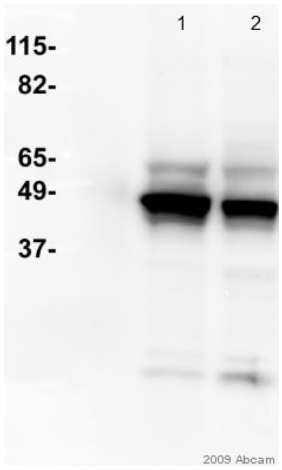
Western blot - Anti-Cytokeratin 8 antibody [M20] - Cytoskeleton Marker (ab9023)

The image shows different concentration of total protein from normal and tumour colon tissues.

This picture was kindly supplied as part of the review submitted by Semona Rupchand.

The image shows different concentration of total protein from normal and tumour colon tissues.

This picture was kindly supplied as part of the review submitted by Semona Rupchand.



Western blot - Anti-Cytokeratin 8 antibody [M20] - Cytoskeleton Marker (ab9023)

This image is a courtesy of Anonymous Abreview

All lanes : Anti-Cytokeratin 8 antibody [M20] - Cytoskeleton Marker (ab9023) at 1/2000 dilution

Lane 1 : Lysate prepared from human prostate cancer LNCaP cells
Lane 2 : Lysate prepared from human prostate cancer PC3 cells

Lysates/proteins at 50 µg per lane.

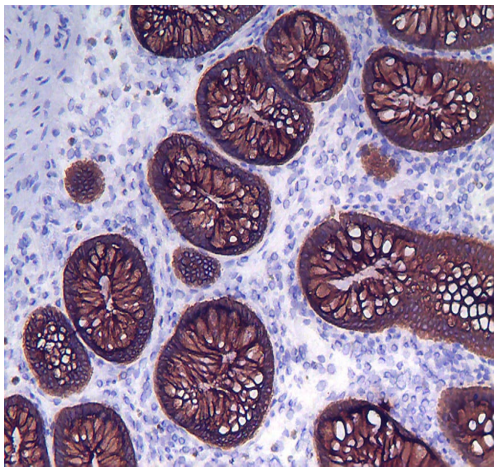
Secondary
All lanes : HRP-conjugated goat polyclonal to mouse IgG at 1/10000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

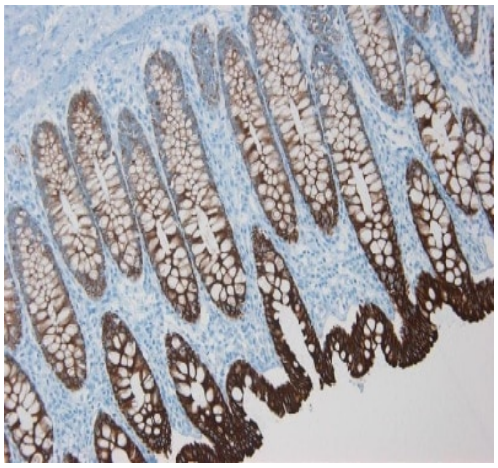
Observed band size: 50 kDa
Additional bands at: 26 kDa, 65 kDa. We are unsure as to the identity of these extra bands.

Exposure time: 4 minutes



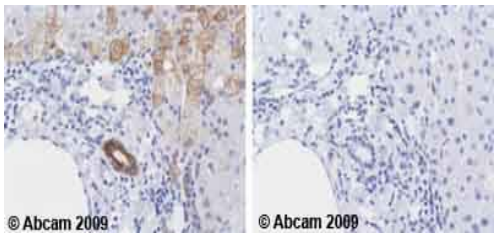
Immunohistochemistry (Frozen sections) - Anti-Cytokeratin 8 antibody [M20] - Cytoskeleton Marker (ab9023)

Immunohistochemistry analysis of frozen section of human colon labeling Cytokeratin 8 with ab9023.



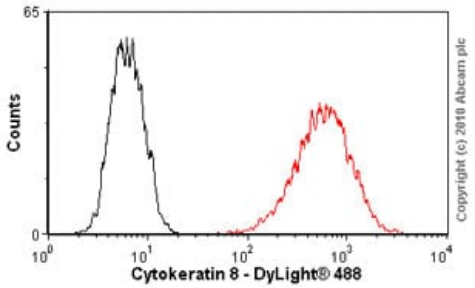
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Cytokeratin 8 antibody [M20] - Cytoskeleton Marker (ab9023)

Immunohistochemistry on paraffin section of human colon



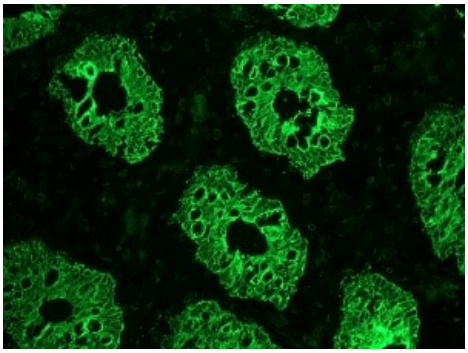
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Cytokeratin 8 antibody [M20] - Cytoskeleton Marker (ab9023)

Ab9023 staining human normal liver parenchima tissue. Staining is localized to cell membrane.
Left panel: with primary antibody at 4 ug/ml. Right panel: isotype control.
Sections were stained using an automated system DAKO Autostainer Plus , at room temperature. Sections were rehydrated and antigen retrieved with the Dako 3-in-1 AR buffer EDTA pH 9.0 in a DAKO PT Link. Slides were peroxidase blocked in 3% H2O2 in methanol for 10 minutes. They were then blocked with Dako Protein block for 10 minutes (containing casein 0.25% in PBS) then incubated with primary antibody for 20 minutes and detected with Dako Envision Flex amplification kit for 30 minutes. Colorimetric detection was completed with Diaminobenzidine for 5 minutes. Slides were counterstained with Haematoxylin and coverslipped under DePeX. Please note that for manual staining we recommend to optimize the primary antibody concentration and incubation time (overnight incubation), and amplification may be required.



Flow Cytometry - Anti-Cytokeratin 8 antibody [M20] - Cytoskeleton Marker (ab9023)

Overlay histogram showing MCF-7 cells stained with ab9023 (red line). The cells were fixed with 4% paraformaldehyde (10 min) and then permeabilized with 0.5% PBS-Tween 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 (ab9023, 2µg/1x10^6 cells) for 30 min at 22ºC. The secondary antibody used was DyLight® 488 goat anti-mouse IgG (H&L) (**ab96879**) at 1/250 dilution for 30 min at 22ºC. Isotype control antibody (black line) was mouse IgG1 [ICIGG1] (**ab91353**, 2µg/1x10^6 cells) used under the same conditions. Acquisition of >5,000 events was performed. This antibody gave a positive signal in MCF-7 cells fixed with methanol (5 min)/permeabilized in 0.5% PBS-Tween used under the same conditions.



Immunohistochemistry (Frozen sections) - Anti-Cytokeratin 8 antibody [M20] - Cytoskeleton Marker (ab9023)

Immunohistochemistry on frozen section of human colon

Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

Our Abpromise to you: Quality guaranteed and expert technical support

- Replacement or refund for products not performing as stated on the datasheet
- Valid for 12 months from date of delivery
- Response to your inquiry within 24 hours
- We provide support in Chinese, English, French, German, Japanese and Spanish
- Extensive multi-media technical resources to help you
- We investigate all quality concerns to ensure our products perform to the highest standards

If the product does not perform as described on this datasheet, we will offer a refund or replacement. For full details of the Abpromise, please visit **https://www.abcam.com/abpromise** or contact our technical team.

Terms and conditions

- Guarantee only valid for products bought direct from Abcam or one of our authorized distributors