Genetic Screen for and Molecular Characterization of Novel Suppressors of Mutations Affecting the SUP-9/SUP-10/UNC-93 Two-Pore Domain K^+ Channel Complex

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sup-9, sup-10 and *unc-93* encode components of a presumptive *C. elegans* two-pore domain K^+ channel complex. Rare gain-of-function mutations of each of these three genes cause abnormal body muscle contraction and are thought to activate the SUP-9 K^+ channel. The mutant animals are defective in egg laying, sluggish and exhibit the "rubberband" phenotype: when prodded on the head, an animal contracts and relaxes along its entire body without moving backwards. The SUP-9 protein is similar to the mammalian Two-pore Acid Sensitive K^+ channels TASK-1 and TASK-3. *sup-10* encodes a novel single transmembrane protein. *unc-93* encodes a multiple transmembrane protein that defines a novel family of proteins conserved from *C. elegans* to mammals.

To seek essential genes that interact with *unc-93*, we screened ~10,000 EMS-mutagenized F1 *unc-93(e1500sd)* animals clonally for progeny with improved locomotion and identified four new suppressors. *n4562* and *n4563* are partial suppressors and cause sterility or carry closely linked mutations that cause sterility. *n4564* and *n4588* are strong recessive suppressors and cause temperature-sensitive (ts) lethality or carry closely linked ts-lethal mutations. We mapped *n4564, n4588* and *n4562* to small regions of LGI, LGIII and LGIV, respectively. We identified a missense mutation in the *let-418* gene, which encodes a *C. elegans* homolog of the chromatin-remodeling factor Mi-2, in *n4563* mutant animals. *let-418* might regulate the activity of the SUP-9/UNC-93/SUP-10 channel complex by affecting the transcription of the channel proteins or regulatory factors.

We also identified *pyp-1*, which encodes a *C. elegans* ortholog of inorganic pyrophosphatase, as a suppressor of *unc-93(e1500sd)* by screening ~1170 RNAi clones reported to cause sterility or lethality from a whole-genome RNAi library[1]. A deletion mutant of *pyp-1, n4599*, arrests at an early larval stage and is suppressed for the locomotory defect of *unc-93(e1500sd)*. A transgene expressing *pyp-1* in body-wall muscles and other cells rescues the suppressor activity of *pyp-1(n4599)*. PYP-1 might function as a subunit of the SUP-9 channel complex or might regulate the channel activity by affecting the expression of the channel proteins or regulatory factors.

[1]Kamath et al. (2003) Nature 421: 231-237.

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