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# Datasheet for ABIN1304779 anti-KCNQ4 antibody

1 Validation

1 Publication



#### Overview

Quantity:	100 μL
Target:	KCNQ4
Reactivity:	Mammalian
Host:	Mouse
Clonality:	Monoclonal
Application:	Western Blotting (WB), Immunohistochemistry (IHC), Immunostaining (ISt)

## Product Details

Clone:	N43-6
Isotype:	lgG1
Characteristics:	K+ channels
Purification:	Purified: Hybridoma in vivo ascites fluid purified by SAS (Saturated Ammonium Sulfate) precipitation followed by anion exchange chromatography. Purified mAbs are >90% specific antibody at 1mg/mL.

### Target Details

Target:	KCNQ4
Alternative Name:	Kv7.4/KCNQ4 (KCNQ4 Products)
UniProt:	P56696
Pathways:	Sensory Perception of Sound

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Application Details	
Restrictions:	For Research Use only
Handling	
Format:	Liquid
Publications	
Product cited in:	Debowska, Poleszczuk, Wojcik-Zaluska, Ksiazek, Zaluska: "Phosphate Kinetics During Weekly
	Cycle of Hemodialysis Sessions: Application of Mathematical Modeling." in: Artificial organs, (
	2015) (PubMed).



NDEPENDENA	Successfully validated (Western Blotting (WB))
	by Dipartimento di Medicina e Scienze della Salute, Università degli Studi del Molise,
	Campobasso
VALIDATION	Report Number: 102037
CUSTOMER VALIDATION N° DATE 102037 31/10/17	Date: Oct 31 2017
Target:	Kv7.4/KCNQ4
Lot Number:	440-2HK-65b
Method validated:	Western Blotting (WB)
Positive Control:	CHO cells transfected with a plasmid containing the cDNA for the longest isoform of the human
	Kv7.4/KCNQ4 channel
Negative Control:	Non-transfected CHO cells (no Kv7 channel expression)
Notes:	Passed. ABIN1304779 specifically detects KCNQ4/Kv7.4 in lysates from CHO cells expressing
	human KCNQ4/Kv7.4.
Primary Antibody:	ABIN654696
Secondary Antibody:	goat anti-mouse IgG (H+L) antibody (ImmunoReagents, GTXMU-003-DHRPX, lot 31-118- 043013)
Protocol:	<ul> <li>Grow CHO cells (Chinese Hamster Ovary cells) in DMEM medium (Microgem, L0106-500) supplemented with 10% fetal bovine serum (Microgem, S1810-500) and antibiotics (1X Penicillin-Streptomycin mix, Microgem, L0022-100), at 37°C and 5% CO<sub>2</sub> in 60mm plates.</li> <li>Transfect CHO cells with 5µg plasmid expressing either Kv7.1, Kv7.2, Kv7.3, Kv7.4, or Kv7.5 and 1µg of plasmid expressing EGFP as a transfection marker using Lipofectamine 2000 (Invitrogen, 11668019) following the manufacturer's instructions.</li> <li>Lyse cells in an appropriate volume of B-buffer, containing: 150mM NaCl, 10mM Tris-HCl pH7.4, 1mM EDTA, 1% SDS and 1x protease inhibitors (Roche, 11836145001, lot 11231400).</li> <li>Determine total protein content of the lysates using a Bradford protein assay (AppliChem, A6932, lot 6J015116).</li> <li>Denature 20µg of total protein for 10min at 100°C in 1/3 volume of 4x Laemmli SDS sample buffer and subsequently separate them on a denaturing 8% SDS-PAGE gel in Mini-PROTEAN Electrophoresis Cells (Bio-Rad) for 2-3h at 100 V.</li> <li>Transfer proteins onto PVDF filter (Bio-Rad, 1620177, lot 28369A01) with transfer buffer (25mM Tris, 192mM glycine, 20% methanol) in a western blotting system at 4°C at 40mA ON.</li> <li>Block the membrane with blocking buffer (5% milk dissolved in PBS + 1% Tween20, PBS-T) for 1h at RT.</li> <li>Incubation with primary</li> </ul>

- monoclonal mouse anti-KCNQ4/Kv7.4 antibody (antibodies-online, ABIN1304779, lot 440-2HK-65b) diluted 1:1000 in blocking buffer ON at 4°C or
- mouse anti-alpha-tubulin (Sigma, T8203, lot 024M4827V) diluted 1:5000 in blocking buffer ON at 4°C.
- Wash membrane 3x for 10min with PBS-T.
- Incubation with secondary goat anti-mouse IgG (H+L) antibody (ImmunoReagents, GTXMU-003-DHRPX, lot 31-118-043013) diluted 1:5000 in blocking buffer for 1h at RT.
- Wash membrane 3x for 10min with PBS-T.
- Treat PVDF filters with chemiluminescence solutions (ECL, Promega) for 5min at RT.
- Images were captured, stored, and analyzed with the ImageLab, version 4.1 analysis software (Bio-Rad).

Image for Validation report #102037



Validation image no. 1 for anti-Potassium Voltage-Gated Channel, KQT-Like Subfamily, Member 4 (KCNQ4) antibody (ABIN1304779)

Western blot analysis of KCNQ4 subunits using ABIN1304779. Total lysates from non-transfected (NT) CHO cells or cells transfected with plasmids expressing human Kv7.1 (Q1; NM\_019842), Kv7.2 (Q2; NM\_004518.4), Kv7.3 (Q3; NM\_004519.3), Kv7.4 (Q4; NM\_004700.3), or Kv7.5 (Q5; NM\_019842) cDNAs were prepared and separated as described in the protocol. Alpha-tubulin served as loading control.

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