

Monoclonal Antibody to Podoplanin - Aff - Purified

Alternate names:	Aggrus, GP36, Glycoprotein 36, PA.2.26 antigen, PDPN, PSEC0003, PSEC0025, T1-alpha
Catalog No.:	DM3500P
Quantity:	0.1 mg
Background:	Podoplanin (PDPN) is a small mucin-like transmembrane protein, widely expressed in various specialized cell types throughout the body. This is a type-I integral membrane glycoprotein with diverse distribution in human tissues. The physiological function of this protein may be related to its mucin-type character.
Uniprot ID:	Q86YL7
NCBI:	NP_001006625.1
GeneID:	10630
Host / Isotype:	Mouse / IgG1
Clone:	18H5
Immunogen:	GP36 (Podoplanin) expressed by MDCK cells.
Format:	State: Lyophilized purified IgG fraction. Purification: Affinity Chromatography on Protein G. Buffer System: PBS, pH 7.4 without preservatives or stabilizers. Reconstitution: Restore in sterile water to a concentration of 0.1-1.0 mg/ml.
Applications:	FACS analysis (1-10 µg/ml). Immuofluorescence (1-2 µg/ml). Western blot analysis (1-5 µg/ml). Immunohistochemistry on Frozen and Paraffin Sections (6-30 µg/ml). Other applications not tested. Optimal dilutions are dependent on conditions and should be determined by the user.
Specificity:	This antibody reacts with Podoplanin (gp36).
Species Reactivity:	Tested: Human.
Storage:	Prior to reconstitution store at 2-8°C for one month. Following reconstitution store undiluted at 2-8°C for one month or (in aliquots) at -20°C for longer. Avoid repeated freezing and thawing. Shelf life: one year from despatch.
Product Citation:	1. Nerbil Kilic, Leticia Oliveira-Ferrer, Samira Neshat-Vahid, Ster Irmak, Kirstin Obst-Pernberg, Jan-Henner Wurbach, Sonja Loges, Ergin Kilic, Joachim Weil, Heidrun Lauke, Derya Tilki, Bernhard B. Singer, and Süleyman Ergün. Lymphatic reprogramming of microvascular endothelial cells by CEA-related cell adhesion molecule-1 via interaction with VEGFR-3 and Prox1. Blood, Dec 2007; 110: 4223-4233.

For research and in vitro use only. Not for diagnostic or therapeutic work.

Material Safety Datasheets are available at www.acris-antibodies.com or on request.

Antibody Hotline - Technical Questions - Antibody Location Service
Free Call: 0800-2274746 (Germany only) - www.acris-antibodies.com

2. Rainer Riesenberger, Christoph Weiler, Oliver Spring, Martin Eder, Alexander Buchner, Tanja Popp, Mirna Castro, Robert Kammerer, Osamu Takikawa, Rudolf A. Hatz, Christian G. Stief, Alfons Hofstetter, and Wolfgang Zimmermann. Expression of Indoleamine 2,3-Dioxygenase in Tumor Endothelial Cells Correlates with Long-term Survival of Patients with Renal Cell Carcinoma. *Clin. Cancer Res.*, Dec 2007; 13: 6993-7002.
3. Claudius Conrad, Hanno Niess, Ralf Huss, Stephan Huber, Irene von Luetlichau, Peter J. Nelson, Harald C. Ott, Karl-Walter Jauch, and Christiane J. Bruns. Multipotent Mesenchymal Stem Cells Acquire a Lymphendothelial Phenotype and Enhance Lymphatic Regeneration In Vivo. *Circulation*, Jan 2009; 119: 281-289.
4. Joseph D. Turner, R. Stuart Langley, Kelly L. Johnston, Katrin Gentil, Louise Ford, Bo Wu, Maia Graham, Faye Sharpley, Barton Slatko, Eric Pearlman, and Mark J. Taylor: Wolbachia Lipoprotein Stimulates Innate and Adaptive Immunity through Toll-like Receptors 2 and 6 to Induce Disease Manifestations of Filariasis *J. Biol. Chem.*, Aug 2009; 284: 22364 - 22378.
5. Kerstin Birke, Elke Lütjen-Drecoll, Donscho Kerjaschki, and Marco T. Birke: Expression of Podoplanin and Other Lymphatic Markers in the Human Anterior Eye Segment; *Invest. Ophthalmol. Vis. Sci.*, Jan 2010; 51: 344-354.

- General References:**
1. Breiteneder-Geleff et al., *Am. J. Pathol.* 151:1141,1997.
 2. Zimmer et al., *Biochem. J.* 341:277, 1999.
 3. Schacht et al., *EMBO J.* 22:3546-56, 2003.

Protocols:

Sample protocol for Formalin-Fixed Paraffin-Embedded Sections (as a guide only)

- 1) Put Paraffin sections (2-4 µm) onto a Poly-Lysin object holder
- 2) Deparaffination:
 - Xylol (10 min)
 - Acetone (10 min)
 - Acetone / buffer mixture (10 min)
 - Rinse 3x with wash buffer
- 3) Microwave: boil up 3x 10 min in the appropriate buffer (put the whole glass curette into the microwave)
 - a) EDTA buffer: 0,1 M EDTA in aqua dest, pH 8,0
 - b) Citrate buffer/10 mM, pH 6,0A: citric acid, 19,2 g in 1L aqua dest
B: Na-Citrate, 29,41 g in 1L aqua dest
9 ml stock solution A + 50 ml stock solution B, in 500 ml aqua dest (pH adjustment only in solution B)
Cool down the sections to RT for 25 min in wash buffer
- 4) Addition of primary ABs (dilute e.g. in ready to use buffer from DAKO [S3022])
 - Shortly drip off the sections on paper (take 3-4 sections at once)
 - Dust off the liquid beside the sections
 - Add primary Abs
 - incubate 0.5 h at RT
 - Rinse 3x with wash buffer
 - Shortly drip off the sections
- 5) Addition of the by-pass AB (monoclonal mouse-anti-rabbit IgG, e.g. DAKO [M0737] 1: 250)
 - incubate 0.5 h at RT
 - Rinse 3x with wash buffer
 - Shortly drip off the sections
- 6) Addition of anti-mouse-IgG from rabbit 1:
 - Incubate 0.5 h at RT
 - Rinse 3x with wash buffer
 - Shortly drip off the sections

For research and in vitro use only. Not for diagnostic or therapeutic work.

Material Safety Datasheets are available at www.acris-antibodies.com or on request.

Antibody Hotline - Technical Questions - Antibody Location Service
Free Call: 0800-2274746 (Germany only) - www.acris-antibodies.com

7) Addition of the APAAP complexes

- Incubate 0.5 h at RT
- Rinse 3x with wash buffer
- Shortly drip off the sections

8) Addition of the Chromogens (NaphtolAS-MXPhosphat plus FastredTR-Salz)

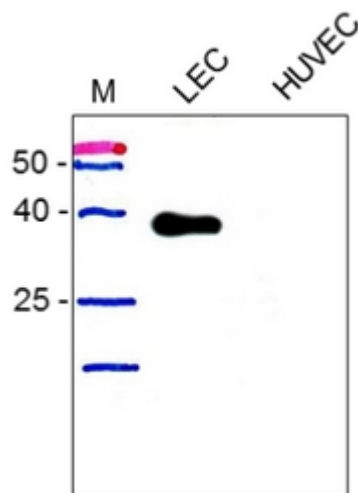
- Incubate for 25 min at RT
- Rinse 3x with wash buffer
- Shortly drip off the sections
- Counterstain: Hämalaun
- 45 sec
- rinse 3x 5 min in lukewarm tap water
- rinse 1x 5 min in dest. H₂O

9) Cover the sections in Glycergel (DAKO)

Note: For steps 6,7,8 use kit from DAKO: Universal-DAKO APAAP Kit Mouse

Note: After each step the sections could stay in the buffer!

Wash buffer (for 40 L): 351g NaCl, 274g Tris-HCL, 36g Tris-Base

Pictures:

Western blot analysis of Podoplanin expression in human lymphatic endothelial cells (LEC) and HUVECs. Total lysate of both cell types were subjected to SDS-PAGE and subsequent Western analysis with the Podoplanin Antibody Cat.-No DM3500P. The antibody recognizes a protein of about 36 kDa in total lysate from LECs but not from HUVEC.

For research and in vitro use only. Not for diagnostic or therapeutic work.

Material Safety Datasheets are available at www.acris-antibodies.com or on request.

Antibody Hotline - Technical Questions - Antibody Location Service
Free Call: 0800-2274746 (Germany only) - www.acris-antibodies.com