CELL-BASED THERAPIES TO LIMIT PHOTORECEPTOR DEGENERATION

TERAPIAS CELULARES PARA LIMITAR LA DEGENERACIÓN DE LOS FOTORRECEPTORES

LUND R

Subretinal transplantation of cells into the mammalian eye for the treatment of retinal degeneration was first investigated in the 1980’s. In these studies, retinal pigment epithelium (RPE) cells were introduced into the subretinal space of the Royal College of Surgeons (RCS) rat, an animal model of photoreceptor degeneration caused by RPE dysfunction. Interestingly, the transplanted RPE cells slowed the progress of photoreceptor loss presumably functioning by replacing the dysfunctional RPE (1). The RCS rat now serves a generic model of photoreceptor degeneration; however, it is especially relevant to age related macular degeneration (AMD) where the developed vision loss is believed to result from compromised RPE cell function. Given the success of the RPE cells in slowing degeneration in the RCS rat, the approach was applied to late stage AMD patients however, with little evidence of success.

The results of the human studies point to several major issues that require special attention before further progress can be expected. First, careful evaluation must be given to the use of suitable animal models for initial investigation into the efficacy of the cell-based therapies. In addition, consideration must be given as to the source of cells to be used in experiments. Specific emphasis should be placed on functional measures of efficacy and safety issues. Finally, one must carefully consider how to transition from rodent models in the laboratory to patients in a clinical setting. This includes delineating the specific patient group that would most benefit from the treatment.

FUNCTIONAL MEASUREMENTS

In early studies using RCS rats, efficacy of cell transplantation was assessed solely by measuring the thickness of the outer nuclear layer, which in rodents comprises more than 95% rods. Recent studies now place stronger emphasis on measuring visual function, particularly cone-related vision in these animals. Three clinically relevant measures are used to assess visual function – visual acuity, luminance thresholds across the visual field and electroretinogram (ERG).

Visual acuity is assessed by optomotor testing, a behavioral test that measures the optomotor response of an animal to a rotating sine-wave grating. The test provides a noninvasive way to measure visual acuity over a period of time in a large number of animals. Luminance threshold recordings determine the relative sensitivity of the retina over the visual field. Here, luminance threshold responses are recorded from electrodes placed across the area of the superior colliculus, a brain region with a well-delineated map of the visual field. This test can be used to determine the local efficacy of the cell treatment in maintaining retinal function. Finally, visual function is measured at the cellular level using full-field electroretinogram recordings (ERG). ERGs assess the

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electrical activity most importantly of photoreceptors, bipolar and amacrine cells.

**CELL SOURCE**

Early work using freshly harvested RPE cells determined that it was necessary to harvest cells from fetal eyes. However, the use of fetal tissue not only raises logistical and ethical issues, but also presents the potential for transfer of infective agents that cannot be adequately detected within the time frame available. These concerns prompted exploration of a range of renewable cell sources, which can be produced on a commercial scale and rigorously screened prior to implantation.

A range of such cell types has been examined for their efficacy in cell-based therapies for retinal degenerations (2). With the macular degeneration patient groups in mind, a first approach was to replace RPE cells with RPE cell lines. Subsequently, other cell types have been explored, some of which clearly function or can be engineered to function as cells delivering a range of growth factors. Because there is ample evidence to show that growth factor injection plays an important role in slowing degeneration in animal models of retinitis pigmentosa, examination of growth factor delivering cells provides an alternative and continuous delivery system for such factors.

Of the cells with an RPE phenotype, the one that is effective in rescuing acuity, luminance threshold and ERG measures as well as answering the various safety issues is a set of cell lines derived from embryonic stem cells (3). These cells satisfy the safety criteria outlined below and they appear to be effective in mouse models of macular degeneration. They are now being explored for potential clinical application.

A second and more varied group of cells function by delivering growth factors. The first of these to be explored was the Schwann cell (2), which makes a number of factors known to support photoreceptor survival. It is effective in rescuing vision and has the potential of being delivered as an autologous graft, so minimizing safety concerns, including immune responses, but preparation requires a more complex procedure, involving removal of a peripheral nerve, isolating the cells and growing them in culture. Other cells that may function similarly are cells isolated from the umbilical cord stroma, which are effective, readily renewable and stable (4). An additional cell type, derived from fetal forebrain tissue (5), can function on its own or can be transfected with factors such as GDNF to be more effective. Interestingly, this cell takes on some of the characteristics of RPE cells after transplantation raising the possibility of it expressing two separate functions.

This is far from a complete list and other cell types are being explored and may be suitable for injection into the subretinal space, including ones that have been used in clinical trial for other tissues. Important too is the fact that some cells such as fibroblasts and cells isolated from the placenta have no effect in preserving photoreceptor rescue: there are selective behaviors.

**SAFETY CONSIDERATIONS**

With all cell-types, several safety issues must be considered. How long do donor cells survive, do they show any sign of uncontrolled growth over time, including tumorous transformations (in the case of embryonic stem cell-derived cells, whether they may form teratomas)? Do the cells spread from the site of injection, perhaps invading compartments of the eye or body in which they could cause ancillary effects? The particular cell line must be assessed for stability of phenotype, evidence of chromosome damage with progressive cell divisions, and as far as possible screened for transmittable pathogens. Finally the cells must be manufactured under defined Good Manufacturing Conditions (GMP).

**TRANSLATION OF CELL-BASED THERAPIES FROM RODENTS TO HUMANS**

The translation of therapies from the use in rodent models to a clinical setting is not simple, a fact that is highlighted by the lack of success in the human clinical trials for AMD mentioned earlier. Clearly the safety issues outlined above and the question of immune tolerance must be addressed. Furthermore, the technique of introducing cells into the rodent eye via a trans-scleral approach would not be appropriate for use in humans. Rather, a transvitreal approach is ideal. To address these issues, some consideration to doing studies in non-human primates is of value given that structure of the non-human primate eye closely resembles that
of a human including the presence of macula. Furthermore, the ability of cell-based therapies to deliver trophic factors allows the cells to be applicable to a larger patient group unlike gene therapy, which is restricted to correcting a specific genetic disorder. As with any clinical extrapolation, properly designed and reported clinical trials must be adhered to with appropriate objective evaluation criteria. Because the scale of such endeavors is substantial, it can best be done with commercial partners.

Overall this field is showing real momentum towards generating effective clinical treatments. Useful cells have been identified, the issues of translation from laboratory to clinic have been largely resolved and clinical trials are underway or planned. The approach would help stabilize the eye in patients with progressive photoreceptor loss and pave the way for the more complex goal of replacing photoreceptors once they are lost.

REFERENCES