Stem cell transplantation for stroke

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The past decade has seen impressive advances in the prevention and treatment of cerebrovascular disease. Nevertheless, stroke remains the leading cause of serious adult disability in the United States, as more than 4 million Americans are estimated to be stroke survivors. Physical therapy, occupational therapy, and speech therapy are the mainstay of rehabilitative efforts, but in many cases significant disabilities remain following stroke.

Several new therapies are under investigation to address the long-term disability of stroke survivors. Growth factors, amphetamines, cortical stimulation, and new approaches to physical therapy (eg, constraint-induced therapy) offer the possibility of improving neurologic deficits months or years after the recovery process has reached a plateau. Cell transplantation was pioneered for the treatment of Parkinson disease (PD) and has now been applied to other neurologic diseases, including stroke. However, treatment with transplanted cells is somewhat more complex for stroke than for PD. In PD, cellular therapy is aimed at replacing dopaminergic cells in the substantia nigra, whereas in stroke, multiple cell types and neurotransmitters are lost.

There is uncertainty about the mechanism by which cell transplantation might improve stroke deficits. Transplanted cells would ideally replace cells that are damaged by ischemia and take over the function of these cellular elements. However, it is also possible that transplanted cells secrete trophic factors that help to maintain marginally surviving cells or otherwise enhance the local environment sufficiently to improve function. Transplantation might also conceivably produce a host reaction that could include sprouting of new axons and synapse formation.

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POTENTIAL CELL SOURCES

A number of sources of transplanted cells are available.

Transplanted fetal stem cells survive and integrate into host brains in animal models of stroke. Functional improvement occurs as long as the cells remain immature. In human studies of PD, initial improvement in neurologic disability was mitigated by severe late dyskinesias. Whether similar problems might occur with stroke is unknown. Use of fetal stem cells is constricted by ethical concerns and limited availability.

Neuroprogenitor cells are found in the periventricular region of developing brains and in adults. After brain injury, including ischemia, these cells react by migrating to the area of injury and undergoing differentiation. When transplanted into brains of rats subjected to ischemia, these cells survive, differentiate, and proliferate. Because neuroprogenitor cells are derived from fetal brains, ethical issues similar to those with fetal stem cells limit availability.

Bone marrow stromal cells are capable of differentiating into multiple cell types, including neuronal cells. Transplantation of these cells into striatum of rats subjected to ischemia significantly improves function. Similar results have been achieved with intravenous infusion. Unfortunately, the yield of stromal cells from bone marrow is low and the safety of these cells is uncertain.

Multipotential cells have also been isolated from umbilical cord blood. This source of cells for transplantation is appealing because of ready availability and the lack of ethical issues. Functional improvement has been demonstrated in animal stroke models with implantation or intravenous injection similar to bone marrow stromal cells.

Immortalized cell lines offer a ready source of cells for transplantation without the ethical concerns that surround fetal tissues. One source of cells is the NTera 2/cl.D1 (NT2) human embryonic carcinoma-derived cell line. These cells proliferate in culture and differentiate into pure, postmitotic human neuronal cells (LBS-Neurons, Layton BioScience, Sunnyvale, Calif.) upon treatment with retinoic acid.

Thus, NT2 precursor cells appear to function as central nervous system (CNS) progenitor cells with the capacity to develop diverse mature neuronal phenotypes. When transplanted, these neuronal cells survive, extend processes, express neurotransmitters, form functional synapses, and integrate with the host. The final product is greater than 95% pure populations of human neuronal cells that appear virtually indistinguishable from terminally differentiated postmitotic neurons. The cells are capable of differentiation to express different neuronal markers characteristic of mature neurons, including all three neurofilament proteins (NFL, NFM, and NFH); microtubule-associated protein 2 (MAP2), the somal/dendritic protein; and tau, the axonal protein. Their neuronal phenotype makes them a promising candidate for replacement in CNS disorders, as a virtually unlimited supply of pure, postmitotic, terminally differentiated human neuronal cells.

■ TRANSPLANTED LBS-NEURONS: EVIDENCE TO DATE

Sanberg, Borlongan, and colleagues were the first to show that transplants of LBS-Neurons could reverse the deficits caused by stroke. Animals that received transplants of LBS-Neurons (and cyclosporine treatment) showed amelioration of ischemia-induced behavioral deficits throughout a 6-month observation period. They demonstrated recovery in the passive avoidance test, as well as recovery of motor function in the elevated body swing test. In comparison, control groups receiving transplants of rat fetal cerebellar cells, medium alone, or cyclosporine failed to show significant behavioral improvement. A second study that evaluated response relative to the number of cells transplanted confirmed the efficacy of transplanted LBS-Neurons in reversing the behavioral deficits resulting from transient ischemia in a middle cerebral artery occlusion rat model.

The first clinical study using LBS-Neurons included 12 patients with stroke primarily involving the basal ganglia and producing significant motor deficits. In the first 4 subjects, 2 million cells were implanted. The next 8 patients were randomized to receive 2 million or 6 million cells. The major objective of this study was to assess safety, but patients were also assessed for outcome using the European Stroke Scale (ESS) and the National Institutes of Health Stroke Scale. No complications occurred related to the implantation procedure. After more than 36 months of follow-up, there were no complications attributable to the implanted cells.

Though efficacy was not the major focus of this initial study, neurologic improvement occurred in some patients and a trend toward improved ESS scores was seen after 12 months. Changes were greater in the group receiving 6 million cells. The significance of such findings in this small uncontrolled trial is uncertain. FDG-PET studies were performed in all patients at baseline and at 6 and 12 months. Improvement in metabolism in the region of implantation was observed in 6 patients. Whether such changes represent metabolism in the grafted cells, increased function of host cells, or simply an inflammatory response to the implanted cells is unknown. In a few patients, the metabolism increases disappeared at 12 months, but in others the changes persisted. Two patients died of unrelated causes, and in 1 an autopsy was obtained. Surviving implanted LBS-Neurons were identified at the injection site within the area of infarction.

A phase 2 dose-response trial of patients with basal ganglia stroke and significant motor deficits was recently completed comparing implantation of 5 million and 10 million cells. An additional 4 control patients received no cell implants. There were no serious complications from the procedures.

Longer-term safety and efficacy results should enhance our understanding of cell implantation therapy for the treatment of stroke.

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