

## Supplementary Materials for

### Casein Kinase I $\delta$ Mutations in Familial Migraine and Advanced Sleep Phase

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Published 1 May 2013, *Sci. Transl. Med.* **5**, 183ra56 (2013)  
DOI: 10.1126/scitranslmed.3005784

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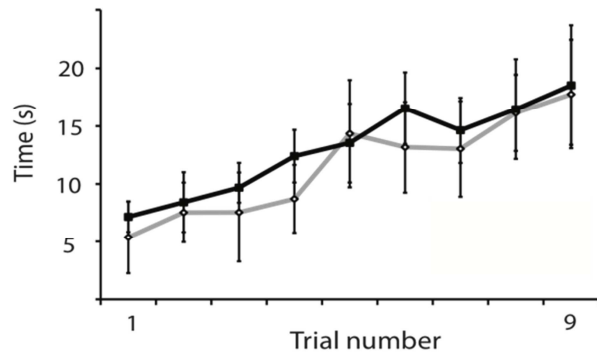
Supplementary Figures S1-S5; Supplementary Tables S1-3.

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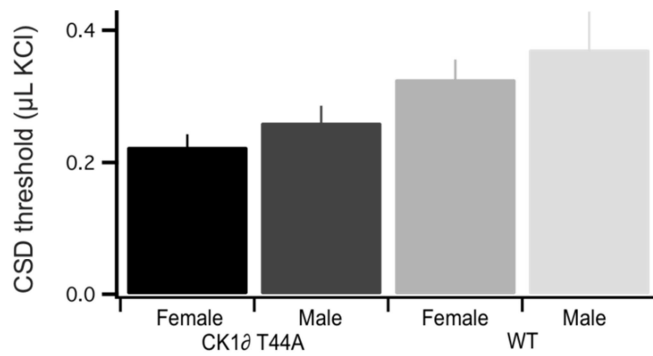
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                K5579
                A R
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DBT     GEEVAIKLECIRTKHPQLHIESKFKYKTMQ
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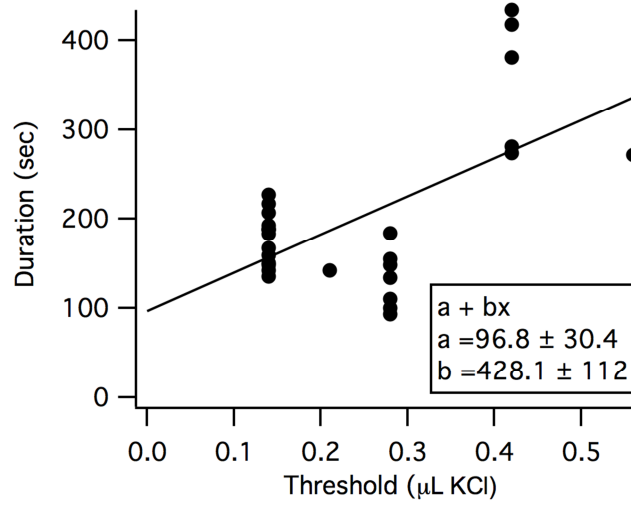
**Fig. S1.** Alignments for *Drosophila* Dbt and mouse (m) and human (h) CKI $\delta$  and CKI $\epsilon$  proteins. The T44A and H46R mutation sites are highlighted.



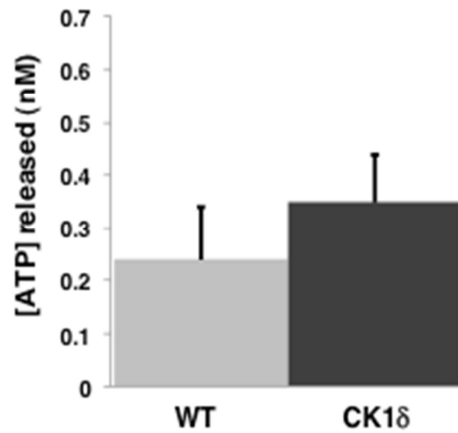
**Fig. S2.** Motor function in wild-type and CKIδ-T44A mice. Motor function was measured in CKIδ-T44A mice and wild-type littermates on Rotarod testing



**Fig. S3.** Gradient of CSD thresholds by sex and genotype. There was an increase in CSD threshold from female CKI $\delta$ -T44A to male CKI $\delta$ -T44A to female WT to male WT animals, which was significant overall, but individually only female CKI $\delta$ -T44A and male wild-type differences were significant on post-hoc testing (see text).



**Fig. S4.** Scatterplot of CSD dc shift duration and threshold in all animals. There is a positive correlation between volume of KCl ejected and duration of CSD. See text for details.



**Fig S5.** Astrocyte ATP release evoked by low divalent cation solution. Average concentration of ATP released in response to exposure of astrocytes to medium with no added  $\text{Ca}^{2+}$  or  $\text{Mg}^{2+}$  for 1 minute (error bars, SEM). There was no significant difference in the amount ATP released by astrocytes from CK1δ T44A mice vs. wild-type mice under these conditions.

**Table S1:** Mechanical sensitivity thresholds. Average difference in mechanical response (in grams) in wild-type and CKI $\delta$ -T44A **mice** at each time point. Estimates from the mixed model are presented with *P* values.

Dose	Baseline		60 Minutes		120 Minutes		240 Minutes	
	WT-CK1 $\delta$	<i>p</i>	WT-CK1 $\delta$	<i>p</i>	WT-CK1 $\delta$	<i>p</i>	WT-CK1 $\delta$	<i>p</i>
Saline	0.03	0.74	-0.01	0.94	0.20	0.05	0.05	0.68
1 mg/kg NTG	0.10	0.36	0.17	0.06	0.32	<0.01	0.13	0.14
3 mg/kg NTG	0.13	0.15	0.15	0.03	0.22	<0.01	0.20	0.02
5 mg/kg NTG	0.03	0.84	0.34	<0.01	0.51	<0.01	0.35	<0.01
7 mg/kg NTG	0.08	0.39	0.07	0.18	0.05	0.28	0.09	0.06
10 mg/kg NTG	-0.05	0.48	0.08	0.01	0.02	0.34	0.07	0.08

**Table S2:** Thermal responses. Average difference in thermal response (in seconds) comparing wild-type and CKI $\delta$ -T44A mice.

Dose	Baseline		30 Minutes		60 Minutes		90 Minutes		120 Minutes		240 Minutes	
	WT-CKI $\delta$	<i>p</i>	WT-CKI $\delta$	<i>p</i>	WT-CKI $\delta$	<i>p</i>	WT-CKI $\delta$	<i>p</i>	WT-CKI $\delta$	<i>p</i>	WT-CKI $\delta$	<i>p</i>
Saline	0.75	0.17	0.05	0.93	0.81	0.13	0.97	0.06	-0.08	0.89	1.28	0.02
1 mg/kg NTG	-0.40	0.66	0.38	0.64	-0.61	0.48	-1.07	0.21	0.69	0.42	0.63	0.47
3 mg/kg NTG	0.46	0.58	1.24	0.07	1.27	0.07	2.09	0.01	3.39	<0.01	2.84	<0.01
5 mg/kg NTG	-0.27	0.74	0.91	0.21	1.23	0.11	1.42	0.07	0.42	0.59	0.29	0.73
7 mg/kg NTG	0.91	0.34	-0.31	0.71	1.89	0.02	0.21	0.81	-0.34	0.70	1.17	0.21
10 mg/kg NTG	0.48	0.57	1.26	0.07	1.28	0.07	2.11	0.01	3.41	<0.01	2.86	<0.01



**Table S3:** CSD-associated measures in CKI $\delta$ -T44A mice and wild-type littermates.

	Measure (mean +/- SEM)	<b>CKI<math>\delta</math>- T44A</b>	N (CSD; animals)	<b>WT</b>	N (CSD; animals)	p value
<b>OIS</b>	<b>Duration</b> (sec.)	311+/-20	29; 9	433+/-35	17; 6	0.003
	<b>AUC</b> (a.u.)	61.3+/-16	29; 9	61.6+/-21	17; 6	0.49
<b>FP</b>	<b>Duration</b> (sec.)	166+/-9	16; 6	253+/-32	12; 5	0.01
	<b>Amplitude</b> (mV)	-12.1+/-0.6	16; 6	-12.8+/-0.9	12; 5	0.24

OIS: optical intrinsic signal; FP: field potential; AUC: area under the curve. P values:

Student's t-test.