Adrenal Chromaffin Cell Co-Grafts With NGF-Secreting Fibroblasts

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Autografts offer distinct advantages over neural grafts of non-self origin. While adrenal chromaffin cells (ACC) have been used extensively in grafts to the CNS, their potential for neuronal differentiation has been underutilized. ACC are mitotically active, multipotential neuronal progenitors that respond to instructive differentiation signals by adopting any of several neuronal phenotypes. For instance, these cells are potential sources for cholinergic as well as catecholaminergic neuronal grafts (Mahanthappa, Gage and Patterson, *Prog Brain Res* 1990; 82: 33-39).

One approach for enhancing ACC survival and neurite outgrowth in the CNS is to provide a source of nerve growth factor (NGF). We find that skin fibroblasts genetically engineered to secrete high levels of NGF (Kawaja and Gage, *Neuron* 1991; 7: 1019-1030) promote such neuronal differentiation by perinatal rat ACC when cogenerated into the striatum of adult rats. These ACC exhibit greatly enlarged soma size, extensive process outgrowth, and are positive for tyrosine hydroxylase, neurofilament (NF200), NGF receptor (p75), as well as the neuron-specific markers SCG10 and MAP2. The effect

of NGF secretion on graft volume and the total number of ACC, as defined by morphology with Nissl and tyrosine hydroxylase staining, was determined using unbiased stereology. The number of ACC surviving at 8 weeks was minimal without co-grafted fibroblasts, and ACC survival with the NGF-secreting fibroblasts was enhanced 2.5-fold over the co-grafting with control fibroblasts. The ability of control fibroblasts to enhance survival dramatically without inducing neuronal differentiation merits further investigation. The NGF transgene induces virtually all TH⁺ cells to express the neuronal markers SCG10 and MAP2. Neurite outgrowth by these cells was largely confined to within the graft, however. The survival rate (a mean of 42% of initial ACC grafted) and differentiation (virtually all TH⁺ cells) observed in this co-graft paradigm are the most significant yet reported for adrenal grafts.

The effects of these long-term grafts on the animals' behavior in Parkinson's disease paradigms are currently being tested.