22. Genes Involved In Serotonergic Neurotransmission

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Wild-type animals that have been acutely food deprived slow their locomotory rate upon encountering bacteria more than do well-fed animals. This behavior, called the enhanced slowing response, is serotonin (5-HT) dependent. Animals mutant for the 5-HT reuptake transporter *mod-5* slow even more than wild-type animals because endogenous serotonin activity is potentiated. We call this behavior the hyperenhanced slowing response. *mod-5* animals are hypersensitive to immobilization by exogenous 5-HT. To identify additional genes involved in 5-HT signaling and possibly in the enhanced slowing response, we screened for suppressors of the 5-HT hypersensitivity of *mod-5* animals. We also used a candidate gene approach, testing for 5-HT resistance of strains containing deletions in genes that encode proteins similar to metabotropic serotonin receptors.

Using Mos1 transposon mutagenesis (Bessereau *et al*. *Nature*, 413: 70-74, 2001), we screened worms corresponding to 46,200 haploid genomes and identified three suppressors. Two contain insertions in genes that when mutated are known to suppress *mod-5* for both the exogenous 5-HT hypersensitivity and the hyperenhanced slowing response. One of these suppressors is an allele of *mod-1*, which encodes a 5-HT-gated chloride channel, and the other is an allele of *goa-1,* a predicted alpha subunit of a heterotrimeric G-protein. The third suppressor, *n4094*, partially suppresses the 5-HT hypersensitivity of *mod-5* animals but does not suppress the hyperenhanced slowing response of *mod-5* animals. *n4094* animals contain an insertion in a gene with similarity to bicarbonate transporters. A deletion allele of this gene phenocopies *n4094*. Experiments are currently underway to determine whether this deletion mutant displays other defects and to determine the expression pattern of this gene.

Using our candidate gene approach, we found a deletion, *ser-4(ok512),*that confers resistance to 5-HT and defects in the enhanced slowing response. *ser-4* likely encodes a metabotropic serotonin receptor (Olde and McCombie, *J. Mol. Neurosci*., 8:53-62, 1997). *ser-4(ok512)* suppressed both the 5-HT hypersensitivity and the hyperenhanced slowing response of *mod-5* animals. We will place *ser-4* in a genetic pathway with other genes known to function in the hyperenhanced slowing response, including *mod-1*, *goa-1*, and *dgk-1* (diacylglycerol kinase).

We also hope to define the neural circuit(s) through which *mod-5* and suppressors of *mod-5* act to affect the enhanced slowing response. Using antibodies raised against MOD-5,we have identified two pairs of head neurons in which MOD-5 is expressed: the NSMs and either the AIMs or the AIYs. Additionally, a translational *mod-1*::*rfp* reporter has been constructed and studied (by Eric Miska), and the expression pattern of SER-4 has been reported (Tsalik et al., *Dev. Biol*., 263(1): 81-102, 2003) using a translational GFP reporter. We will confirm these expression patterns by raising antibodies against these proteins. We will then express these genes in subsets of the neurons in which they are expressed to determine where these genes are required to function for a normal enhanced slowing response.