REVIEW ARTICLE

Cell Therapy Strategies vs. Paracrine Effect in Huntington's Disease

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ABSTRACT

Huntington's disease (HD) is a genetic neurodegenerative disorder. The most common symptom of HD is abnormal involuntary writhing movements, called chorea. Antipsychotics and tetrabenazine are used to alleviate the signs and symptoms of HD. Stem cells have been investigated for use in neurodegenerative disorders to develop cell therapy strategies. Recent evidence indicates that the beneficial effects of stem cell therapies are actually mediated by secretory molecules, as well as cell replacement. Although stem cell studies show that cell transplantation provides cellular improvement around lesions in *in vivo* models, further work is required to elucidate some issues before the clinical application of stem cells. These issues include the precise mechanism of action, delivery method, toxicity and safety. With a focus on HD, this review summarizes cell therapy strategies and the paracrine effect of stem cells.

Key Words
Huntington's disease; Chorea; Cell therapy; Stem cell; Paracrine effect.

INTRODUCTION

Huntington's disease (HD) is an autosomal dominant neurodegenerative disorder. It is caused by an abnormal number of coronary angiography (CAG) trinucleotide repeats in the Huntingtin gene (HTT), which encodes a 350 kDa ubiquitously expressed protein, Huntingtin (Htt).[1] HD is characterized by movement disorder, cognitive impairment, dementia, and affective disturbances.[2] HD patients have more than 40 CAG repeats and show abnormal involuntary writhing movements. Juvenile HD patients have more than 60 CAG repeats.[1,3] The age of onset of HD is typically between 35 and 44 years old.

A HTT comprising more than 40 CAG repeats is translated into mutant Huntingtin (mHtt) protein, which causes the death of medium spiny neurons in the striatum. Normal Htt is ubiquitously expressed and is essential for embryonic development.[4] The mechanism of neuronal cell death by mHtt has not been clearly established although previous studies report that it has been linked with mitochondrial dysfunction, transcriptional dysregulation, altered protein-protein interactions, abnormal protein aggregations, and excitotoxicity.[5-7]

TREATMENT OF HUNTINGTON'S DISEASE

Many pharmacological drugs are used in the treatment of HD (Table 1). The treatment paradigm for HD patients depends on 3 main clinical domains: movement, psychiatric, and cognitive abnormalities. Tetrabenazine is the most commonly used drug for chorea. Antipsychotic agents, including Haloperidol, Pimozide, and Clozapine, are used to treat patients with psychiatric/behavioral comorbidities. Rivastigmine and Donepezil are the preferred treatments for improving cognitive function. These treatments are the result of limited evidence presented in the literature. Furthermore, comparison of the available treatment studies is problematic due to differences in study populations, variable outcomes, the use of different instruments, and the confounding effects of drugs.

CELL THERAPY FOR HUNTINGTON'S DISEASE

The pharmacological treatment of HD can alleviate symptoms, but it cannot cure the disease. Cell therapy strategies have been actively studied as a potential cure for HD. The ultimate goal of cell therapy is the replacement or neuroprotection of dead or dying cells. Cell therapy strategies can be classified into two broad categories based on the use of either fetal tissues/cells or stem cells. Studies using fetal brain tissue were performed using animal models of HD prior to 1990. Several clinical trials on HD patients have been performed with fetal tissues or cells. However, effective recovery has not been reported in any clinical trials, although some studies showed

Table 1. Pharmacological drugs on symptoms of HD

Treatment of chorea	
Antidopaminergic agents[8,9]	Tetrabenazine
Antipsychotic agents[10-17]	Haloperidol, Pimozide, Clozapine, Olanzapine, Ziprasidone, Aripiprazole, Risperidone, Quetiapine
N-methyl-D-aspartic acid receptor antagonists[18,19]	Amantadine, Memantine
Omega-3 fatty acids[20]	Ethyl-eicosapentaenoic acid
Treatment of cognitive dysfunction associated with HD	
Cognition[21,22]	Rivastigmine, Donepezil
Treatment of behavioral disturbances associated with HD	
Depression[23-26]	Fluoxetine, Venlafaxine, Mirtazapine, Clozapine
Psychosis[27]	Risperidone
Irritability, agitation[28-33]	Olanzapine, Quetiapine, Sertraline, Buspirone, Valproate, Propranolol

HD: Huntington's disease.

that fetal tissue transplantation provided cellular improvement around lesions.[34,35] Moreover, fetal tissue transplantation led to localized effects only and did not persist long-term.[34,36]

Stem cells are being studied in various disease models, in preference to fetal tissue or cells due to the limited availability of the latter. Stem cell research focuses primarily on neurodegenerative disorders. Several types of stem cells, such as embryonic stem cells (ESCs), bone marrow mesenchymal stem cells (BM-MSCs), neural stem cells (NSCs), adipose stem cells (ASCs), and induced pluripotent stem cells (iPSCs), are used to develop cell therapy strategies.

Embryonic stem cells are pluripotent, and mouse ESCs can differentiate into neurons, astrocytes, and oligodendrocytes.[37] It has been reported that human ESCs (hESCs) can differentiate into neurons in the lesions of HD animal models, attenuating progressive symptoms.[38] Despite these benefits of hESCs, complications arising from their use include immune rejection, ethical concerns, and tumor formation.[38] On the other hand, somatic stem cells such as BM-MSCs, NSCs, ASCs, and iPSs are ideal sources for clinical trials because these stem cells do not present the above mentioned immune rejection and ethical problems.

Murine and human NSCs (hNSCs) have been studied *in vivo* as cell therapy sources for HD. A study involving an hNSC treated HD animal group investigated the migration of transplanted hNSCs around a lesion site. Following tail vein or ventricle injection, a significantly greater volume of striatum was observed in the treatment group compared to the control group. Other studies reported that transplanted NSCs differentiated into neurons, oligodendrocytes, and predominantly, astrocytes, in *in vivo* HD models, resulting in partial functional recovery.[38-42]

Bone marrow mesenchymal stem cells and ASCs are easily obtained multipotent somatic stem cells that can be differentiated into neuronal cells. Moreover, these stem cells have the ability to secrete neuroprotective factors, such as growth factors, chemokines, and cytokines. Recent studies have shown that intrastriatal transplantation of BM-MSCs reduced striatal atrophy, although transplanted cells only survived for up to 7 days in transgenic HD mice. BM-MSCs can be genetically modified to provide sustained and long-term delivery of neuroprotective factors, which increase neurogenesis and protect against cell death.[43-45] Genetically modified MSCs are currently under consideration for use in the treatment of neurodegenerative disorders, including HD.[46]

Adipose stem cells are a feasible source for cellular therapy due to ease of isolation, manipulation, and a strong safety profile in the clinic. The intrastriatal transplantation of normal human ASCs reduced lesion volumes in an HD rat model.[47] To examine the long-term effect of ASC transplantation and investigate the possibility of autologous ASC transplantation in HD patients, HD patient-derived ASCs have been investigated over a period of 4 months in the YAC128 model.[48] The results showed similar expression levels of growth factors, such as brain derived neurotrophic factor (BDNF), hepatocyte growth factor (HGF), vascular endothelial growth factor (VEGF), and leukemia inhibitory factor (LIF), in HD ASCs compared with normal human ASCs. However, no long-term effects of transplantation with either HD or normal ASCs were observed in YAC128.

Embryonic stem cells have two limitations regarding their clinic application: the ethical issues surrounding their use and allogenic immune rejection. iPSCs provide a potential solution because they have the ability to differentiate into various cell types and can be induced from the fibroblasts of an HD patient.[49,50] iPSCs from an HD patient with 72 CAG repeats have been efficiently induced to form gamma-Aminobutyric acid neurons and were functional following transplantation into a rat model of HD.[51]

STEM CELL PARACRINE EFFECT BASED THERAPY IN HUNTINGTON'S DISEASE

Although stem cells have the ability to differentiate into any type of cell, recent studies indicate that the beneficial effects of stem cell therapies actually occur via secretory molecules in addition to cell replacement, the so-called paracrine effect.[47,52] Stem cells secrete a variety of growth factors, cytokines, and chemokines that regulate their biology in an autocrine/paracrine manner, and they interact with the surrounding microenvironment.[53,54] VEGF, HGF, insulin-like growth factor-1 and -2 (IGF-1, -2) and stromal-derived factor-1 secreted from stem cells are important to neuronal survival, neurogenesis, and
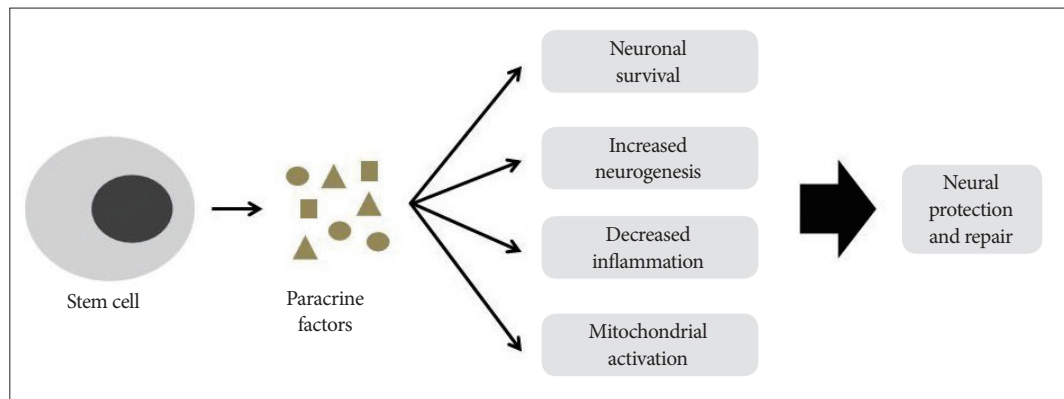
Figure 1. Possible mechanism of neural protection and repair by paracrine effects of stem cells. Stem cells release secretory molecules, including anti-inflammatory cytokines, various growth factors and extracellular vesicles. These factors could positively influence cell survival, neurogenesis, inflammation and mitochondrial function, leading to neural protection and repair.

mitochondrial activation via a bystander-like mechanism.[47,55,56] These positive effects on recipient neural cells result in protection and repair, leading to the inhibition of HD progression (Figure 1).

Adipose stem cells are multipotent somatic stem cells. They secrete multiple antiapoptotic growth factors, including VEGF, HGF, BDNF, basic fibroblast growth factor, and IGF-1.[57-59] One solution to the problem of stem cell availability may be the paracrine effect of ASCs.

The paracrine effects of human ASCs on HD pathology were investigated in cell culture experiments and HD R6/2 mouse models.[47] Transplantation of ASCs resulted in reduced lesion volume and fewer apoptotic striatal cells in the HD rat model compared with control animals. The ASC transplanted group showed significant improvement in apomorphine-induced rotation tests via the paracrine effect. ASCs have been injected into the R6/2 HD mouse model, and treated mice exhibited a significantly longer survival time than control mice.

The paracrine effect of ASCs in the R6/2 HD mouse model was also investigated.[60] ASC extracts were isolated and used to treat R6/2 mice via intraperitoneal injection. The results were similar to those obtained from stem cell transplantation, suggesting that the injection of these stem cell extracts could also slow HD progression.[60]

Taken together, the use of growth factors in HD could be an ideal stem cell strategy to protect against neuronal death, given that stem cells from an HD patient have the genetic components for autologous transplantation therapies. To implement this therapy, further works are required to elucidate the precise mechanism of the paracrine effects of ASC extracts. Prior to clinical application, thorough *in vivo* studies examining the delivery method, toxicity, and pharmacokinetics of therapeutic candidates are required.

CONCLUSION

Pharmacological drugs to cure HD are in development. Most of these drugs do not demonstrate significant effects, although several drugs are currently undergoing clinical trials. Stem cell therapy is an effective strategy for curing HD, and many preclinical trials show encouraging results. Although the precise mechanism of the stem cell paracrine effect has not been completely elucidated, this strategy has potential for clinical application.

Conflicts of Interest

The authors have no financial conflicts of interest.

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