Datasheet for ABIN3042435

anti-GRK6 antibody (C-Term)

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Overview

Quantity:	100 µg
Target:	GRK6
Binding Specificity:	AA 382-417, C-Term
Reactivity:	Human, Rat
Host:	Rabbit
Clonality:	Polyclonal
Conjugate:	This GRK6 antibody is un-conjugated
Application:	Western Blotting (WB)

Product Details

Purpose:	Rabbit IgG polyclonal antibody for G protein-coupled receptor kinase 6(GRK6) detection. Tested with WB in Human,Rat.
Immunogen:	A synthetic peptide corresponding to a sequence at the C-terminus of human GRK6 (382-417aa QSPFQQRKKKIKREEVERLVKEVPEEYSERFSPQAR), different from the related mouse sequence by four amino acids, and from the related rat sequence by three amino acids.
Sequence:	QSPFQQRKKK IKREEVERLV KEVPEEYSER FSPQAR
Isotype:	IgG
Cross-Reactivity (Details):	No cross reactivity with other proteins.
Characteristics:	Rabbit IgG polyclonal antibody for G protein-coupled receptor kinase 6(GRK6) detection. Tested with WB in Human,Rat. Gene Name: G protein-coupled receptor kinase 6

Product Details

	Protein Name: G protein-coupled receptor kinase 6
Purification:	Immunogen affinity purified.

Target Details

Target:	GRK6
Alternative Name:	GRK6 (GRK6 Products)
Background:	G protein-coupled receptor kinase 6 is an enzyme that in humans is encoded by the GRK6 gene. It is mapped to 5q35. This gene encodes a member of the guanine nucleotide-binding protein (G protein)-coupled receptor kinase subfamily of the Ser/Thr protein kinase family. The protein phosphorylates the activated forms of G protein-coupled receptors thus initiating their deactivation. Several transcript variants encoding different isoforms have been described for this gene. Also, GRK6 appears to be involved in responses to morphine. Synonyms: FLJ32135 antibody|G protein coupled receptor kinase 6 antibody|G protein coupled receptor kinase GRK6 antibody|G protein-coupled receptor kinase 6 antibody|G protein-coupled receptor kinase GRK6 antibody|Gprk6 antibody|Grk6 antibody|GRK6_HUMAN antibody
Gene ID:	2870
UniProt:	P43250
Pathways:	Myometrial Relaxation and Contraction, Regulation of G-Protein Coupled Receptor Protein Signaling, CXCR4-mediated Signaling Events, Negative Regulation of Transporter Activity

Application Details

Application Notes:	WB: Concentration: 0.1-0.5 µg/mL, Tested Species: Human, Rat Notes: Tested Species: Species with positive results. Other applications have not been tested. Optimal dilutions should be determined by end users.
Comment:	Antibody can be supported by chemiluminescence kit ABIN921124 in WB.
Restrictions:	For Research Use only

Handling

Format:	Lyophilized
Reconstitution:	Add 0.2 mL of distilled water will yield a concentration of 500 µg/mL.

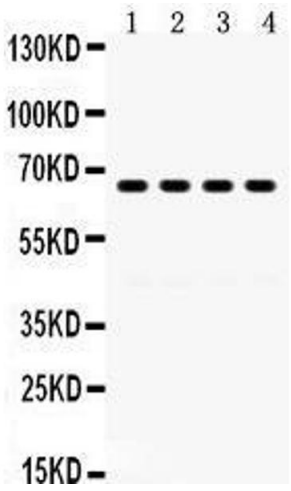
Handling

Concentration:	500 µg/mL
Buffer:	Each vial contains 5 mg BSA, 0.9 mg NaCl, 0.2 mg Na2HPO4, 0.05 mg Sodium azide.
Preservative:	Sodium azide
Precaution of Use:	This product contains Sodium azide: a POISONOUS AND HAZARDOUS SUBSTANCE which should be handled by trained staff only.
Handling Advice:	Avoid repeated freezing and thawing.
Storage:	4 °C/-20 °C
Storage Comment:	At -20°C for one year. After reconstitution, at 4°C for one month. It can also be aliquotted and stored frozen at -20 °C for a longer time. Avoid repeated freezing and thawing.

Publications

Product cited in:	Reichel, Weitzel, Klement, Hoffmann, Drube: "Suitability of GRK Antibodies for Individual Detection and Quantification of GRK Isoforms in Western Blots." in: **International journal of molecular sciences**, Vol. 23, Issue 3, (2022) (PubMed).

Images



Western Blotting

Image 1.

Successfully validated (Western Blotting (WB))

by Institut für Molekulare Zellbiologie, Universitätsklinikum Jena

Report Number: 103590

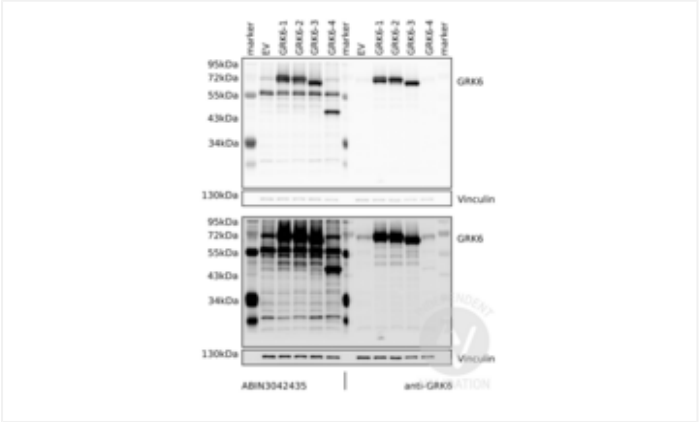
Date: Jan 31 2019

Target:	GRK6
Lot Number:	0971512Da9609109
Method validated:	Western Blotting (WB)
Positive Control:	HEK293 cells transiently transfected with pcDNA3 vectors containing GRK6 transcript variant 1 (NM_001004106.3), transcript variant 2 (NM_002082.3), transcript variant 3 (NM_001004105.2), or transcript variant 4 (NM_001364164.1)
Negative Control:	HEK293 cells transiently transfected with empty pcDNA3 vector
Notes:	The GRK6 antibody ABIN3042435 is able to detect all four tested GRK6 isoforms, but gives strong background-signals around 55kDa.
Primary Antibody:	ABIN3042435
Secondary Antibody:	goat-anti-rabbit Peroxidase-labeled antibody (SeraCare, 5220-0336)
Protocol:	• Grow 7.5x10^5 HEK293 cells in DMEM-medium (Sigma-Aldrich, D6429)) supplemented with 10% Fetal Bovine serum (Sigma-Aldrich, F7524) and 1% penicillin-streptomycin (Sigma-Aldrich, P0781), at 37°C and 5% CO_2 in 2ml on a 6-well-dish (Greiner Bio-One) ON. • Transfect cells with 2µg of respective pcDNA3 construct using self-prepared PEI transfection reagent. • After 24h lyse the cells in 250µl per well cold RIPA lysis buffer (1 % NP-40, 1mM EDTA, 50mM Tris-HCl pH7.4, 150mM NaCl, 0.25 % Sodium-deoxycholate, PhosSTOP tablet (Roche, 04906845001) and cOmplete tablet (Roche, 04693132001) diluted in RIPA following the manufacturer´s instructions. • Denature the cleared lysate of total protein for 5min at 95°C in 50µl of 6x SDS sample buffer and subsequently separate 4µl of each sample on a 10% polyacrylamide gel. • Transfer proteins onto nitrocellulose membrane (Biostep 01-14-101) with a Tank Blotting System at 10V ON. • Block the membrane with 1x Casein Blocking Buffer (Sigma-Aldrich, B6429) for 1h at RT with gentle shaking. • Wash membrane 3x for 10min with TBST. • Cut the membrane and incubate fragments separately with primary ◦ rabbit anti-GRK6 antibody (ABIN3042435, antibodies-online, lot 0971512Da9609109)

diluted 1:1000 in 5% BSA-TBST at 4°C ON.
    - rabbit anti-GRK6 antibody (Cell Signaling Technology, 5878, lot 1) diluted 1:1000 in 5% BSA-TBST at 4°C ON.
    - rabbit anti-Vinculin antibody (Biozol, BZL03106, lot 0401) diluted 1:1000 in 5% BSA-TBST at 4°C ON.
- Wash membrane 3x for 10min with TBST.
- Incubate membrane with secondary goat-anti-rabbit Peroxidase-labeled antibody (SeraCare, 5220-0336) diluted 1:10000 in 1x Casein Blocking Buffer) for 1h at RT with gentle shaking.
- Wash membrane 3x for 10min with TBST.
- Reveal protein bands using Western Lightning Plus ECL reagent (Perkin Elmer, NEL103001EA, Luminol reagent Lot 275-17431, Oxidizing reagent, lot 265-17431) on a LAS-4000 Luminescence Imager (Fujifilm), exposure for 15 and 90 seconds.

Experimental Notes:	If the detection of the smallest isoform (GRK6-4, transcript variant 4, NM_001364164.1) is required, ABIN3042435 is a good choice, as this isoform is not detected by the alternative GRK6 antibody tested in parallel. Nevertheless, the strong background signal around 55kDa produced by ABIN3042435 diminishes its usefulness.

Image for Validation report #103590



Validation image no. 1 for anti-G Protein-Coupled Receptor Kinase 6 (GRK6) (AA 382-417), (C-Term) antibody (ABIN3042435)

Western blot analysis of HEK293 cell lysates subsequently to transient transfection with pcDNA3 constructs containing no insert (EV), GRK6 transcript variant 1, NM_001004106.3, transcript variant 2, NM_002082.3, transcript variant 3, NM_001004105.2, or transcript variant 4, NM_001364164.1 as indicated. Equal amounts of the lysates were loaded twice on the same gel and after blotting, the membrane was cut and incubated with ABIN3042435, a reference anti-GRK6 antibody (GRK6), and Vinculin loading control antibody (Vinculin) in parallel. Exposure for the upper panel was 15 seconds and for the lower 90 seconds.