Characterization of a Novel Conserved Neuronal Protein Possibly Involved in Synaptic Vesicle Exocytosis

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Upon entering a bacterial lawn, well-fed wild-type *C. elegans* reduce their locomotion rate by about 30 percent. This response, known as the basal slowing response, requires dopaminergic modulation of the locomotory circuit. Worms that have been starved briefly prior to crawling onto a bacterial lawn slow by more than 75 percent. This behavior, dependent on the past feeding experience of the animal, is termed the enhanced slowing response and is modulated by serotonin. Animals lacking *mod-5*, a serotonin reuptake transporter, display a hyper-enhanced slowing response upon reaching a bacterial lawn, slowing by more than 90 percent.

By mutagenizing *mod-5* animals, we identified mutations that cause well-fed worms to behave as if they had been deprived of food. These mutations might affect food-sensing or satiety mechanisms and make the enhanced slowing response independent of past feeding experience. We cloned a gene defined by one such mutation, *n4022*. This gene encodes a novel and highly conserved protein, C44B9.1. A rescuing *C44B9.1* translational GFP reporter that contains the endogenous *C44B9.1* promoter is expressed in the HSNs, multiple head and tail neurons and the ventral nerve cord. *C44B9.1* expression is observed mainly in cell bodies but also in processes as puncta. The locomotion defects of *n4022* worms are rescued by expressing *C44B9.1* from a pan-neuronal promoter (*unc-119*) but not from a body-wall muscle promoter (*myo-3*), indicating a neuronal function for *C44B9.1*.

The *n4022* strain shows decreased sensitivity to aldicarb, an acetylcholinesterase inhibitor, and increased sensitivity to levamisole, an acetylcholine receptor agonist, suggesting a presynaptic role for *C44B9.1*. In addition to their locomotion defect, *n4022* mutants retain eggs *in utero* for an abnormally long period of time, which leads to eggs being laid at a later developmental stage. Furthermore, *C44B9.1* mutants display high temperature (27°C) induced dauer formation (Hid) and synthetic dauer formation at 25°C in combination with *unc-31(e928)* (Sdf). This phenotype and the drug sensitivity profile are similar to those of strains mutant in genes involved in the regulation of synaptic vesicle exocytosis, such as *unc-64*, *unc-31*, *hid-1* and *aex-3*.

We are currently performing suppressor screens to place *C44B9.1* in a genetic pathway and are investigating the genetic interactions of *C44B9.1* with genes involved in neurotransmission. We plan to use a genetically-encoded calcium indicator and electrophysiology to assess the role of *C44B9.1* in synaptic vesicle exocytosis and cell excitability.

Poster
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