Anti-Cytokeratin 14 Antibody [A2C10]

EM1901-33

Product Type:	Mouse monoclonal IgG1, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IHC-P, IF-Cell, IF-Tissue
Molecular Wt:	Predicted band size: 52 kDa
Clone number:	A2C10

Description: This gene encodes a member of the keratin family, the most diverse group of intermediate filaments. This gene product, a type I keratin, is usually found as a heterotetramer with two keratin 5 molecules, a type II keratin. Together they form the cytoskeleton of epithelial cells. Mutations in the genes for these keratins are associated with epidermolysis bullosa simplex. 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. Expressed in the corneal epithelium (at protein level). 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. A form of epidermolysis bullosa simplex, a group of skin fragility disorders characterized by skin blistering due to cleavage within the basal layer of keratinocytes, and erosions caused by minor mechanical trauma. There is a broad spectrum of clinical severity ranging from minor blistering on the feet, to subtypes with extracutaneous involvement and a lethal outcome. EBS1A is an autosomal dominant form characterized by generalized intraepidermal skin blistering that begins and is very prominent at birth. EBS1A may be life-threatening in the first year of life. Tendency to blistering diminishes in adolescence.

Immunogen: Synthetic peptide within Human Cytokeratin 14 aa 423-472 / 472.

Positive control: A431 cell lysate, mouse skin tissue lysate, rat skin tissue lysate, A431, human breast tissue, human esophagus tissue, human lung squamous carcinoma tissue, human tonsil tissue, human skin tissue, mouse esophagus tissue.

Subcellular location: Cytoplasm, Nucleus.

Database links: SwissProt: P02533 Human

Recommended Dilutions:

WB	1:500-1:2,000
IHC-P	1:200-1:1,000
IF-Cell	1:100
IF-Tissue	1:200

Storage Buffer: 1*PBS (pH7.4), 0.2% BSA, 50% Glycerol. Preservative: 0.05% Sodium Azide.

Storage Instruction: Shipped at 4℃. Store at +4℃ short term (1-2 weeks). It is recommended to aliquot into single-use upon delivery. Store at -20℃ long term.

Purity: Protein G affinity purified.

Images

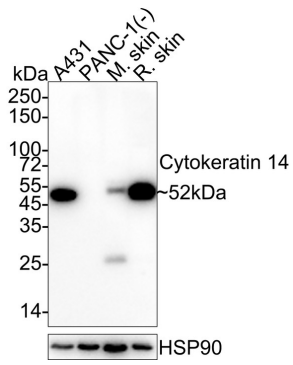

Fig1: Western blot analysis of Cytokeratin 14 on different lysates with Mouse anti-Cytokeratin 14 antibody (EM1901-33) at 1/1,000 dilution.

Lane 1: A431 cell lysate (20 µg/Lane)
Lane 2: PANC-1 cell lysate (negative) (20 µg/Lane)
Lane 3: Mouse skin tissue lysate (40 µg/Lane)
Lane 4: Rat skin tissue lysate (40 µg/Lane)

Predicted band size: 52 kDa
Observed band size: 52 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 (EM1901-33) at 1/1,000 dilution was used in 5% NFDM/TBST at 4℃ overnight. Goat Anti-Mouse IgG - HRP Secondary Antibody (HA1006) at 1/50,000 dilution was used for 1 hour at room temperature.

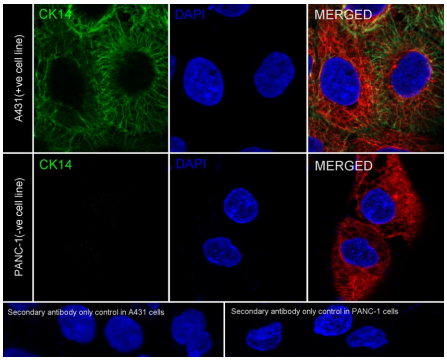

Fig2: Immunocytochemistry analysis of A431 (positive) and PANC-1 (negative) labeling Cytokeratin 14 with Mouse anti-Cytokeratin 14 antibody (EM1901-33) at 1/100 dilution.

Cells were fixed in 4% paraformaldehyde for 20 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 5 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Mouse anti-Cytokeratin 14 antibody (EM1901-33) at 1/100 dilution in 1% BSA in PBST overnight at 4 ℃. Goat Anti-Mouse IgG H&L (iFluor™ 488, HA1125) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

beta Tubulin (ET1602-4, red) was stained at 1/100 dilution overnight at +4℃. Goat Anti-Rabbit IgG H&L (iFluor™ 594, HA1122) were used as the secondary antibody at 1/1,000 dilution.

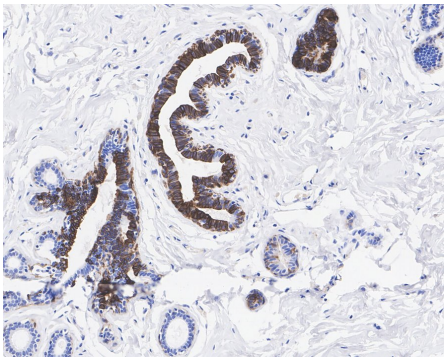

Fig3: Immunohistochemical analysis of paraffin-embedded human breast tissue with Mouse anti-Cytokeratin 14 antibody (EM1901-33) at 1/1,000 dilution.

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 (EM1901-33) at 1/1,000 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.

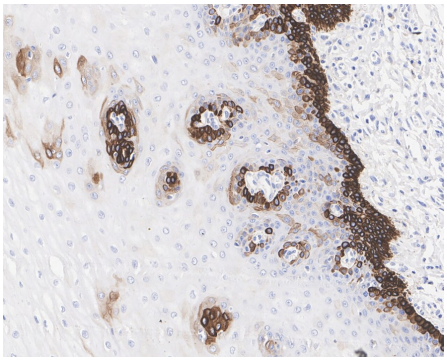

Fig4: Immunohistochemical analysis of paraffin-embedded human esophagus tissue with Mouse anti-Cytokeratin 14 antibody (EM1901-33) at 1/1,000 dilution.

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 (EM1901-33) at 1/1,000 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.

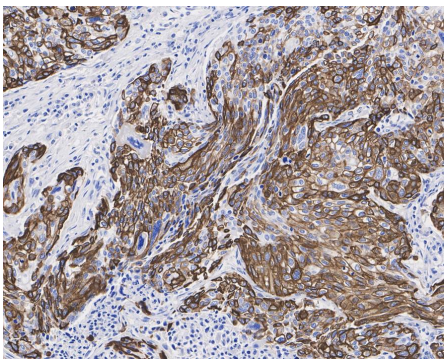

Fig5: Immunohistochemical analysis of paraffin-embedded human lung squamous carcinoma tissue with Mouse anti-Cytokeratin 14 antibody (EM1901-33) at 1/1,000 dilution.

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 (EM1901-33) at 1/1,000 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.

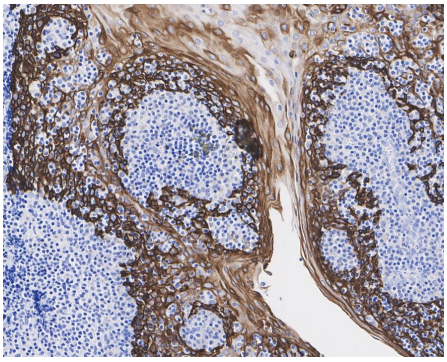

Fig6: Immunohistochemical analysis of paraffin-embedded human tonsil tissue with Mouse anti-Cytokeratin 14 antibody (EM1901-33) at 1/1,000 dilution.

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 (EM1901-33) at 1/1,000 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.

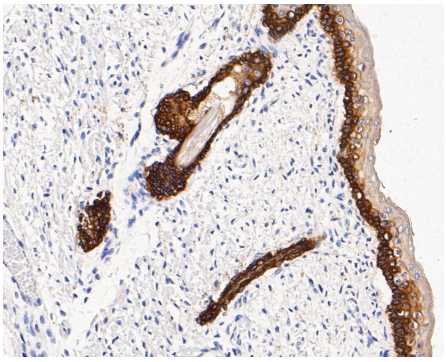

Fig7: Immunohistochemical analysis of paraffin-embedded human skin tissue using anti-Cytokeratin 14 antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes.The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (EM1901-33, 1/200) 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.

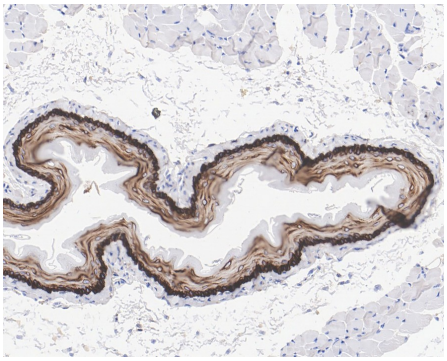

Fig8: Immunohistochemical analysis of paraffin-embedded mouse esophagus tissue with Mouse anti-Cytokeratin 14 antibody (EM1901-33) at 1/1,000 dilution.

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 (EM1901-33) at 1/1,000 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.

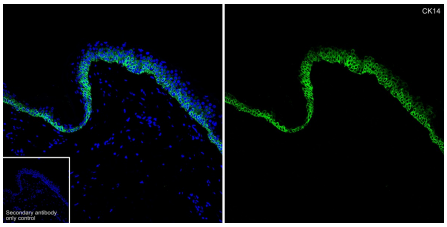

Fig9: Application: IF-Tissue

Species: Human

Site: skin

Sample: Paraffin-embedded section

Antibody concentration: 1/200

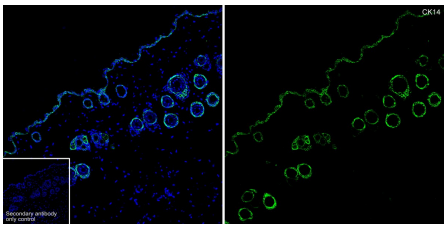

Fig10: Application: IF-Tissue

Species: Mouse

Site: skin

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. Bousquet O. et al. The nonhelical tail domain of keratin 14 promotes filament bundling and enhances the mechanical properties of keratin intermediate filaments in vitro. J. Cell Biol. 155:747-754(2001).
2. Schweizer J et al. "New consensus nomenclature for mammalian keratins". The Journal of Cell Biology. 174 (2): 169–74(2006).