

MONOCLONAL ANTIBODY

Anti-Desmoglein 3 (Mouse) mAb (No Azide)

Code No.	Clone	Subclass	Quantity	Concentration
D218-3	AK18	Mouse IgG1	100 µL	1 mg/mL

BACKGROUND: Pemphigus vulgaris (PV) is a life-threatening autoimmune blistering disease of the skin and mucous membranes. In patients of PV, IgG autoantibodies against the cell surface of keratinocytes in stratified squamous epithelia play a major pathogenic role in loss of cell-cell adhesion. The autoimmune target of PV is desmoglein 3 (Dsg3) which is a member of the cadherin family of calcium-dependending, transmembrane glycoprotein.

SOURCE: This antibody was purified from hybridoma (clone AK18) supernatant using protein A agarose. This hybridoma was established by fusion of splenocytes from PV model mouse and P3 mouse myeloma cells.

FORMULATION: 100 µg IgG in 100 µL volume of PBS, pH 7.2. No preservative is contained.

STORAGE: This antibody solution is stable for one year from the date of purchase when stored at 4°C.

REACTIVITY: This antibody reacts with human and mouse Dsg3.

APPLICATIONS:

Western blotting; Not tested

Immunoprecipitation; Clone AK18 is used in reference 6).

Immunohistochemistry; Mouse Dsg3: 0.1 µg/mL

Immunocytochemistry; Mouse Dsg3: 0.1 µg/mL

Detailed procedure is provided in the following **PROTOCOLS**.

INTENDED USE:

For Research Use Only. Not for use in diagnostic procedures.

REFERENCES:

- 1) Culton, D. A., *et al.*, *J. Invest. Dermatol.* **135**, 1590-1597 (2015) [WB, IHC]
- 2) Kamiya, K., *et al.*, *Br. J. Dermatol.* **167**, 252-261 (2012)
- 3) Sharma, P. M., *et al.*, *J. Invest. Dermatol.* **129**, 2309-2312 (2009)
- 4) Yamamoto, Y., *et al.*, *J. Biol. Chem.* **282**, 17866-17876 (2007)
- 5) Yamamoto, Y., *et al.*, *J. Dermatol. Sci.* **47**, 119-125 (2007)
- 6) Tsunoda, K., *et al.*, *J. Immunol.* **170**, 2170-2178 (2003)

Clone AK18 is used in this references.

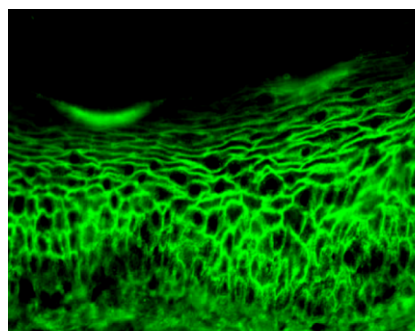
SPECIES CROSS REACTIVITY:

Species	Human	Mouse	Rat
Tissues	Skin	Oral mucosa	Not tested
Reactivity on IHC	weak*	+	

*Clone AK 18 has weak reactivity in human.

RELATED PRODUCTS:

- D217-3 Anti-Desmoglein 3 (Mouse) mAb (AK9)
- D219-3 Anti-Desmoglein 3 (Mouse) mAb (AK23)
- W331-3 Anti-Desmoglein 2 (Human) mAb (2B2C1A)



Immunohistochemical detection of Dsg3 on frozen section of mouse oral mucosa with D218-3.

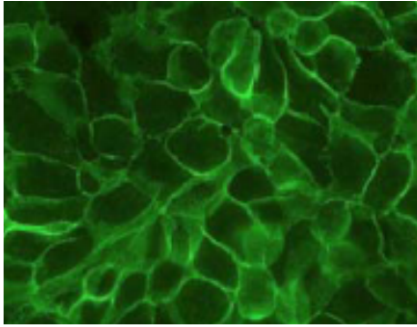
This data was kindly provided by Dr. Amagai, M.D. Ph.D. and Dr. Tsunoda Ph.D. (The Department of Dermatology, School of Medicine, Keio University, Tokyo)

PROTOCOLS:

Immunohistochemical staining for frozen sections

- 1) Cover tissues with the primary antibody diluted with dilution buffer (TBS containing 0.5 mM CaCl₂, 1% BSA) as suggest in the **APPLICATIONS**.
- 2) Incubate the sections for 1 hour at room temperature.
- 3) Wash the slides with washing buffer (TBS containing 0.5 mM CaCl₂) (5 minutes x 3 times).
- 4) Wipe gently around each section and cover tissues with FITC-conjugated anti-mouse IgG antibody diluted by dilution buffer.
- 5) Incubate the sections for 1 hour at room temperature.
- 6) Wash the slides with washing buffer (5 minutes x 3 times).
- 7) Now ready for mounting.

(Positive control for Immunohistochemistry; Mouse oral mucosa)



Immunocytochemical detection of Dsg3 on living mouse keratinocyte cell line (PAM212 cells) with D218-3.

This data was kindly provided by Dr. Amagai, M.D. Ph.D. and Dr. Tsunoda Ph.D. (The Department of Dermatology, School of Medicine, Keio University, Tokyo)

Immunocytochemistry for living keratinocytes

- 1) Prepare 80-90% confluent mouse keratinocytes in chamber slide.
- 2) Wash the cells 2 times with DMEM (FCS free).
- 3) Add the primary antibody diluted with medium as suggest in the **APPLICATIONS** onto the cells and incubate for 30 minutes on ice (Optimization of antibody concentration or incubation condition is recommended if necessary).
- 4) Aspirate medium by aspirator.
- 5) Wash the cells with PBS (5 minutes x 3 times).
- 6) Apply Methanol into each well.
- 7) Incubate for 20 minutes at -30°C.
- 8) Aspirate Methanol by aspirator.
- 9) Wash the cells with PBS (5 minutes x 3 times).
- 10) Add FITC-conjugated anti-mouse IgG antibody diluted with PBS for 30-60 minutes at room temperature. Keep out light by aluminum foil.
- 11) Wash the cells with PBS (5 minutes x 3 times).
- 12) Promptly add mounting medium onto the slide, then put a cover slip on it.

(Positive control for Immunocytochemistry; PAM212)