Glaucma

Development of a Stem Cell Therapy for Glaucma

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Abstract

Glaucma is the most common form of optic neuropathy and a major cause of blindness worldwide. Currently, there is no cure for glaucoma. Damage to retinal ganglion cells and optic nerve caused by the diseases is permanent. Stem cells, which can divide indefinitely and differentiate into various cell types, offer a great premise for cell-replacement therapy. Recent breakthroughs in stem cell research that enable generation of pluripotent stem cells from adult somatic cells by reprogramming technology have opened new therapeutic possibilities. However, besides the common factors in inducing directed and homogenous cell differentiation and integration into the host tissue and establishing functionality, stem cell therapy for glaucoma is especially challenged by difficulties in driving optic nerve regeneration or repair. On the contrary, stem cell transplantation may still gain functional benefits by secreting neurotrophic factors and promoting neural plasticity. This review will discuss these advances and challenges in stem cell therapy.

Keywords

Glaucma, stem cells, mesenchymal stem cells, embryonic stem cells, induced pluripotent stem cells, transplantation, cell replacement therapy, neuroprotection

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Glaucma is the most common form of optic neuropathy and a leading cause of blindness worldwide.1 Currently, no treatment is available to reverse glaucomatous retinal ganglion cell (RGC) damage or vision loss. In clinic, glaucoma is diagnosed by optic disc pallor, central depression, excavation, and an increased cup-to-disc ratio.2 Many patients with glaucoma display an elevated intraocular pressure (IOP),3,4 but they do not normally experience blurry vision5 and visual field loss until a late stage of the disease.6 Treatment for glaucoma is solely directed at lowering the IOP through pharmacologic, surgical, or laser-based approaches.2 Although these treatments slow down the disease progression, they do not repair the damaged optic nerve or reverse vision loss.

New treatment strategies for glaucoma are in demand. Stem cell therapy presents a new intervention that holds great promise for reversing vision loss. Particularly, as pluripotent stem cells (SCs) can divide indefinitely and differentiate into almost any types of cells in the body, it makes them a superior source of cell therapy aimed at replacing cells lost due to disease or injury. In addition, SCs or progenitor cells can also work through a neuroprotective mechanism by secreting neurotrophic factors that prevent cell loss. In this review, we will provide an overview of recent advances in stem cell approaches for the treatment of glaucoma.

Types of Stem Cells and their Therapeutic Potentials

Sources of Stem Cells—From Bone Marrow-derived Cells to Induced Pluripotent Stem Cells

Several cell populations may be regarded as potential sources of retinal SCs. Primarily, these include adult SCs, embryonic SCs (ESCs), and induced pluripotent SCs (iPSCs). Adult SCs are found in small numbers in adult tissues, for example, mesenchymal SCs (MSCs), a widely studied human adult stem cell population, which can be obtained from bone marrow, blood, and adipose and dental tissue. On the other hand, adult somatic cells can be reprogrammed via nuclear transfer or cell fusion. Recently, a set of genes for cell reprogramming that allow somatic cells to return to their pluripotent state to form iPSCs was identified by Yamanaka’s group.7 This method enables the reprogramming of a patient’s own cells to differentiate into any cell types in vitro.

Stem Cell Treatment in Ophthalmology

Stem cell therapy may benefit patients with retinal degeneration by replacing degenerated cells or preventing cell death via secreting neuroprotective agents. Subretinal transplantation of retinal precursor cells derived from human ESCs (hESCs) has been shown to improve vision in a mouse model.
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Stem cell replacement therapy has also been shown to benefit ocular surface diseases. Corneal diseases or wound healing can be compounded by opacification of the transparent epithelium. Although transplantation of donor corneas is commonly practiced in clinics, increasing shortage of corneal donors and the risk for immune rejection present significant challenges. Regular turnover and repair of corneal epithelial cells occurs through limbal SCs, and transplantation of limbal SCs thus has brought opportunities for reconstructing the ocular surface and reversing vision loss. Successful restoration of a transparent cornea with differentiated epithelial cells has been observed in animal studies of undifferentiated ESC transplantation. Emerging evidence indicates that application of cultivated SCs for corneal surface reconstruction is coming of age.

**Stem Cell Treatment for Glaucoma**

Development of stem cell therapies for glaucoma has been focused on the replacement of two cell types: cells of the trabecular meshwork (TM) and RGCs. TM cells decrease with age, and it is evident that there is more cell loss in the TM of glaucomatous eyes compared with age-matched normal eyes. A recent study in an animal model of glaucoma suggests that transplantation of SCs to the TM lowers IOP through TM cell recovery, enhanced ocular outflow, and progenitor cell recruitment. In an independent study, injection of TM SCs or fibroblasts into normal C57BL/6 mice does not alter IOP compared with noninjected controls, further supporting the therapeutic potential for TM stem cell transplantation without impacting TM's normal function. Replacing damaged RGCs with SCs faces several obstacles, but evidence suggests that SCs may release trophic factors and work to the benefit of existing RGCs through a neuroprotective mechanism without requiring optic nerve reformation and functional integration. In this review, research attempts trying to replace both TM and RGCs, the two cell types mainly affected by glaucoma, will be discussed.

**Trabecular Meshwork Cells Derived from Stem Cells**

Disruption of the TM, where aqueous humor is drained, is thought to cause elevated IOP leading to glaucoma. These disruptions are reflected in both cell motility and endothelial function, which may include changes in cell shape, size, and permeability, and accumulation of damaged and cross-linked proteins with malfunctioning proteolytic cellular systems. Increases in oxidative stress also induces mitochondrial damage and trigger cell apoptosis. Treatment of dysfunctional TM thus may present a viable therapy. The aqueous humor circulates from the ciliary body through the anterior chamber to the TM. The primary function of TM is to maintain the aqueous flow and remove foreign objects through phagocytosis.

Cellular senescence of the TM and decreases in numbers of TM-cells with aging have been reported and are speculated to contribute to the pathophysiology of glaucoma. Accumulating evidence suggests that stem-like cells reside in the transitional area between the peripheral corneal and TM. SCs, including adult TM SCs and MSCs, are found in the human TM (hTM). Stem-like cells in the TM were first discovered by Raviola in the early 80s, which were later shown to possess stem-like properties and express distinct biomarkers. Interestingly, MSCs derived from the TM express TM-specific markers, suggesting that they are TM-specific progenitors, forming a reservoir for TM repairs. A recent report suggests that TM cells may also be derived from iPSCs.

Adult TM SCs can be differentiated into cells characteristic of corneal epithelium, phagocytotic TM cells, and retinal neurons in vitro. Phagocytosis is an important function of TM cells. Moreover, transplanted TM SCs home to the TM following injection into the healthy mouse anterior chambers. Human iPSC-derived TM cells also resemble native hTM, with similar morphology and the ability of phagocytosis. Whereas, transplanted bone marrow-derived MSCs primarily exert their benefits through a paracrine effect that recruits progenitor cells to promote TM regeneration. Injection of MSCs into the anterior chamber of rats with laser-induced glaucoma significantly decreased IOP and improved the histology of the angle structure without observing permanently integrated MSCs in the host tissue. This paracrine effect of MSCs was confirmed using the medium conditioned by cultured MSCs. To date, proof of concept studies for a stem cell therapy for TM repair have been carried out in animals. Importantly, transplantation of iPSC-cell-derived TM cells into the anterior segments reestablished IOP homeostatic function in perfused human outflow pathway organ cultures. Together, these studies establish the conceptual feasibility of a cell-replacement approach for restoring IOP regulatory function, supporting a potential benefit for human patients with open-angle glaucoma.

**Cell Therapy for Treating RGC Damage**

The neuroretina, specifically RGCs, is irreversibly damaged in glaucoma. Thus, neuroprotection focused on improving neuron survival and preventing progressive RGC damage is critical. Multiple studies have reported beneficial effects on the survival of injured RGCs and improved visual function in animal models of optic neuropathy, suggesting the nature of neuroprotective effects by transplanted SCs. The beneficial effects could also be seen by injecting secretome or conditioned medium of SCs, associated with the production of platelet-derived growth factors and extracellular matrix proteins. Embryonic mouse neural SCs modified to secrete ciliary neurotrophic factor were shown not only attenuating RGC axon loss but stimulating long-distance axonal regrowth. Therefore, the stem cell approach is appealing because it presents a source of sustained production of neuroprotective and growth-stimulating factors.

Ideally, beyond neuroprotection, a more sophisticated use of stem cell technology could be applied to replace RGCs that have been lost and reverse vision loss. However, this approach is faced with several obstacles. Present protocols for pluripotent stem cell differentiation often generate a heterogeneous population of neurons and glial cells. Moreover, differentiated neurons, in contrast to early-stage SCs, have limited ability...
to integrate into the host tissue following transplantation. Although, in some circumstances, partial integration was detected when cells were injected into the epiretinal space, functional improvement, which likely requires formation of new optic nerve fibers that can reach their brain targets and form appropriate synapses, has not been observed. Despite these limitations, cultivated SCs present an invaluable resource for the understanding of disease pathology and drug screening.

Conclusion
Glucoma is a leading cause of irreversible blindness worldwide. Stem cell technology is a new and rapidly developing field that presents a great potential to replace or revive damaged neurons in glaucoma. While the TM stem cell transplantation and neoproneurogenic property of the SCs holds more promise for the near future, additional research is needed to further characterize the secretome compositions for more efficient design of cell grafts. With the endless possibilities of stem cell division and differentiation, iPSCs present a unique opportunity for exploration of diverse causative mechanisms of glaucoma as well as for cell-replacement therapy. While more research on stem cell therapy is warranted, encouraging results in animal models point to the likelihood of human trials in patients with glaucoma in the foreseeable future.