# abcam

## Product datasheet

## Anti-Cytokeratin 15 antibody [EPR1614Y] ab52816





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#### Overview

**Product name** Anti-Cytokeratin 15 antibody [EPR1614Y]

**Description** Rabbit monoclonal [EPR1614Y] to Cytokeratin 15

**Host species** Rabbit

**Tested applications** Suitable for: ICC/IF, Flow Cyt (Intra), WB, IHC-P

Unsuitable for: IP

Species reactivity Reacts with: Mouse, Human

**Immunogen** Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

Positive control WB: Mouse skin tissue lysates; A-431 cell lysate. IHC-P: Human squamous cervical carcinoma,

normal skin, cervical carcinoma tissue ICC/IF: HeLa cells. Flow Cyt (intra): A-431 cells.

**General notes** 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**.

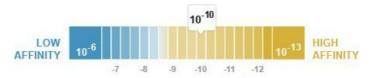
#### **Properties**

**Form** Liquid

Storage instructions Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C.

Stable for 12 months at -20°C.

 $K_D = 5.60 \times 10^{-10} M$ Dissociation constant (K<sub>D</sub>)



Learn more about K<sub>D</sub>

Storage buffer pH: 7.20 Preservative: 0.05% Sodium azide

Constituents: 0.1% BSA, 40% Glycerol (glycerin, glycerine), 9.85% Tris glycine, 50% Tissue

culture supernatant

Purity Protein A purified

Clonality Monoclonal
Clone number EPR1614Y

**Isotype** IgG

#### **Applications**

The Abpromise guarantee Our <u>Abpromise guarantee</u> covers the use of ab52816 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
ICC/IF		1/50 - 1/100.
Flow Cyt (Intra)		1/100.  ab172730 - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody.
WB		1/10000. Detects a band of approximately 45 kDa (predicted molecular weight: 45 kDa).
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

**Application notes** Is unsuitable for IP.

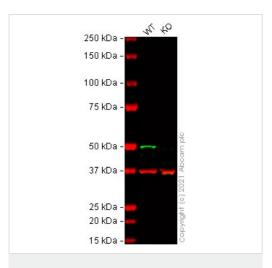
## **Target**

Tissue specificity

Expressed in a discontinuous manner in the basal cell layer of adult skin epidermis, but continuously in the basal layer of fetal skin epidermis and nail. Also expressed in the outer root sheath above the hair bulb in hair follicle (at protein level). Expressed homogeneously in all cell layers of the esophagus and exocervix, but detected in the basal cell layer only of oral mucosa, skin and in the basal plus the next two layers of the suprabasal epithelium of the palate.

**Sequence similarities** Belongs to the intermediate filament family.

## **Images**



Western blot - Anti-Cytokeratin 15 antibody [EPR1614Y] (ab52816)

**All lanes :** Anti-Cytokeratin 15 antibody [EPR1614Y] (ab52816) at 1/10000 dilution

Lane 1: Wild-type A431 cell lysate

Lane 2: KRT15 knockout A431 cell lysate

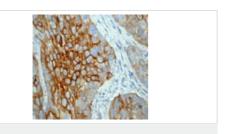
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

**Predicted band size:** 45 kDa **Observed band size:** 50 kDa

**Lanes 1 - 2:** Merged signal (red and green). Green - ab52816 observed at 50 kDa. Red - loading control <u>ab8245</u> (Mouse anti-GAPDH antibody [6C5]) observed at 37 kDa.

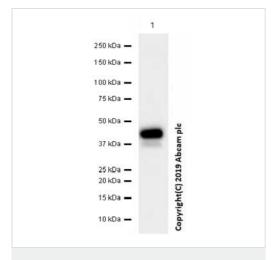
ab52816 was shown to react with Cytokeratin 15 in wild-type A431 cells in Western blot with loss of signal observed in KRT15 knockout cell line <a href="mailto:ab263922">ab263922</a> (knockout cell lysate <a href="mailto:ab262484">ab262484</a>). Wild-type A431 and KRT15 knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3 % milk in TBS-T (0.1 % Tween<sup>®</sup>) before incubation with ab52816 and <a href="mailto:ab8245">ab8245</a> (Mouse anti-GAPDH antibody [6C5]) overnight at 4 °C at a 1 in 10000 dilution and a 1 in 20000 dilution respectively. Blots were incubated with Goat anti-Rabbit lgG H&L (IRDye<sup>®</sup> 800CW) preabsorbed (<a href="mailto:ab216773">ab216773</a>) and Goat anti-Mouse lgG H&L (IRDye<sup>®</sup> 680RD) preabsorbed (<a href="mailto:ab216776">ab216776</a>) secondary antibodies at 1 in 20000 dilution for 1 h at room temperature before imaging.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Cytokeratin 15 antibody [EPR1614Y] (ab52816)

ab52816 at 1/100 dilution staining Cytokeratin 15 in human squamous cervical carcinoma by Immunohistochemistry, Paraffin embedded tissue.

Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



Western blot - Anti-Cytokeratin 15 antibody [EPR1614Y] (ab52816)

Anti-Cytokeratin 15 antibody [EPR1614Y] (ab52816) at 1/100000 dilution + Mouse skin tissue lysates at 20 µg

#### **Secondary**

Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/20000 dilution (Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated)

**Predicted band size:** 45 kDa **Observed band size:** 45 kDa

Blocking buffer and concentration: 5% NFDM/TBST Diluting buffer and concentration: 5% NFDM /TBST

Exposure time: 5.5 seconds

kDa
250150150503725201510-

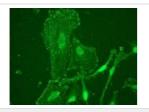
Western blot - Anti-Cytokeratin 15 antibody [EPR1614Y] (ab52816)

Anti-Cytokeratin 15 antibody [EPR1614Y] (ab52816) at 1/10000 dilution + fetal thymus lysate at 10  $\mu g$ 

#### Secondary

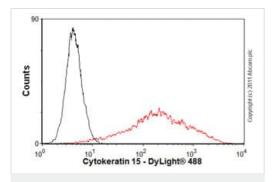
goat anti-rabbit HRP at 1/2000 dilution

**Predicted band size:** 45 kDa **Observed band size:** 45 kDa



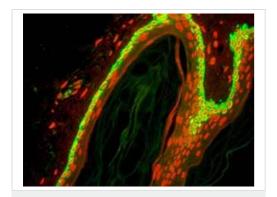
Immunocytochemistry/ Immunofluorescence - Anti-Cytokeratin 15 antibody [EPR1614Y] (ab52816)

ab52816 at 1/50 dilution staining Cytokeratin 15 in HeLa cells by Immunocytochemistry.



Flow Cytometry (Intracellular) - Anti-Cytokeratin 15 antibody [EPR1614Y] (ab52816)

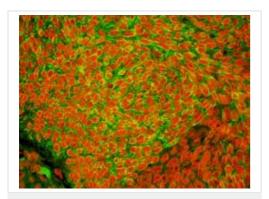
Overlay histogram showing A431 cells stained with ab52816 (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Triton for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab52816, 1/100 dilution) for 30 min at 22°C. The secondary antibody used was DyLight® 488 goat anti-rabbit lgG (H+L) (ab96899) at 1/500 dilution for 30 min at 22°C. Isotype control antibody (black line) was was rabbit lgG (monoclonal) (1µg/1x10<sup>6</sup> cells) used under the same conditions. Acquisition of >5,000 events was performed. This antibody gave a positive signal in A431 cells fixed with 4% paraformaldehyde/permeabilized in 0.1% PBS-Triton used under the same conditions.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Cytokeratin 15 antibody [EPR1614Y] (ab52816)

Fluorescent immunohistochemical analysis of paraffin-embedded human normal skin tissue using ab52816. Green-CK15 red-PI

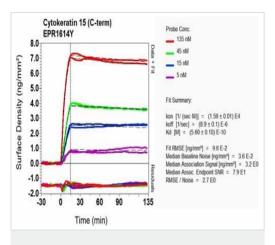
Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Cytokeratin 15 antibody [EPR1614Y] (ab52816)

Fluorescent immunohistochemical analysis of paraffin-embedded human cervical carcinoma tissue using ab52816. Green-CK15 red-Pl.

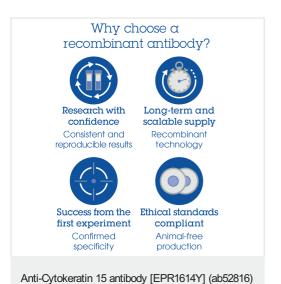
Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



OI-RD Scanning - Anti-Cytokeratin 15 antibody [EPR1614Y] (ab52816)

Equilibrium disassociation constant ( $K_D$ ) Learn more about  $K_D$ 

Click here to learn more about KD



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