Supplementary information

Inactivation of the Kv2.1 channel through electromechanical coupling

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Inactivation of the Kv2.1 channel through electromechanical coupling

Ana I. Fernández-Mariño ^{1†}, Xiao-Feng Tan ^{1†}, Chanhyung Bae ^{1†}, Kate Huffer ^{1,2}, Jiansen Jiang ³ and Kenton J. Swartz ^{1*}

¹ Molecular Physiology and Biophysics Section, Porter Neuroscience Research Center, National Institute of Neurological Disorders and Stroke, National Institutes of Health, Bethesda, MD 20892

² Department of Biology, Johns Hopkins University, 3400 N. Charles Street, Baltimore, MD 21218

³ Laboratory of Membrane Proteins and Structural Biology and Biophysics Center, National Heart, Lung, and Blood Institute, National Institutes of Health, Bethesda, MD 20892

+ These authors contributed equally to this work

* Corresponding author. Email: swartzk@ninds.nih.gov

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Supplementary Information Figure 1|

Data processing workflow for the structure of Kv2.1 (1-598)



Supplementary Information Figure 2|

Data processing workflow for the structure of the L403A mutant of Kv2.1 (1-598)

Supplementary Information Table 1. Gating properties of epileptic encephalopathy mutations in Kv2.1.

	V _{1/2} (mV)	SEM	Z	SEM	n	У
Kv2.1 WT	-4.8	2.0	3.7	0.2	4	2
S198F	51.1	3.3	1.1	0.1	5	2
T206K	13.7	4.5	1.0	0.1	3	2
T206M	11.1	4.6	1.7	0.3	3	2
L207P	-9.4	5.4	1.2	0.2	3	2
R302C	-82.0	3.5	1.6	0.4	5	2
R308H	71.6	3.3	0.9	0.0	6	4
K387N	-15.1	1.0	5.1	0.7	6	6

Voltage-activation (G-V) relations were obtained using tail current measurements in 100 mM external K⁺. The Boltzmann equation was fit to G-V relations to obtain values of $V_{1/2}$ (midpoint) and z (steepness). Values of n indicate the number of cells studied and values of y indicate the number of independent experiments.

			S4-S5 linker	S5		Pore helix		S6	
					~~~~~	~~~~~	~~~~~~	$\sim\sim\sim\sim$	$\sim\sim\sim\sim$
Kup and I	) from Cr	w and New shappels	1316	1329				1403	F412
KVS anu L		av and hav charmers						1.00	· · · -
		(0.4.0. 40.0)						I	
KV2.1	(65044)	(310-428)	STGLQSLGFTLRR	SYNELGLLILFLAMGIN	IIF SSLVFFAEKDEDDTKF	KSIPASEWWATITMTTVG	YGDIYPKTILGKIVGGLCCIAGVLVI	ALPIPLIVNN	FSEFYKEQKRQEKAIK
Shakar	(0EBINI) (7SID)	(307-417)	SKGLQILGQTLKA	SMRELGLLIFFLFIGVI	LESSAVYFAEADERDSQF	PSIPDAFWWAVVSMTTVG	YGDMVPTTIGGKIVGSLCAIAGVLTI	ALPVPVIVSN	FNYFYHRE
Kud 2	(755)	(379-400)	SKOLQILORTLKA	SMRELOLLIFFLFIOV	LISSAVYFAEAGSENSF	<b>RS I PDAFWWAVVTMTTVG</b>	YODMTPVOVWORIVOSECAIAOVETI	ALPVPVIVSN	FNYFYHR
Ku1 3	(7557)	(381 400)	SKOLOILOUILKA	SMRELGLLIFFLFIGVI	LESSAVYFAEADDPISGF	SSTPDAFWWAVVIMITVG	YODMHPVTTGGKTVGSLCATAGVLTT	ALPVPVIVSN	PNYFYHR
Kv1.3	(755Y)	(381-490)	SKOLOLLOOTLKA	SMRELGLL IFFLFIGVI	LESSAVTFAEADDPISG	SSTPDAFWWAVVTMTTVG	YODMHPVTTGGKTVGSLCATAGVLTT	ALPVPVIVSN	
Kv1.3	(7557)	(381-490)	SKGLOLLGOTLKA	SMPELGILIEFIEIGVI	L ESSAVYEAEADDRTSG	SS I PDA FWW AVV TMTTVG	YONHPYTICCKIVOSI CALACVITI	AL PVPVIVSN	ENVEYUP
Kv3.1	(7PHH)	(327-452)	FVGLRVLGHTLRA	STNEFLLLIIFLALGVI	IFATMIYYAERASENTHE	KNIPIGEWWAVVTMTTLG	YGDMYPO TWSGML VGAL CALAGVLTI	AMPVPVIVNN	FGMYYSLAMAKOK
Kv3.1	(7PQT)	(327-452)	FVGLRVLGHTLRA	STNEFLLLIIFLALGVI	IFATMIYYAERASENTHE	KNIPIGEWWAVVTMTTLG	YODMYPO TWSGMLVGALCALAGVLTI	AMPVPVIVNN	FOMYYSLAMAKOK ···
Kv4.2	(7UKĆ)	(307-424)	SOGLEILGYTLKS	CASELGFLLFSLTMAI	I I FATVMFYAEKGSSASK	TSIPAAFWYTIVTMTTLG	YGDMVPKTIAGKIFGSICSLSGVLVI	ALPVPVIVSN	FSRIYHQNQRADKRR-
Kv4.2	(7UKD)	(307-424)	SOGLRILGYTLKS	CASELOFLLFSLTMAII	I I FATVMFYAEKGSSASKF	TS I PAAFWYT I VTMTTLG	YGDMVPKTIAGKIFGSICSLSGVLVI	ALPVPVIVSN	FSRITHQNQRADKRR
Kv4.2	(7UKE)	(307-424)	SQGLRILGYTLKS	CASELOFLLFSLTMAII	II <mark>F</mark> ATVM <mark>FY</mark> AEKGSSASKF	TSIPAAFWYTIVTMTTLG	YGDMVPKT I AGK I FGSICSLSGVLVI	ALPVPVIVSN	FSRIYHQNQRADKRR-
Kv4.2	(7UKF)	(307-424)	SQGLRILG <mark>Y</mark> TLKS	CASELGFLLFSLTMAII	I I <mark>F A T V M F Y A E KG S S A S K F</mark>	TS I PAA <mark>FWY</mark> T I VTMTTLG	YGDMVPKTIAGKI <mark>F</mark> GSICSLSGVLVI	ALPVPVICSN	F S R I Y HQ NQ R A D K R R -
Kv4.2	(7UKG)	(307-424)	SQGLRILGYTLKS	CASELGFLLFSLTMAI	I I FATVMFYAEKGSSASKF	TS I PAAEWYT I VTMTTLG	YGDMVPKTIAGKIFGSICSLSGVLVI	ALPVPVIVSN	FSRIYHQNQRADKRR.
Kv7.1	(6022)	(244-359)	GGTWRLLGSVVFI	HRQELITTLYIGELGLI	FSSYEVYLAEKDAVRVE	GSYADALWWGVVTVTTIG	YGDKVPQTWVGKTIASCESVEAISFE	ALPAGILGSG	FALKVQQ···K·····
Cav1.1	(68YO)	(180-344)	PS-LQVVLNSIFK	AMLPLEHIALLVLEMVI	I I YA I IGLELFKKMKPGP1	HFFGFSMLTVYQCITM-E	GWTDVLYNEWPWIYFVTLILLGSFFI	LNLVLGVLSG	EFTKERE
Cav1.1	(8000)	(100-344)	PS-LQVVLNSTPK	AMEPERHIALLVERMV	TYATIGLELFKKMKPGPT	HFFGFSMLTVYQCITM-E	GWTDVLYNEWPWIYFVTLILLGSFFI	LNLVLGVLSG	EFTKERE K
Cav1.1	(6/P8)	(180-344)	PSLOVVINCIE		TTATIGLELF KKMKPPTP	HEEGERMI TVYOCITM	GWTDVLTNNWPWIYFVTLILLSFFTL	NEVEGVESGE	ETVEREN.
Cav1.1	(6.IPA)	(180-344)	PS-LOVVINSIE K	AME PERHIALI VIEMVI	TATIOLEL FRAMESPET	HEEGESML TYYOCITM.E	OWTOVETNNWPWITFVTETEESFFTE	NIVIOVISOE	TVEREV
Cav11	(6.IPR)	(180-344)	PSLOVVINSIE	AMI PLENIALLYLEMY	I VALIGI I FROKMKSPPT	HEEGESMI TVYOCITM.E	OWTOVI YNEWPWLYEVTLILL SEE LL	NUVIOVISOE	FTKEREK
Cav2.3	(8EPL)	(205-395)	SL-QIVLKSIMKA	WV-PLLQIGLLLFFAIL	MEALIGLEEYSKLPGPTO	FDILFAVLTVFQCITM-E	GWTTVLYNWLYF I PLI I G SFFVLNLV	LOVLSGEFAK	ERERVENRRAF
Cav1.3	(7UHĠ)	(255-417)	PSLQVVLNSII-K	AMVPLLHIALLVLFVII	INALIGLELFIKMPOPT	FOFAFAML TVFQCITM-E	GWTDVLYNWVYFVSLVIG SFFVLNLV	LOVLSGEFSK	EREKAKARGD K
Cav1.3	(7UHF)	(255-417)	PSLQVVLNSIIKAI	WVPL - LHIALLVLFVII	II <mark>Y</mark> AIIGLL <mark>E</mark> IG <mark>KMK</mark> PPTN	F D F A F AML T V F Q C I T M - E	GWTDVLYNELPWVYEVSLVIESEEVL	NLVLGVLSGE	FSERKAKARGD . QK
Cav2.2	(7MIY)	(207-364)	PSLQVVLKSIM-K	AMVPLLQIGLLL <mark>FF</mark> AIL	MFAIIGLEFYMKFPGPTN	FDILFAILTVFQCITM-E	GWTDILYNNWNWLYFIPLIII SFFML	NLVLGVLSGE	FAKE-R
Cav3.3	(7WLI)	(193-413)	- PSMRILVNLLLD	TLPMLGNVLLLCFFVFF	IFGIIGVQLWALLF IPI	FD IGYAWIVIFQVITL - E	GWVEIMYMFIYFILLIIGSFFMINLC	LVVIATOFSE	TKOREHRL
Nav1.1	(7DTD)	(232-438)	GL - KT IVGALIQS	VK - KLSDVMILTVFCLS	V FALIGLL FNLRNKANP 1	ISDF SWAFLSLFRLMTQDF	WENLYQLLKTYMIFFVLVIFLSFYLI	NLILAVVAMA	YEEQNQATLEEA-E
Nav1.2	(6J8E)	(232-441)	POLKTIVGALIQ	SVKKLSDVMILTVFCLS	VFALIGLLFMGNNKANP1	SDF SWAFLSLFRLMTQDF	WENLYQLLKTYMIFFVLVIFOSFYLI	NLILAVVAMA	YEEQNOATLEEA - E
Nav1.4	(6AGF)	(234-463)	POLKTIVGALIOS	VKKL - SDVMILTVFCLS	SVFALVGLLFMGNQKLNP1	SDESWAFLALFRLMTODY	WENLFOLLKTYMIFFVVIIFGSFYLI	NLILAVVAMA	YAEQNEATLAED . K
Nav1.5	(0020)	(230-437) (225-420)	GL . KTIVGALI . Q	SVERLADVMVLTVFCLS	SVFALIGLLENL RHKVNPI	SD SAWAFLALFRLMTODC	WERLYGOLKTYMIFFMLVIFGSFYLV	NLILAVVAMA	YEEQNOAT TAETEE
Nav1.5	(61 0 4)	(236 434)	SOLKTIVOAL LOS		VEAL IGLIEN DHEVER	SO SAWAFLALF RUMTODO	WERLYGOLKIYMIFFMLVIFOSFTLV		VEEQNOAT LAET
Nav1.6	(BEHD)	(236-443)	GL KTIVGAL LOS	VKKL SDVMIL TVECIS	VEAL IGLLEMONNEL NP	SOTSWAFLALERIMTODY	WENLYOLLKTYMLEEVLVLEVSEYLV	NULLAVVAMA	YEEONOATI EEAEKEE
Nav1 7	(7W9K)	(229-407)	POLKTIVGALLO	SVKKLSDVMILTVFCLS	VEALIGLEEMONHKANPI	SDTSWAFLALFRLMTODY	WENLYOOLKTYMIEEVVV IELSEYLI	NLILAVVAMA	YE
Nov1.8	(7M/EM/)	(232-407)	L. KVIVGALL. H	NUMBER ADVITER TIPOL					and the second se
14441.0	(,,)	(=-=)		SVARCADVIICII	VFALVOLL KONNKLNP	FDSAWAFLSLFRLMIUDS	WERLYQQLKIYMIFFVLVIFLSFYLV	NLILAVVTMA	YEEQNQA
Kvs and [	DII from C	av and Nav channels	L316	L329	ALVOLL KONNKLAPS	I	WERLYQOLKIYMIFFVLVIFLSFYLV	L403	F412
Kvs and [	DII from C	(310-428)	L316		IFSSLVFFAEKDEDDTKF	KS I PAS FWWAT I TMTTVG	YOB IYPRTILOKIVGOLCCIAGVLV		F412
Kvs and [ Kv2.1 Kv1.2/2.1	OII from C	av and Nav channels (310-428) (307-417) (370-489)	L316	L329	IIFSSLVEFAEKDEDDTKF	KSIPAS PWAT ITATTVG	VGBIYPRTILÖKIVGÖLCCIÄGVLVI VGBIYPRTILÖKIVGÖLCCIÄGVLVI	L403	F412
Kvs and E Kv2.1 Kv1.2/2.1 Shaker	(16EBM) (75IP)	av and Nav channels (310-428) (307-417) (391-488) (381-400)	L316	L329	LFSSLVFFAEKDEDDTK LFSSLVFFAEKDEDDTK LFSSLVYFAEADERDSOF	KSIPAS FUWATITMITVG PSIPDAFUWAVVSMTTVG KSIPDAFUWAVVSMTTVG	VODIYPKTILGKIVGOLOCIAGVLVI Vodiypktilgkivgolociagvlti Vodiyptigokivgolociagvlti	L403	F412 FSEFYKEOKROEKAIK FNYFYHRE
Kvs and E Kv1.2/2.1 Shaker Kv1.3 Kv1.3	(1500 C) (1500 C) (15	av and Nav channels (310-428) (307-417) (381-490) (381-490)	L316 STOLOSLOE TLERS SKOLQILGOTLERAS SKOLQILGOTLERAS	L329	IFSSLVFFAEKDEDDTK LFSSLVFFAEKDEDDTK LFSSLVFAEADERDSOF LFSSLVFAEADERDSOF LFSSLVFAEADPTSG	KSIPAS WATITMTTVG PSIPDAF WAVYSMTTVG KSIPDAF WAVYSMTTVG SSIPDAF WAVYMTTVG	VGBIYPRTILGRIVGGLCCIAGVLV VGBIYPRTILGRIVGGLCCIAGVLV VGBYPRTILGRIVGSLCAIAGVLV VGBYPRTILGRIVGSLCAIAGVLT VGBYPRTIGGNIGSLCAIAGVLT	L403 ALPIPLIVNN ALPVPVIVSN ALPVPVIVSN ALPVPVIVSN	
Kvs and E Kv2.1 Kv1.2/2.1 Shaker Kv1.3 Kv1.3 Kv1.3	(6EBM) (7SIP) (7SSV) (7SSX) (7SSX) (7SSY)	av and Nav channels (307-417) (307-417) (381-480) (381-490) (381-490) (381-490) (381-90)	L316	L329	IFSSLVFFAEKÖEDDTK FSSLVFFAEKÖEDDTK FSSLVFFAEADERDSOF LFSSLVFAEADERDSOF LFSSLVFAEADERDSOF LFSSLVFAEADERDSOF LFSSLVFAEADERTSOF	KSIPASFWATITMTTVO PSIPDAFWAVTMTTVO KSIPDAFWAVVSMTTVO KSIPDAFWAVVTMTTVO SSIPDAFWAVVTMTTVO SSIPDAFWAVVTMTTVO	VOBINETIOKIVOLOTIASVLV VOBINETIOKIVOSLOTIASVLV VODMETOSKIVOSLOTIASVLV VODMETOSKIVOSLOTIASVLT VODMETOSKIVOSLOTIASVLT	L403 L403 ALPIPLIVNN ALPVPVIVSN ALPVPVIVSN ALPVPVIVSN ALPVPVIVSN ALPVPVIVSN	
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#### Supplementary Information Figure 3 | Structure-based sequence alignment of Kv, Cav and Nav channels

Structure-based sequence alignment of residues from the S4-S5 linker to the end of S6 with residues colored based on side chain character. Residues identified within the hydrophobic coupling nexus are highlighted as well as other regions of interest (IFM motif).



Supplementary Information Figure 4 Uncropped gel from Extended Data Fig. 1d