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Generation of cell-type-specific RNA and gene expression profiles for Mechanosensation Neurons in *C. elegans*

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One of the great advantages of *C. elegans* as an experimental system is the ability to characterize mutant phenotypes at the level of single type of cells. With the sequencing of the genome, the use of DNA microarrays offers the opportunity to look at the genome-wide transcriptional activity under different experimental conditions. Unfortunately, array experiments using whole worm RNA do not afford the resolution needed to look, for example, at the differences in expression between specific wild type and mutant nerve cells. We have developed a technique to generate cell-type-specific RNA to use with cDNA microarrays to study gene expression profiles for a single type of *C. elegans* cells and have applied it to study the differentiation and function of the embryonic touch receptor neurons (ALM and PLM). While saturation mutagenesis for touch insensitive mutants have identified several genes needed for touch cell development and function, those screens could not have identified redundant genes, pleiotropic genes or genes that mutate to a touch super-sensitive phenotype. DNA array data should reveal such genes as well as provide candidates for as yet uncloned *mec* genes.

To identify *mec-3*-dependent genes expressed in the touch cells, we collected late-stage embryos from strains with integrated GFP arrays: either P_{*mec-18*} *gfp* in a wild-type background for differentiated touch cells or P_{*mec-3*} *gfp* in a *mec-3* (*e1338*) background for *mec-3* mutant cells that should have become touch cells. Cells from dissociated embryos were cultured overnight (many extended processes in culture) and the GFP-positive cells were enriched 100-fold by fluorescence-activated cell sorting. Typical post-sort values from cell sorting are 4x10⁶ cells with 40-50% being GFP positive. mRNAs from these cells were linearly amplified 10⁶-fold using a modified Eberwine's method, labeled and applied to cDNA microarrays (in Stuart Kim's lab) representing virtually all of the *C. elegans* genes.

The reliability and sensitivity of the expression profiles is suggested by the finding that 6 of the 10 known *mec-3*-dependent genes were among the top 15 genes and 8 out of 10 known *mec-3*-dependent genes (including *mec-3* itself) had ratios above 4.0 (the two other genes had ratios higher than 2.0). In addition, the profile also provides several groups of interesting candidates besides unknown function genes with ratios equal to or higher than ratios of known *mec-3*-dependent genes: 1) the gene with the highest ratio

