

AUTOLOGOUS BONE MARROW-DERIVED MESENCHYMAL STEM CELL TRANSPLANTATION AS A THERAPY FOR NEURONAL CEROID LIPOFUSCINOSIS

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ABSTRACT

The Neuronal Ceroid Lipofuscinoses (NCLs) are a group of rare genetic diseases characterized by neurodegeneration and accumulation of autofluorescent lysosomal storage bodies in numerous cell types. There are currently no effective clinical treatments for the NCLs, though research indicates that stem cell transplant could be efficacious. The bone marrow stroma is composed of a heterogeneous population of cells including endothelial cells, fibroblasts, adipocytes, osteocytes and two types of multipotent adult stem cells. Mesenchymal stem cells (MSCs) are stroma-derived adult stem cells which are known to give rise to cells of mesodermal origin including bone, cartilage, fat, tendon, muscle and possibly germ cells. MSCs may also be capable of differentiating into cells of a non-mesodermal origin, including neural cells. We have isolated a population of untransformed, self-renewing, multipotent, stroma-derived cells from both murine and canine bone marrow. These cells express protein markers associated with MSCs and grow quickly in culture, with a doubling time of 24-36 hours., Murine cells have been expanded in culture for more than 150 population doublings without evidence of senescence or loss of differentiation potential. Upon differentiation, these MSCs express appropriate protein and RNA markers and display histochemical features consistent with terminally differentiated cells.

Using a suicide gene knockout model, we demonstrate genetic modification of our MSC isolate through small fragment homologous replacement (SFHR) as a precursor to repairing NCL-specific genes in autologous cells. Intraocular injection of eGFP expressing murine MSCs into the eye of a murine model of human infantile NCL (INCL) show that transplanted MSCs are capable of surviving in the eye of an allogeneic host for up to 5 weeks. Within 2 weeks after injection, cells were observed to form close associations with the retinal tissue and at 5 weeks, MSCs had integrated into host tissue with no obvious signs of immune rejection. We also report that MSCs cultured on brain slices from NCL-affected mice are able to reduce the amount of autofluorescent storage material in surrounding cells through enzymatic cross-correction. Isolation of canine MSCs and characterization of several models of canine NCL lay the foundation for similar experiments in canine models.

Our characterization of these cells indicates they may be ideal candidates for studying autologous adult stem cell transplants as a therapy in patients with neurodegenerative diseases. Further experiments in animal model systems will help to determine the most effective method of implantation, the potential for cellular replacement and survival time of donor cells after transplant. Because of their multipotent phenotype, ease of isolation and growth in culture, ready ability to engraft in a multitude of tissues and susceptibility to genetic manipulation, autologous MSCs offer many advantages over other potential therapies as a treatment for NCLs. Development of a standardized approach to using autologous MSC transplant as a therapy could lead to vast improvements in clinical outcome not just for NCLs but for numerous other diseases including common ailments such as heart disease, stroke, diabetes and Parkinson's as well as other rare inherited genetic disorders.