Expression profiling of identified spinal neurons before and after cord injury (SCI)

D, Cui[1], K. Doughtery[2], M. Sawchuk[2], D. Machacek[2], S. Hochman[2], D.J. Baro[1]
[1]Biology, Georgia State University and [2]Physiology, Emory University, Atlanta GA.

To understand the complex properties of unique neuronal subpopulations and their response to SCI, expression profiling of identified neurons was performed by combining the technologies of fluorescent reporters, laser capture microdissection and DNA microarrays. Complete transections (T8-T10) were performed on postnatal day 24 and mice were sacrificed 3 weeks later (n=5). Fluorogold injections identified motor efferents. Sham operations were done on littermate controls. Three neuronal populations [lateral motoneurons (LMN) medial motoneurons (MMN) and intermediolateral horn neurons (IML)] were isolated from frozen sections of the spinal cord (T11-L4). For each animal we isolated 400 neurons per cell type and extracted RNA. The RNA was divided in half and processed separately to produce 2 targets for 2-4 Affymetrix oligonucleotide arrays. Preliminary analysis of these data (n=2 animals) suggest that: 1.GFAP, an astrocyte marker, is reduced by 93-100% in isolated populations relative to the entire spinal cord; thus, specific cell types are excluded. 2.Myelin basic protein and proteolipid protein are detected at significant levels suggesting unexpected expression in neurons, or perineural oligodentrocyte contamination. If contamination levels are equivalent, then differences between cell types reflect differences in neuronal gene expression. 3.Intermediate to low abundance transcripts expressed in MN and IML are detected in our assay [glutamate, $GABA_A$, dopamine D2, 5HT(2B, 1D, 5A, and 4) receptors]. 4.Differences between cells and their responses to SCI can be identified. Example: Preproenkephalin expression is highest in IML, but after SCI it increases to IML levels only in MMN. Future work will attempt to establish molecular fingerprints for identified cell types. Deviations from these fingerprints after SCI may illuminate untrodden paths toward functional recovery.

Supported by NIH NS38770 and NIH NS40440
Keywords: identified neuron, bioinformatics, preganglionic sympathetic, glia