

A Multiplex Human Syndrome Implicates a Key Role

for Intestinal Cell Kinase in Development

of Central Nervous, Skeletal, and Endocrine Systems

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Table S1. Variations found in directly sequenced genes.

Gene symbol	Gene name	No. of exons	Variation site	dbSNP ID	AA/splice site change ^{a,b}	Notes ^c
<i>IL17F</i>	interleukin 17F	3	E3, c.228A>G	rs11465553	CAT-H; CGT-R; H161R; probably damaging	U:AA, A:AA, C:AA, Con:GG
<i>LOC647163</i>	similar to intestinal mucin-2 precursor	2	IVS2, +94A>G E2, c.15C>A	rs875142 n/a	n/a CGC-R; AGC-S; R61S; this change is benign (Polyphen) **	U:GG, A:GG, C:GG, Con:AA U:TT, A:TT, C:TT, Con:GG
<i>EFHC1</i>	EF-hand domain (C') containing 1	11	E3, c.190C>T E3, c.260G>A	n/a n/a	CGG-R; TGG-W; R159W; this variant is possibly damaging CGC-R; CAC-H; R182H; this is a benign change (Polyphen)**	U:CC, A:CC, C:CT, Con:CC U:GG, A:GG, C:GG, Con:GA
			IVS5, +166C>T	rs614570	no change to splice site	U:CC, A:CC, C:CT, Con:CC
			IVS5, -118G>T	n/a	T-616% increased binding of spliceosome at acceptor site	U:TT, A:TT, C: GT, Con:GG
			IVS10, +58T>C	rs2273120	no change to splice site	U:CC, A:CC, C:TC, Con:CC
<i>TMEM14A</i>	transmembrane protein 14A	5	NOTHING FOUND			
<i>GSTA3</i>	glutathione S-transferase A3	7	NOTHING FOUND			
<i>GSTA4</i>	glutathione S-transferase A4	7	IVS5, -48C>G IVS6, -87T>C IVS7, +137C>A	rs316133 rs375887 rs367836	no change to splice site no change to splice site no change to splice site	U:GG, A:GG, C:GG, Con:CC U:CC, A:TT, C:CC, Con:TT U:AA, A:AA, C:AA, Con:CA
<i>ICK</i>	intestinal cell (MAK-like) kinase	13	E7, c.815G>A	n/a	CGA-R; CAA-Q; R272Q; change is probably damaging	U:AG, A:AA, C:AG, Con:GG
<i>FBXO9</i>	F-box protein 9	13	IVS1, -227C>T IVS5, +61C>T IVS10, +120A>G	n/a rs17616311 n/a	no change to splice site T-16% increased binding by spliceosome at donor site no change to splice site	U:CT, A:CC, C:CC, Con:CC U:CT, A:CC, C:CC, Con:CT U:GG, A:GG, C:GG, Con:GG
<i>GCM1</i>	glial cells missing homolog 1	6	NOTHING FOUND			
<i>ELOVL5</i>	elongation of long chain fatty acids	8	IVS4, +49A>T	n/a	no change to splice site	U:AT, A:AA, C:AT, Con:TT

GCLC	glutamate-cysteine ligase, catalytic subunit	16	IVS3, +101G>A	n/a	A-4% increased binding by spliceosome at acceptor site	U:AG, A:GG, C:GG, Con:AG
			IVS4, +198G>A	n/a	no effect	U:AA, A:AA, C:AA, Con:AA
			IVS4, -110T>G	n/a	G-676% increased binding of spliceosome at acceptor site	U:GG, A:GG, C:GG, Con:AG
			IVS4, -47G>A	n/a	A-50% decrease in spliceosome binding at acceptor site	U:GG, A:GG, C:GG, Con:AG
			IVS11, -101G>A	rs17886117	no change to splice site	U: AG, A:AA, C:AG, Con: AG
			IVS12, +83A>G	n/a	no change to splice site	U:AG, A:AA, C:GG, Con:AG
			IVS12, -63G>A	rs2066511	A-31% increased binding to splicesosome at the donor site	U:GG, A:GG, C:GG, Con:GA

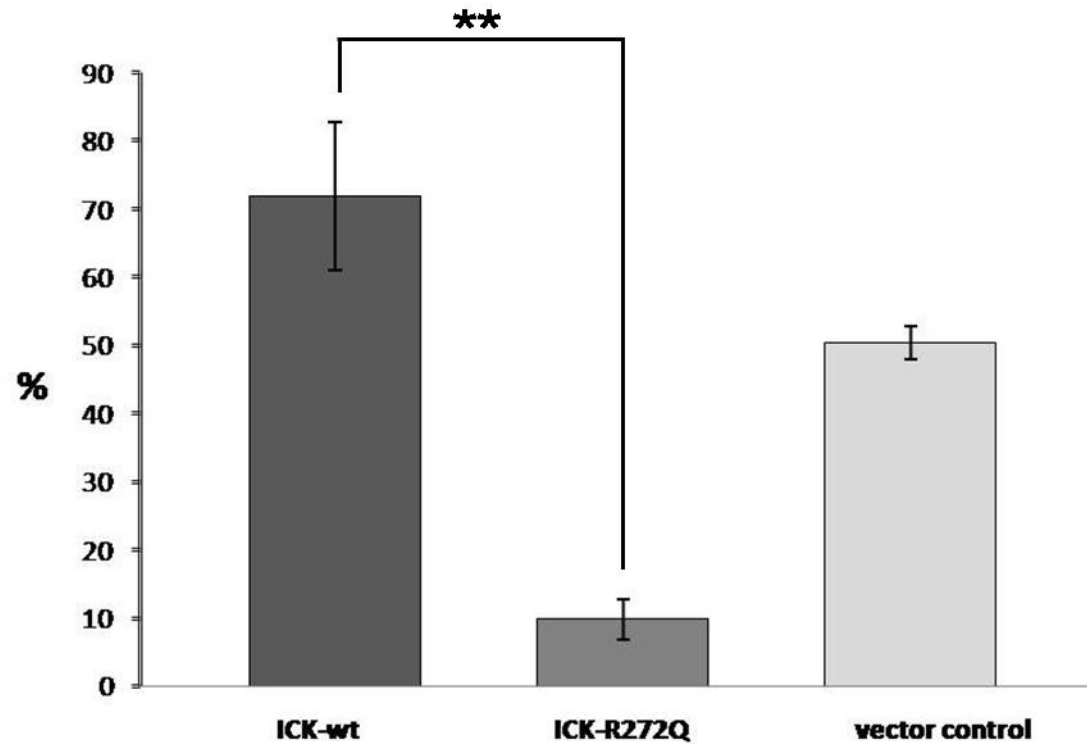
^a Effect of amino acid changes were determined by "Polyphen: prediction of functional effect of human nsSNPs" website (<http://genetics.bwh.harvard.edu/pph/>) from the Bork and Sunyaev labs ²⁸

^b Splice site changes were predicted by the "Automated Splice Site Analyses" website (<https://splice.uwo.ca/>) from the University of Western Ontario ²²

^c U, unaffected sibling; C, obligate carrier; A, affected; Con, control outside Amish population

^d There was no effect on splice site

Figure S1. Bar graph of the percentage (%) of cells with nuclear localization of the three different ICK constructs (wildtype ICK, R272Q mutant ICK, and vector control)



Bar graph of the percentage (%) of cells with nuclear localization of the three different ICK constructs (wildtype ICK, R272Q mutant ICK, and vector control). As demonstrated in the graph, the wildtype ICK and the R272Q mutant ICK protein localizes in the nucleus of 71.9% and 9.78% of the transfected cells, respectively. In total, 502, 501, and 501 transfected cells were counted per construct of wildtype ICK, R272Q mutant ICK, and control, respectively. Bar graph represents cells with nuclear localization \pm standard deviation (in %). **OR (95% CI) = 23.62 (16.59, 33.62); chi-square p-value = 3.9×10^{-97} .