Case presentations from diagnostic exome sequencing results in ion channels

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DASNE meeting 2017, Eisenach

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CASE 1: Clinical picture

- *Patient's presentation*:
  - 38 year-old jewish male with slowly progressive epilepsy and mental decline
  - Late onset (14 years) of generalized seizures, no myoclonus
  - Gradual mental decline until dementia
- Reminiscent of a specific syndrome? Next diagnostic steps?
- Imaging and EEG without any specific changes
- CSF not performed
- Family history negative
- Genetics? Which genes? Patient only? Parents?

CASE 1: Genetics

- Exome Sequencing plus parent testing: ***de novo SCN10A p.*R1582H** mutation
- Gene? Suspicion about a localization?
- *SCN10A encodes the voltage-gated Na^+ channel $Na_V1.8$:*
  - Expressed in the dorsal root ganglion and involved in pain sensation
  - Linked to "AD Familial episodic pain"
  - Mutation is located in the highly conserved voltage sensor (transmembrane segment S4) of the fourth transmembrane domain (D4)
  - Next questions?

Frequency

ExAC/gnomAD:

Not reported.

dbSNP:

2 submissions (rs759164988), both heterozygous

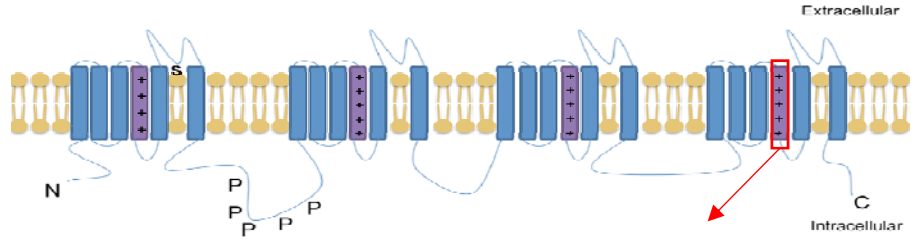
In-Silico pathogenicity prediction

CADD score: 28.3

Damaging predictions:

PolyPhen2, SIFT, MutationTaster, MutationAssessor, Condel, Provean, FATHMM-MLK, LRT

Case 1: Functional consequence / Interpretation - 1

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Nav1.1 (SCN1A)    LFRVIRLARIGRILRLIKGAKGIRTLLFAL
Nav1.2 (SCN2A)    LFRVIRLARIGRILRLIKGAKGIRTLLFAL
Nav1.3 (SCN3A)    LFRVIRLARIGRILRLIKGAKGIRTLLFAL
Nav1.4 (SCN4A)    LFRVIRLARIGRVLRLIKGAKGIRTLLFAL
Nav1.5 (SCN5A)    LFRVIRLARIGRILRLIRGAKGIRTLLFAL
Nav1.6 (SCN8A)    LFRVIRLARIGRILRLIKGAKGIRTLLFAL
Nav1.7 (SCN9A)    LFRVIRLARIGRILRLVKGAKGIRTLLFAL
Nav1.8 (SCN10A)   LFRVIRLARIGRILRLIRAAKGIRTLLFAL
Nav1.9 (SCN11A)   LFRIVRLARIGRILRLVRAARGIRTLLFAL
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- R1582H affects a highly conserved Arginine residue in the D4/S4 voltage sensor

ExAC/gnomAD		Allele count
SCN10A	**R1582C**	3
SCN1A	R1645Q	1
SCN4A	R1457H	1
SCN4A	R1457C	1
SCN5A	R1632H	1
SCN5A	R1632C	1
SCN9A	R1608Q	1
SCN9A	R1608*	1
SCN11A	R1472Q	1
SCN11A	R1472*	21

ClinVar

(likely)pathogenic

SCN1A	R1645Q	germline & de novo
SCN1A	R1645P	de novo
SCN1A	R1645*	germline
SCN2A	R1635Q	germline
SCN2A	R1635*	germline
SCN4A	R1457H	germline (homozygous)

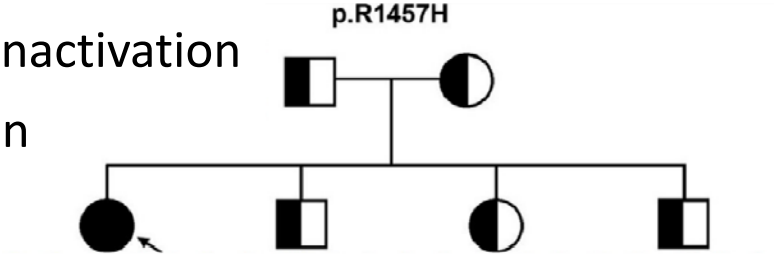
Uncertain significance

SCN5A	R1632H	germline
SCN5A	R1632C	germline
SCN8A	R1626H	germline

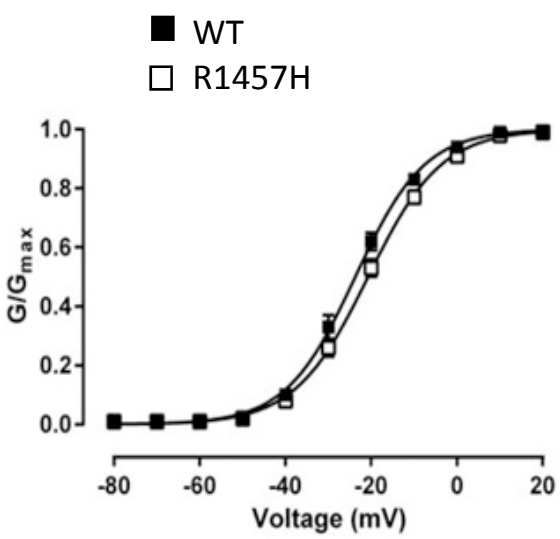
Case 1: Functional consequence / Interpretation - 2

- The corresponding mutation in *SCN4A* causes autosomal recessive congenital myasthenia syndrome
- Subtle depolarizing shift in steady-state activation
- Large hyperpolarizing shift in steady-state fast inactivation
- Strong slowing of recovery from fast inactivation

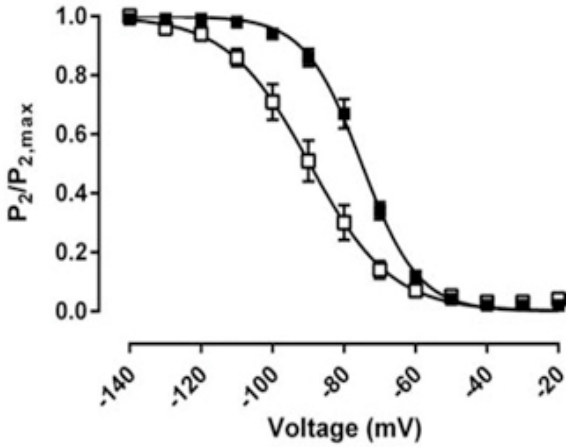
→ Loss of function!



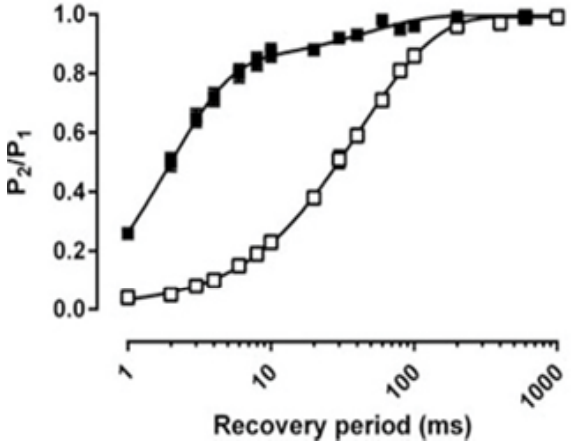
Arnold *et al*. *Ann Neurol. 2015*



Voltage dependence of activation



Steady-state fast inactivation



Recovery from fast inactivation

Summary of case 1: Pathogenicity of the mutations in D4/S4 4th Arginine

- Pathogenic only in homozygous state in
  - *SCN4A* (Loss of Function)
- possibly pathogenic in heterozygous state in
  - *SCN1A*, *SCN2A*, *SCN3A*, *SCN5A*, *SCN8A* (depending on the mutation)
  - likely with reduced penetrance
- **Most probably not pathogenic** in heterozygous state in *SCN10A* & *SCN4A*

Genic intolerance scores

Scores based on ExAC data

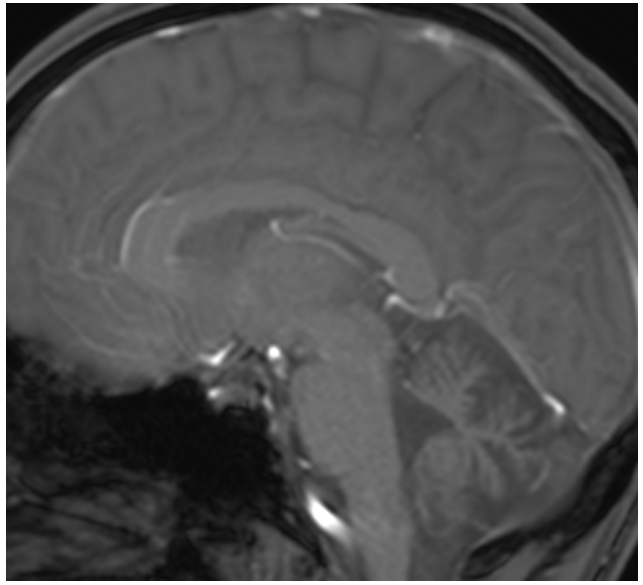
- Residual Variation Intolerance Score (RVIS)
- Intolerance to heterozygous LOF (pLI)
- intolerance to homozygous LOF (pRecessive)

Gene	RVIS %	pLI	pReccessive
SCN8A	0.7%	1	0
SCN3A	1.4%	1	0
SCN2A	1.6%	1	0
SCN1A	2.4%	1	0
SCN5A	21%	1	0
SCN11A	2.4%	0	0.9
SCN4A	53%	0	1
SCN9A	61%	0	1
SCN10A	91%	0	0

CASE 2: Clinical presentation

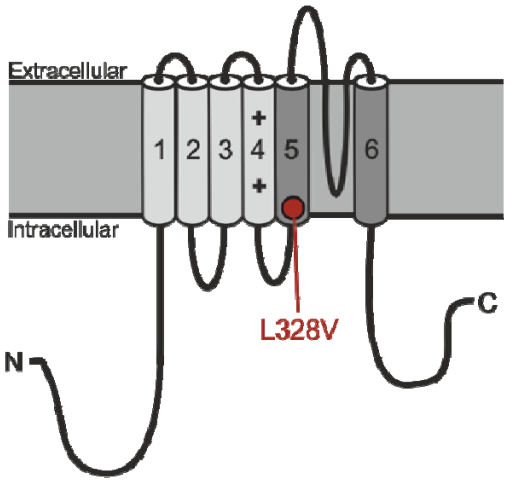
- *Patient's presentation*:
  - 8 year old German male with daily absence seizures, atonic seizures and GTCS
  - Onset at 6 months of age with febrile status epilepticus followed by developmental delay (psychomotor, language) at 1 year.
  - Hyperactivity, severe intellectual disability and ataxia (cerebellar atrophy)
  - EEG: GSW and multifocal SW, sleep activation (SWI < 50%).
  - Medications: lacosamide, lamotrigine, bromide.

- What next?



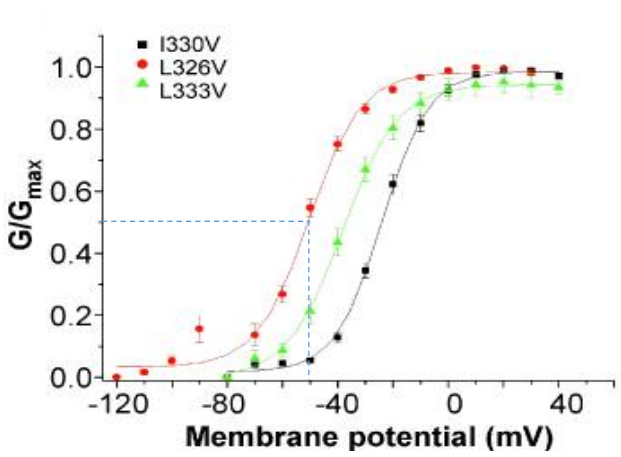
CASE 2: Genetics

- Exome sequencing:
  - ***De novo KCNA2* p.L328V** mutation.
  - In the pore region (S5)
  - Idea how to assess functional effect?



K_V1.1-channel Upadhyay *et al*. *J Physiol.* 2009

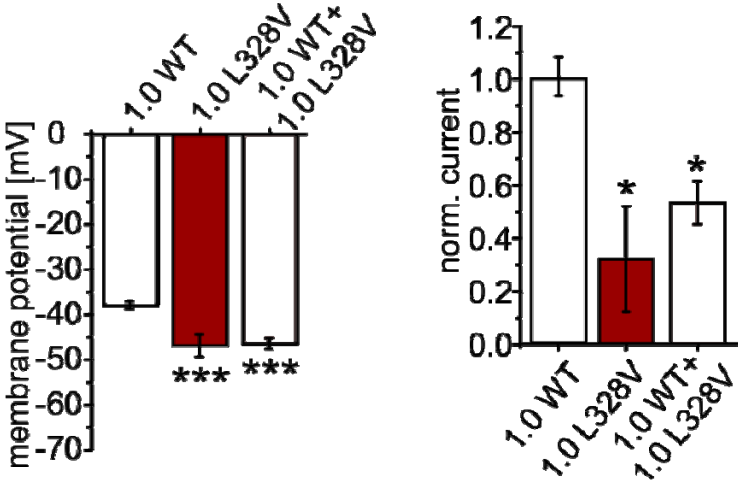
K_V1.1 L326V

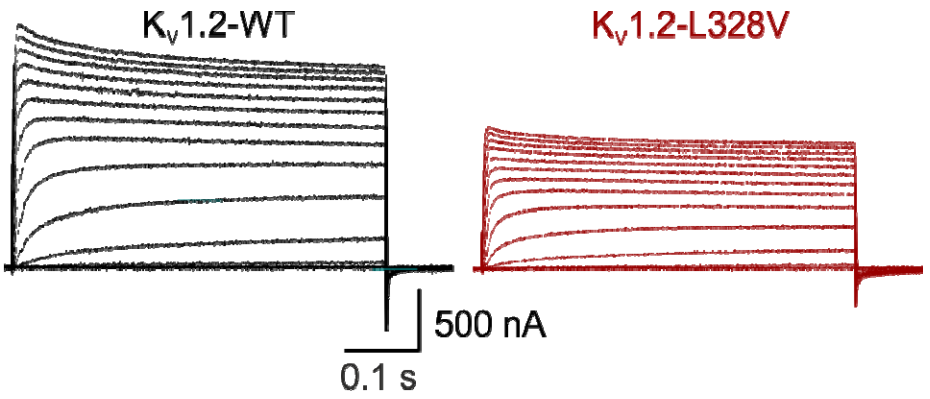


L328V

Homo sapiens	SMRELGLLI
K_v1.1 *H. sapiens*	SMRELGLLI
Rattus norvegicus	SMRELGLLI
Mus musculus	SMRELGLLI
Bos taurus	SMRELGLLI
Xenopus laevis	SMRELGLLI
D. melanogaster	SMRELGLLI

KCNA2 L328V & *KCNA1* L326V

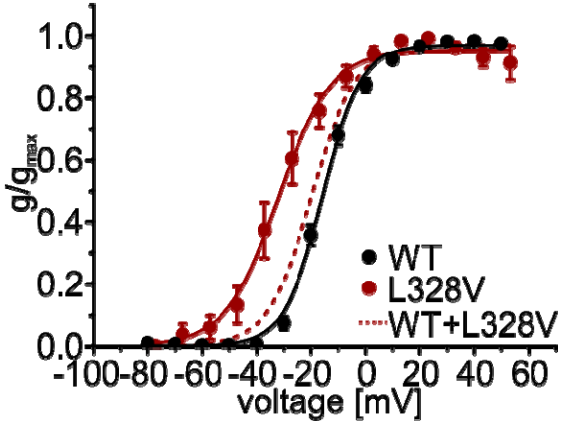




K_V1.2-channel

Masnada *et al. Brain* 2017

K_V1.2 L328V



K_V1.1-channel Upadhyay *et al. J Physiol.* 2009

K_V1.1 L326V