antibodies

Datasheet for ABIN1386389 anti-Plakophilin 2 antibody (AA 801-881)

1 Validation 2 Images 1





Overview

Quantity:	100 μL	
Target:	Plakophilin 2 (PKP2)	
Binding Specificity:	AA 801-881	
Reactivity:	Human	
Host:	Rabbit	
Clonality:	Polyclonal	
Conjugate:	This Plakophilin 2 antibody is un-conjugated	
Application:	ELISA, Immunohistochemistry (Paraffin-embedded Sections) (IHC (p)), Immunocytochemistry (ICC), Immunohistochemistry (Frozen Sections) (IHC (fro)), Immunofluorescence (Cultured Cells) (IF (cc)), Immunofluorescence (Paraffin-embedded Sections) (IF (p))	

Product Details

Immunogen:	KLH conjugated synthetic peptide derived from human Plakophilin 2	
Isotype:	lgG	
Cross-Reactivity:	Human	
Predicted Reactivity:	Mouse,Rat,Dog,Cow,Sheep,Pig,Horse,Rabbit	
Purification:	Purified by Protein A.	
Target Details		

Target:

Plakophilin 2 (PKP2)

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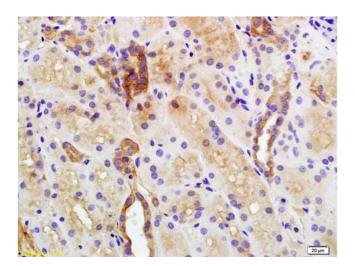
Alternative Name:	Plakophilin 2 (PKP2 Products)	
Background:	Synonyms: ARVD 9, ARVD-9, ARVD9, PKP 2, PKP2, PKP-2, Plakophilin-2. Plakophilin2,	
	PKP2_HUMAN. Background: Plakophilins 1, 2, 3 and 4 (PKP1-4) influence development and participate in linking	
	cadherins to cytoskeletal intermediate filaments. Plakophilins 1-4 contain arm-repeat	
	(armadillo) domains, and localize to nuclei and cell desmosomes (cell-cell junctions found in	
	suprabasal layers of stratifying epithelia that undergo mechanical stress). Plakophilin-1	
	mediates increases in desmosomal protein content, desmosome assembly, and regulation of	
	cell migration. Plakophilin-2 is important for desmosome assembly and is an essential	
	morphogenic factor and architectural component of the heart. Plakophilin-3 plays a role in both	
	desmosome-dependent adhesion and signaling pathways. Plakophilin-4 is a component of	
	desmosomal adhesion plaques that regulates junctional plaque organization and cadherin	
	function.	
Gene ID:	5318	
UniProt:	Q99959	
Pathways:	Cell-Cell Junction Organization, SARS-CoV-2 Protein Interactome, The Global Phosphorylation	
	Landscape of SARS-CoV-2 Infection	
Application Details		
Application Notes:	ELISA 1:500-1000	
	IHC-P 1:200-400	
	IHC-F 1:100-500	
	IF(IHC-P) 1:50-200	
	IF(IHC-F) 1:50-200	
	IF(ICC) 1:50-200	
	ICC 1:100-500	
Restrictions:	For Research Use only	
Handling		
Format:	Liquid	
Concentration:	1 µg/µL	
Buffer:	0.01M TBS(pH 7.4) with 1 % BSA, 0.02 % Proclin300 and 50 % Glycerol.	

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Preservative:	ProClin	
Precaution of Use:	This product contains ProClin: a POISONOUS AND HAZARDOUS SUBSTANCE, which should be handled by trained staff only.	
Storage:	4 °C,-20 °C	
Storage Comment:	Shipped at 4°C. Store at -20°C for one year. Avoid repeated freeze/thaw cycles.	
Expiry Date:	12 months	
Publications		
Product cited in:	Akdis, Medeiros-Domingo, Gaertner-Rommel, Kast, Enseleit, Bode, Klingel, Kandolf, Renois,	
	Andreoletti, Akdis, Milting, Lüscher, Brunckhorst, Saguner, Duru: "Myocardial expression profiles	
	of candidate molecules in patients with arrhythmogenic right ventricular	
	cardiomyopathy/dysplasia compared to those with dilated cardiomyopathy and healthy	
	controls." in: Heart rhythm , Vol. 13, Issue 3, pp. 731-41, (2016) (PubMed).	

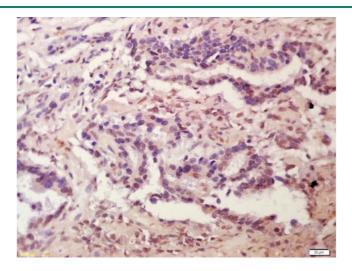
Validation report #103656 for Immunofluorescence (IF)



Immunohistochemistry

Image 1. Formalin-fixed and paraffin embedded human kidney labeled with Anti-Plakophilin 2 Polyclonal Antibody, Unconjugated (ABIN1386389) at 1:200 followed by conjugation to the secondary antibody and DAB staining

Validation report #103656 for Immunofluorescence (IF)



Immunohistochemistry

Image 2. Formalin-fixed and paraffin embedded human colon carcinoma labeled with Anti-Plakophilin 2 Polyclonal Antibody, Unconjugated (ABIN1386389) at 1:200 followed by conjugation to the secondary antibody and DAB staining

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NDEPENDEN	Successfully validated (Immunofluorescence (IF))	
Д	by Gencardio (IDIBGI)	
	Report Number: 103656	
VALIDATION	Date: Mar 13 2019	
103656 13/03/19		
Target:	PKP2	
Lot Number:	AD102900	
Method validated:	Immunofluorescence (IF)	
Positive Control:	HL1 cells	
	HeLa cells	
	mouse heart tissue	
Negative Control:	HL1 cells	
	HeLa cells	
	secondary antibody only control	
Notes:	ABIN1386389 specifically labels the targeted antigen in mouse heart. No signal was detected in	
	sample negative control and the secondary antibody only control.	
Primary Antibody:	ABIN1386389	
Secondary Antibody:	goat anti-rabbit AF488 antibody (Invitrogen, A11008, 1911238)	
Protocol:	HL1 and HeLa cells	
	 Grow HL1 cells in Claycomb medium (sigma, 51800C, SLBX8700) supplemented with 10% FBS (Gibco, 10270-106, 42G5178K), 1% Penicillin-Streptomycin (Sigma, P4333, lot 	
	048M4874V) and 1% Glutamax (Gibco, 35050-061, lot 2037045), at 37°C and 5% CO ₂ in	
	12mm coverslips in a well of 12 well plate.	
	 Grow HeLa cells in MEM/EBSS medium (GE Life Science, SH30244.01, lot AC14563283) 	
	supplemented with 10% FBS (Gibco, 10270-106, lot 42G5178K), 1% Penicillin-Streptomycir	
	(Sigma, P4333, lot 048M4874V) and 1% Glutamax (Gibco, 35050-061, lot 2037045), at	
	 37°C and 5% CO₂ in 12mm coverslips in a well of 12 well plate. Transfect HeLa cells with PKP2a plasmid (p915 PKP2a-pFLAG-cmv5a, provided by Dr. 	
	 Iransfect HeLa cells with PKP2a plasmid (p915 PKP2a-pFLAG-cmv5a, provided by Dr. Green KJ) using Lipofectamine 3000 (Invitrogen, L3000-015, lot 2041107) following the 	
	manufacturer's instructions.	
	 Wash cells 1x with PBS. 	
	 Fix cells on coverslips in 4% PFA for 20min at RT. 	
	 Wash cells 3x for 5min with PBS. 	
	$_{\odot}$ Incubate coverslips with quench solution (PBS with 75mM glycine and 20mM NH ₄ Cl) for	

10min at RT.

- Wash cells 1x with PBS.
- Permeabilize cells and block non-specific binding with PFS-like buffer containing 0.1% BSA,
 0.1% triton and 0.02% NaN₃ for 30min at 37°C.
- Incubate cells with primary anti-Plakophilin 2 (PKP2) (AA 831-881) antibody (antibodiesonline, ABIN1386389, lot AD102900) diluted 1:60 in PFS-like buffer or with anti-Flag-tag antibody (Sigma, monoclonal anti-flag, F316, lot SLBT7654) diluted 1:400 in PFS-like buffer ON at 4°C.
- Wash cells 3x for 5min with PBS.
- Incubate cells with secondary goat anti-rabbit AF488 antibody (Invitrogen, A11008, lot 1911238) or with secondary goat anti-mouse AF568 antibody (Invitrogen, A11031, lot 1841757) diluted 1:200 in PFS-like buffer for 30min at RT.
- Wash cells 2x for 5min with PFS-like buffer.
- Wash cells 2x for 5min with PBS.
- DAPI counterstain for 5min at RT.
- Mount coverslips on glass slides in Calbicochem (Millipore, 345789, lot 2682585).
- Image acquisition with NIS-ELEMENTS (Nikon).
- Tissue: mouse heart
 - Obtain mouse heart mouse and wash it with PBS.
 - $\,\circ\,\,$ Fix tissue in 15ml falcon tube with 4% PFA ON at 4°C.
 - Wash the fixed heart with water.
 - Embed tissue in paraffin.
 - Cut the tissue using a microtrom and put it into slides.
 - Desparaffinize the tissue in the slide (from xylene to water).
- Incubate the slides with retrieval solution for 30min at 95°C to permeabilize the tissue.
- Cool slides down on ice.
- Wash tissue 2x for 3min with PBS.
- Wash tissue 2x for 3min with PBS 0.1% triton.
- Block non-specific binding with PBS 0.1% triton 10% BSA for 1h at RT.
- Incubate tissue with primary anti-PKP2 (AA 831-881) antibody (antibodies-online, ABIN1386389, lot AD102900) diluted 1:100 in PBS 1% BSA solution ON at 4°C.
- Wash tissue 2x for 5min with PBS 0.1% triton.
- Incubate tissue with secondary goat anti-rabbit AF488 antibody (Invitrogen, A11008, 1911238) diluted 1:200 in PBS 1% BSA solution for 1h at RT.
- Wash cells 2x for 5min with PBS.
- Wash tissue 2x for 3min with PBS 0.1% triton.
- Wash tissue 2x for 3min with PBS.
- DAPI counterstain for 5min at RT.
- Mount slides with coverslips in Calbicochem (Millipore, 345789, lot 2682585).
- Image acquisition with NIS-ELEMENTS (Nikon).

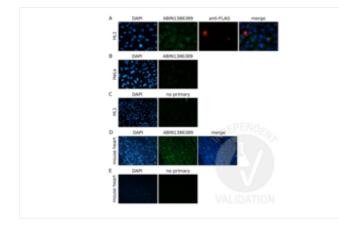
Experimental Notes:

ABIN1386389 did not label recombinantly expressed PKP2-FLAG in transfected HL1 cells. A

hypothesis could be that antibody recognizes the murine protein in mouse heart samples but

not the human sequence of the vector (transfected HL1).

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Validation image no. 1 for anti-Plakophilin 2 (PKP2) (AA 801-881) antibody (ABIN1386389)

A. IF staining of cells expressing PKP2-FLAG using
ABIN1386389 (green) or an anti-FLAG-tag antibody (red).
Nuclear staining with DAPI (blue) (magnification 60x). B.
Staining of HeLa cells not expressing PKP2-FLAG using
ABIN1386389 (magnification 40x). C. Staining of cells
expressing PKP2-Flag with a secondary antibody only
(magnification 40x). D and E. Staining of endogenous Pkp2
in mouse heart tissue with ABIN1386389 (D) or secondary
antibody only (E) (magnification 40x).

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