Mouse P-Cadherin Antibody

Antigen Affinity-purified Polyclonal Goat IgG
Catalog Number: AF761

DESCRIPTION

Species Reactivity	Mouse
Specificity	Detects P-Cadherin in ELISAs and Western blots. In sandwich immunoassays, less than 2% cross-reactivity with recombinant human (rh) P-Cadherin is observed and less than 0.3% cross-reactivity with recombinant mouse E-Cadherin, rhN-Cadherin, and rhCadherin-8 is observed.
Source	Polyclonal Goat IgG
Purification	Antigen Affinity-purified
Immunogen	Mouse myeloma cell line NS0-derived recombinant mouse P-Cadherin Glu100-Gly647 Accession # Q8BSL6
Endotoxin Level	<0.10 EU per 1 µg of the antibody by the LAL method.
Formulation	Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied either lyophilized or as a 0.2 µm filtered solution in PBS.

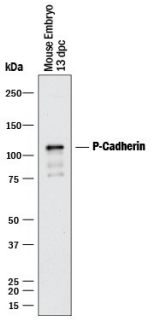
APPLICATIONS

Please Note: *Optimal dilutions should be determined by each laboratory for each application. General Protocols are available in the Technical Information section on our website.*

	Recommended Concentration	Sample
Western Blot	0.5 µg/mL	See Below
Flow Cytometry	0.25 µg/10^6 cells	XB2 mouse teratoma keratinocyte cell line
Immunocytochemistry	5-15 µg/mL	See Below
Immunohistochemistry	0.5-15 µg/mL	See Below
Simple Western	25 µg/mL	See Below
Mouse P-Cadherin Sandwich Immunoassay		**Reagent**
ELISA Capture	0.2-0.8 µg/mL	Mouse P-Cadherin Antibody (Catalog # AF761)
ELISA Detection	0.1-0.4 µg/mL	Mouse P-Cadherin Biotinylated Antibody (Catalog # BAF761)
Standard		Recombinant Mouse P-Cadherin Fc Chimera (Catalog # 761-MP)
CyTOF-ready	Ready to be labeled using established conjugation methods. No BSA or other carrier proteins that could interfere with conjugation.	
Adhesion Blockade	The adhesion of A431 human epithelial carcinoma cells (1 x 10^5 cells/well) to immobilized Recombinant Mouse P-Cadherin Fc Chimera (Catalog # 761-MP, 10 µg/mL, 100 µL/well) was maximally inhibited (80-100%) by 50 µg/mL of the antibody.	

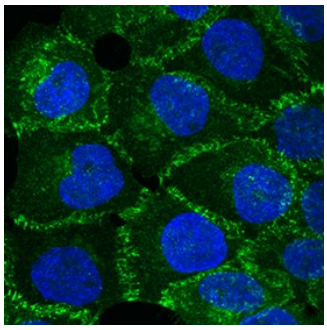
DATA

Western Blot



Detection of P-Cadherin by Western Blot. Western blot shows lysates of mouse embryo tissue. PVDF membrane was probed with 0.5 µg/mL of Goat Anti-Mouse P-Cadherin Antigen Affinity-purified Polyclonal Antibody (Catalog # AF761) followed by HRP-conjugated Anti-Goat IgG Secondary Antibody (Catalog # HAF017). A specific band was detected for P-Cadherin at approximately 115 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.

Immunocytochemistry



P-Cadherin in A431 Human Cell Line. P-Cadherin was detected in immersion fixed A431 human epithelial carcinoma cell line using Goat Anti-Mouse P-Cadherin Antigen Affinity-purified Polyclonal Antibody (Catalog # AF761) at 10 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 493-conjugated Anti-Goat IgG Secondary Antibody (green; Catalog # NL003) and counterstained with DAPI (blue). Specific staining was localized to intercellular junctions. View our protocol for Fluorescent ICC Staining of Cells on Coverslips.

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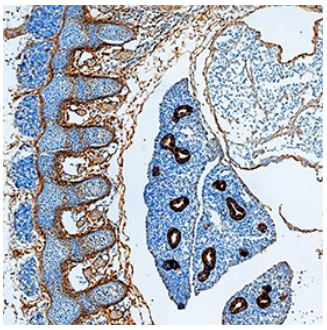
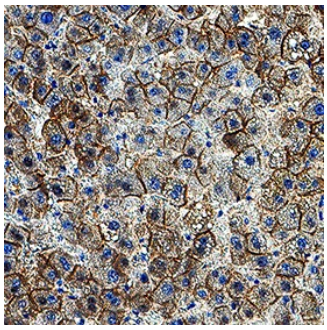
Immunohistochemistry



P-Cadherin in Mouse Embryo. P-Cadherin was detected in immersion fixed frozen sections of mouse embryo (15 dpc) using Goat Anti-Mouse P-Cadherin Antigen Affinity-purified Polyclonal Antibody (Catalog # AF761) at 15 µg/mL overnight at 4 °C. Tissue was stained using the Anti-Goat HRP-DAB Cell & Tissue Staining Kit (brown; Catalog # CTS008) and counterstained with hematoxylin (blue). Specific staining was localized to connective tissue and lungs. View our protocol for Chromogenic IHC Staining of Frozen Tissue Sections.

Immunohistochemistry



Detection of P-Cadherin in Human Liver. P-Cadherin was detected in immersion fixed paraffin-embedded sections of human liver using Goat Anti-Mouse P-Cadherin Antigen Affinity-purified Polyclonal Antibody (Catalog # AF761) at 0.5 µg/ml for 1 hour at room temperature followed by incubation with the Anti-Goat IgG VisUCyte™ HRP Polymer Antibody (Catalog # VC004). Before incubation with the primary antibody, tissue was subjected to heat-induced epitope retrieval using VisUCyte Antigen Retrieval Reagent-Basic (Catalog # VCTS021). Tissue was stained using DAB (brown) and counterstained with hematoxylin (blue). Specific staining was localized to the cell membrane of hepatocytes. View our protocol for IHC Staining with VisUCyte HRP Polymer Detection Reagents.

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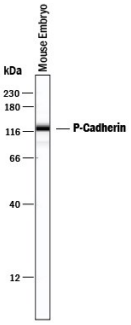
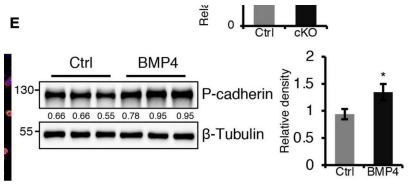
Simple Western



Detection of Mouse P-Cadherin by Simple Western™. Simple Western lane view shows lysates of mouse embryo tissue, loaded at 0.2 mg/mL. A specific band was detected for P-Cadherin at approximately 115 kDa (as indicated) using 25 µg/mL of Goat Anti-Mouse P-Cadherin Antigen Affinity-purified Polyclonal Antibody (Catalog # AF761) followed by 1:50 dilution of HRP-conjugated Anti-Goat IgG Secondary Antibody (Catalog # HAF109). This experiment was conducted under reducing conditions and using the 12-230 kDa separation system.

Western Blot



Detection of Mouse P-Cadherin by Western Blot BMPR1a regulated P-cadherin expression via p63 and Slug. (A) Immunohistochemistry staining for P-cadherin in control (n = 4 mice) and cKO (n = 3 mice) mammary glands at pregnancy day 14.5. Scale bar, 50 µm. (B) Western blotting for P-cadherin in mammary epithelial cells isolated from control and cKO mice at pregnancy day 14.5. β-Tubulin was used as a loading control. Statistical analysis the expression of P-cadherin/β-Tubulin. n = 3 mice. (C) qRT-PCR analysis of Cdh3 in FACS-sorted control and cKO myoepithelial cells at pregnancy day 14.5. n = 4 biological replicates. (D) P-cadherin (red) and K14 (green) double immunofluorescence staining in HC11 mammary epithelial cells treated with BMP4 (50 ng/mL) for 24 h. n = 3 biological replicates. Scale bar, 25 µm. (E) Western blotting for P-cadherin in HC11 mammary epithelial cells treated with BMP4 (50 ng/mL) for 24 h. β-Tubulin was used as a loading control. Statistical analysis the expression of P-cadherin/β-Tubulin. n = 3 biological replicates. (F) Scatter plot showing the correlation between BMPR1A and CDH3 expression in mammary glands from TCGA and GTEx data. Pearson's coefficient test was performed to assess statistical significance. (G) Western blotting for P-cadherin in HC11 cells treated with p63 siRNA (sip63)/Slug siRNA (siSlug) and scramble RNA (NC) at 48 h. β-Actin was used as a loading control. Statistical analysis the expression of P-cadherin/β-Actin. n = 3 biological replicates. (H) Immunofluorescence for P-cadherin (green) in HC11 cells treated with p63 siRNA (sip63)/Slug siRNA (siSlug) and scramble RNA (NC) at 48 h. n = 3 biological replicates. Scale bar, 25 µm. (I) Western blotting for K14, P-cadherin, p63 and Slug in HC11 cells treated with scramble RNA, p63 siRNA, Slug siRNA, or p63 siRNA and Slug siRNA at 48 h. β-Actin was used as a loading control. Statistical analysis the expression of K14/β-Actin, P-cadherin/β-Actin, p63/β-Actin and Slug/β-Actin. n = 3 biological replicates. Data were presented as means ± SD. *p < 0.05, **p < 0.01, ***p < 0.001. Image collected and cropped by CiteAb from the following open publication (https://pubmed.ncbi.nlm.nih.gov/34336839), licensed under a CC-BY license. Not internally tested by R&D Systems.

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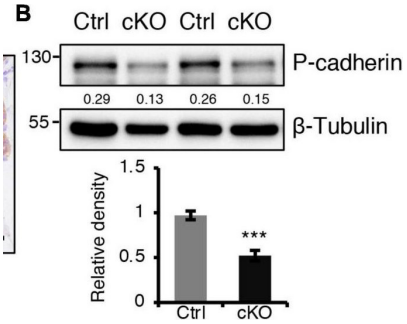

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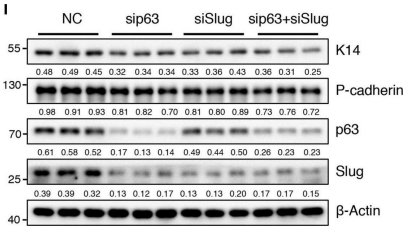
Western Blot



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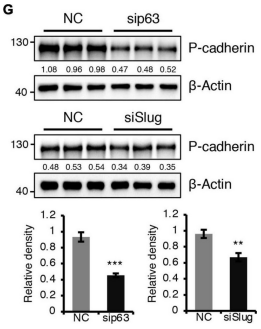

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PREPARATION AND STORAGE

Reconstitution	Reconstitute at 0.2 mg/mL in sterile PBS. For liquid material, refer to CoA for concentration.
Shipping	Lyophilized product is shipped at ambient temperature. Liquid small pack size (-SP) is shipped with polar packs. Upon receipt, store immediately at the temperature recommended below.
Stability & Storage	**Use a manual defrost freezer and avoid repeated freeze-thaw cycles.** • 12 months from date of receipt, -20 to -70 °C as supplied. • 1 month, 2 to 8 °C under sterile conditions after reconstitution. • 6 months, -20 to -70 °C under sterile conditions after reconstitution.

BACKGROUND

Placental Cadherin (P-Cadherin or PCAD) is a member of the cadherin family of cell adhesion molecules. Cadherins are calcium-dependent transmembrane proteins, which bind to one another in a homophilic manner. On their cytoplasmic side, they associate with the three catenins, α, β, and γ (plakoglobin). This association links the cadherin protein to the cytoskeleton. Without association with the catenins, the cadherins are non-adhesive. Cadherins play a role in development, specifically in tissue formation. They may also help to maintain tissue architecture in the adult. P-Cadherin is a classical cadherin molecule. Classical cadherins consist of a large extracellular domain which contains DXD and DXNDN repeats responsible for mediating calcium-dependent adhesion, a single-pass transmembrane domain, and a short carboxy-terminal cytoplasmic domain responsible for interacting with the catenins. Constitutive P-Cadherin expression is found in the epidermis, mesothelium, corneal epithelium, and uterine decidua. Mouse P-Cadherin is an 822 amino acid (aa) protein with a 27 aa signal sequence and a 795 aa propeptide. The mature protein begins at aa 100 and has a 542 aa extracellular region, a 27 aa transmembrane region, and a 153 aa cytoplasmic region.

References:

1. Bussemakers, M.J.G. *et al.* (1993) Mol. Biol. Reports **17**:123.
2. Overduin, M. *et al.* (1995) Science **267**:386.
3. Takeichi, M. (1991) Science **251**:1451.
4. Nose, A. *et al.* (1987) EMBO J. **6**:3655.

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