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

Allan Froehlich, Daniel Omura, Nicolas Paquin, Bob Horvitz
HHMI, Dept. Biology, MIT, Cambridge, (MA), USA

Upon entering a bacterial lawn, well-fed wild-type animals slow 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 for 30 minutes prior to entering a bacterial lawn slow their locomotion by more than 75 percent. This behavior, dependent on the past feeding experience of the animal, is termed the enhanced slowing response and requires serotonin. Animals deleted for *mod-5*, a serotonin reuptake transporter, that have been briefly starved display a hyper-enhanced slowing response upon reaching a bacterial lawn, slowing their locomotion by more than 90 percent.

By mutagenizing *mod-5* animals, we identified mutations that enhance the locomotion defect such that well fed worms behave as if they had been deprived of food. These mutations may affect food-sensing or satiety mechanisms and make the enhanced slowing response independent from past feeding experiences. We have cloned a gene defined by the mutant *n4022* that encodes a novel and highly conserved protein, C44B9.1. 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*. Expression of a GFP reporter under a *C44B9.1* promoter was observed in multiple head neurons.

C44B9.1 is likely involved in synaptic vesicle release, as 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 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 is similar to that of strains mutated for genes involved in the regulation of vesicle exocytosis, such as *unc-64*, *unc-31*, *hid-1* and *aex-3*.

We plan to identify the sites of expression of *C44B9.1* by immunofluorescence and the binding partners of the C44B9.1 protein to further understand this gene. We will also use electrophysiology to assess the role of C44B9.1 in vesicle exocytosis.

Contact: nicolas.paquin@umontreal.ca
Lab: Horvitz