abcam

Product datasheet

Anti-Cytokeratin 14 antibody [EP1612Y] - BSA and Azide free ab243907

KO VALIDATED Recombinant RabMAb

7 Images

Overview

Product name	Anti-Cytokeratin 14 antibody [EP1612Y] - BSA and Azide free
Description	Rabbit monoclonal [EP1612Y] to Cytokeratin 14 - BSA and Azide free
Host species	Rabbit
Tested applications	**Suitable for:** IP, WB, IHC-P, Flow Cyt (Intra), ICC/IF
Species reactivity	**Reacts with:** Human
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: A431 cell lysate. ICC/IF: A431 cells. Flow Cyt (intra): A431 cells. IHC-P: human squamous lung carcinoma IP: A431 whole cell lysate.
General notes	ab243907 is the carrier-free version of **ab51054**. Our **carrier-free** antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for increased conjugation efficiency. This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cell-based assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications. Use our **conjugation kits** for antibody conjugates that are ready-to-use in as little as 20 minutes with <1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold. This product is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar® is a trademark of Fluidigm Canada Inc. 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**.

Mouse, Rat: We have preliminary internal testing data to indicate this antibody may not react with these species. Please contact us for more information.

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C. Do Not Freeze.
Storage buffer	pH: 7.2 Constituent: PBS
Carrier free	Yes
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EP1612Y
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab243907 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
IP		Use at an assay dependent concentration.
WB		Use at an assay dependent concentration. Detects a band of approximately 48 kDa (predicted molecular weight: 52 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.
Flow Cyt (Intra)		Use at an assay dependent concentration.
ICC/IF		1/100.

Target

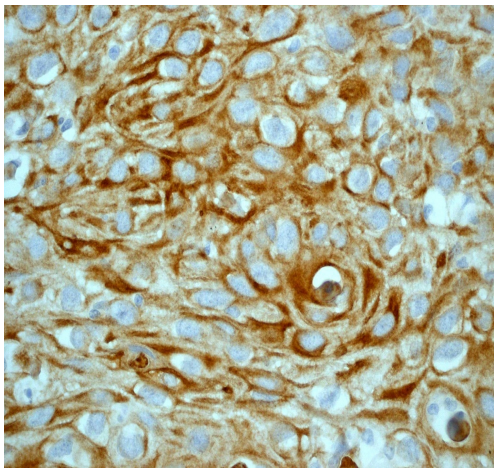
Function	The nonhelical tail domain is involved in promoting KRT5-KRT14 filaments to self-organize into large bundles and enhances the mechanical properties involved in resilience of keratin intermediate filaments in vitro.
Tissue specificity	Detected in the basal layer, lowered within the more apically located layers specifically in the stratum spinosum, stratum granulosum but is not detected in stratum corneum. Strongly expressed in the outer root sheath of anagen follicles but not in the germinative matrix, inner root sheath or hair. Found in keratinocytes surrounding the club hair during telogen.
Involvement in disease	Defects in KRT14 are a cause of epidermolysis bullosa simplex Dowling-Meara type (DM-EBS)

[MIM:131760]. DM-EBS is a severe form of intraepidermal epidermolysis bullosa characterized by generalized herpetiform blistering, milia formation, dystrophic nails, and mucous membrane involvement.
Defects in KRT14 are a cause of epidermolysis bullosa simplex Weber-Cockayne type (WC-EBS) [MIM:131800]. WC-EBS is a form of intraepidermal epidermolysis bullosa characterized by blistering limited to palmar and plantar areas of the skin.
Defects in KRT14 are a cause of epidermolysis bullosa simplex Koebner type (K-EBS) [MIM:131900]. K-EBS is a form of intraepidermal epidermolysis bullosa characterized by generalized skin blistering. The phenotype is not fundamentally distinct from the Dowling-Meara type, although it is less severe.
Defects in KRT14 are the cause of epidermolysis bullosa simplex autosomal recessive (AREBS) [MIM:601001]. AREBS is an intraepidermal epidermolysis bullosa characterized by localized blistering on the dorsal, lateral and plantar surfaces of the feet.
Defects in KRT14 are the cause of Naegeli-Franceschetti-Jadassohn syndrome (NFJS) [MIM:161000]; also known as Naegeli syndrome. NFJS is a rare autosomal dominant form of ectodermal dysplasia. The cardinal features are absence of dermatoglyphics (fingerprints), reticular cutaneous hyperpigmentation (starting at about the age of 2 years without a preceding inflammatory stage), palmoplantar keratoderma, hypohidrosis with diminished sweat gland function and discomfort provoked by heat, nail dystrophy, and tooth enamel defects.
Defects in KRT14 are the cause of dermatopathia pigmentosa reticularis (DPR) [MIM:125595]. DPR is a rare ectodermal dysplasia characterized by lifelong persistent reticulate hyperpigmentation, noncicatricial alopecia, and nail dystrophy.

Sequence similarities Belongs to the intermediate filament family.

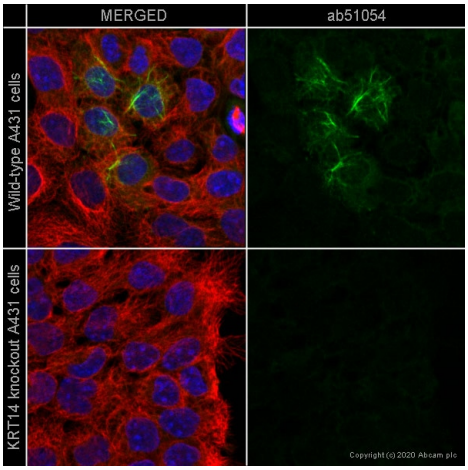
Cellular localization Cytoplasm. Nucleus. Expressed in both as a filamentous pattern.

Images



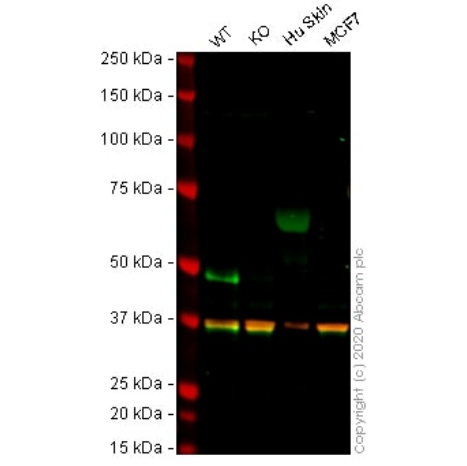
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Cytokeratin 14 antibody [EP1612Y] - BSA and Azide free (ab243907)

This data was developed using the same antibody clone in a different buffer formulation (**ab51054**).
Immunohistochemical analysis of paraffin-embedded human squamous lung carcinoma tissue sections labeling Cytokeratin 14 with purified **ab51054** at 1/100 dilution. ImmunoHistoProbe one step HRP Polymer (ready to use) was used as the secondary antibody. Sections were counterstained with Hematoxylin.
Antigen retrieval was heat mediated antigen retrieval using citrate buffer, pH 6.0).



Immunocytochemistry/ Immunofluorescence - Anti-Cytokeratin 14 antibody [EP1612Y] - BSA and Azide free (ab243907)

This data was developed using the same antibody clone in a different buffer formulation (**ab51054**). **ab51054** staining KRT14 in wild-type A431 cells (top panel) and KRT14 knockout A431 cells (bottom panel). The cells were fixed with 100% methanol (5 min) then permeabilized 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 with **ab51054** at 1/100 dilution and **ab7291** (Mouse monoclonal to alpha Tubulin) at 1/1000 dilution overnight at 4°C followed by a further incubation at room temperature for 1h with a goat secondary antibody to rabbit IgG (Alexa Fluor® 488) (**ab150081**) at 2 µg/ml (shown in green) and a goat secondary antibody to mouse IgG (Alexa Fluor® 594) (**ab150120**) at 2 µg/ml (shown in red). Nuclear DNA was labelled in blue with DAPI.
Image was taken with a confocal microscope (Leica-Microsystems TCS SP8).



Western blot - Anti-Cytokeratin 14 antibody [EP1612Y] - BSA and Azide free (ab243907)

All lanes : Anti-Cytokeratin 14 antibody [EP1612Y] (**ab51054**) at 1/10000 dilution

Lane 1 : Wild-type A431 cell lysate
Lane 2 : KRT14 knockout A431 cell lysate
Lane 3 : Human skin cell lysate
Lane 4 : MCF7 (Human breast adenocarcinoma cell line) whole cell lysate

Lysates/proteins at 20 µg per lane.

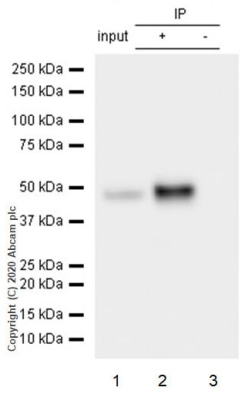
Performed under reducing conditions.

Predicted band size: 52 kDa
Observed band size: 49 kDa

This data was developed using the same antibody clone in a different buffer formulation (**ab51054**).

Lanes 1 - 4: Merged signal (red and green). Green - **ab51054** observed at 49 kDa. Red - loading control, **ab8245** (Mouse anti-GAPDH antibody [6C5]) observed at 37kDa.

ab51054 was shown to react with Cytokeratin 14 in wild-type A431 cells in Western blot. Loss of signal was observed when KRT14 knockout sample was used. Wild-type A431 and KRT14 knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3% milk in TBS-T (0.1% Tween®) before incubation with **ab51054** and **ab8245** (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 IgG H&L (IRDye® 800CW) preabsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Immunoprecipitation - Anti-Cytokeratin 14 antibody [EP1612Y] - BSA and Azide free (ab243907)

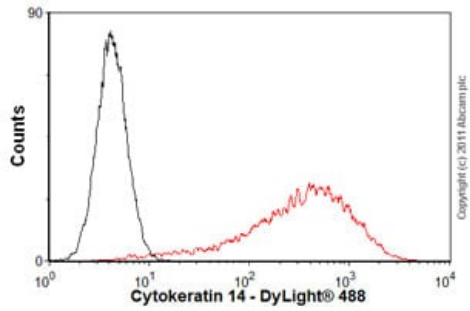
This data was developed using **ab51054**, the same antibody clone in a different buffer formulation.
Purified **ab51054** at 1/20 dilution (0.5µg) immunoprecipitating Cytokeratin 14 in A431 whole cell lysate.
Lane 1 (input): A431 (Human epidermoid carcinoma epithelial cell) whole cell lysate 10µg
Lane 2 (+): **ab51054** + A431 whole cell lysate.
Lane 3 (-): Rabbit monoclonal IgG (**ab172730**) instead of **ab51054** in A431 whole cell lysate.
VeriBlot for IP Detection Reagent (HRP) (**ab131366**) (1/1000 dilution) was used for Western blotting.
Blocking Buffer and concentration: 5% NFDM/TBST.
Diluting buffer and concentration: 5% NFDM/TBST.
Observed band size: 48 kDa

Immunocytochemistry/ Immunofluorescence - Anti-Cytokeratin 14 antibody [EP1612Y] - BSA and Azide free (ab243907)

ICC/IF image of **ab51504** stained A431 (Human epidermoid carcinoma cell line) cells.

The cells were fixed in 100% methanol (5 min) and then incubated in 1% BSA / 10% normal goat serum / 0.3M glycine in 0.1% PBS-Tween for 1h to permeabilize the cells and block non-specific protein-protein interactions. The cells were then incubated with the antibody (**ab51504**, 1/100 dilution) overnight at +4°C. The secondary antibody (green) was **ab96899**, DyLight® 488 goat anti-rabbit IgG (H+L) used at a 1/250 dilution for 1h. Alexa Fluor® 594 WGA was used to label plasma membranes (red) at a 1/200 dilution for 1h. DAPI was used to stain the cell nuclei (blue) at a concentration of 1.43 µM.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab51054**)



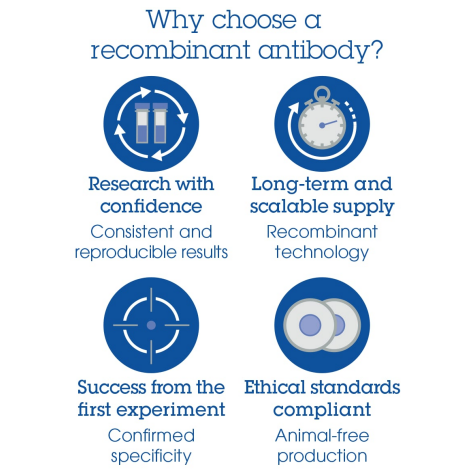
Flow Cytometry (Intracellular) - Anti-Cytokeratin 14 antibody [EP1612Y] - BSA and Azide free (ab243907)

Overlay histogram showing A431 (Human epidermoid carcinoma cell line) cells stained with **ab51054** (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 (**ab51054**, 1/100 dilution) for 30 min at 22°C. The secondary antibody used was DyLight®488 goat anti-rabbit IgG (H+L) (**ab96899**) at 1/500 dilution for 30 min at 22°C. Isotype control antibody (black line) was was rabbit IgG (monoclonal) (1µg/1x10^6cells) 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.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab51054**).



Anti-Cytokeratin 14 antibody [EP1612Y] - BSA and Azide free (ab243907)

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