A0124

Phase I/IIa Clinical Trial of Human Embryonic Stem Cell (hESC)-Derived Retinal Pigmented Epithelium (RPE, OpRegen) Transplantation in Advanced Dry Form Age-Related Macular Degeneration (AMD): Interim Results

Eyal Banin[1], Yitzchak Hemo[1], Tareq Jaouni[1], Devora Marks-Ohana[1], Maria Gurevich[2], Oscar Cuzzani[3], David S. Boyer[4], Benjamin Reubinoff[5]

[1]Center for Retinal and Macular Degenerations, Department of Ophthalmology, Hadassah-Hebrew University Medical Center, Jerusalem, Israel; [2]Cell Cure Neurosciences Ltd, Jerusalem, Israel; [3]BioTime, Inc., Alameda, California, United States; [4]Retina-Vitreous Associates Medical Group, Los Angeles, California, United States; [5]Center for Embryonic Stem Cells and the Department of Gynecology and Obstetrics, Hadassah-Hebrew University Medical Center, Jerusalem, Israel

INTRODUCTION

- Age related macular degeneration (AMD) is the leading cause of blindness in people >50y in the developed world. Approximately 90% of these patients suffer from the dry form and currently there are no FDA-approved therapies beyond nutritional supplements.
- In dry-AMD, there is dysfunction and loss of retinal pigment epithelial (RPE) cells in the macular region. In the advanced stage, widespread loss of RPE and photoreceptors in the macular area evolves into geographic atrophy (GA), leading to severe vision loss.
- Attempts to transplant human embryonic stem cells (hESC)-derived as well as iPSC-derived RPE cells in patients with AMD, in suspension or on scaffolds, are being conducted by a number of groups.[1-4]
- Our directed differentiation protocol allows derivation of RPE cells from hESCs.[5] These NIH-approved cells, grown under xeno-free conditions, underwent rigorous characterization and extensive safety and efficacy testing, and were FDA-approved for use in the current clinical trial.
- In the Royal College of Surgeons (RCS) rat model of retinal degeneration, our hESC-derived RPE cells (called OpRegen) settled into monolayers, polarized, & begin functioning (**Figure 1**), improving both structure and function compared with untreated controls.[6]

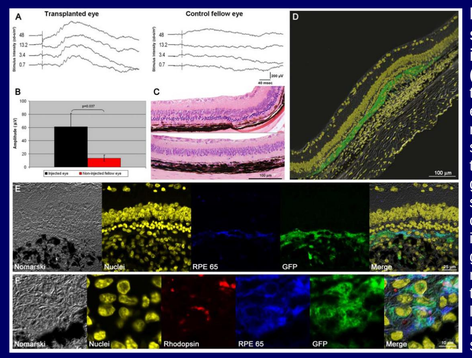


Figure 1. hESC-derived RPE cells survive and provide long term rescue in RCS rat eyes
(A,B) Relative preservation of ERG responses in transplanted versus control fellow eyes 13w after engraftment. (**C**) H&E stained sections, showing relative preservation of ONL in proximity to a subretinal graft (upper panel) as compared with thin degenerated ONL in an area distant from the graft in the same eye (lower panel). (**D**) GFP+ subretinal graft (green). Nuclei are counterstained with DAPI (yellow). (**E**) Confocal images showing GFP+ cells within a subretinal graft co-expressing RPE65. (**F**) Higher magnification confocal images of subretinal transplanted RPE cells showing the co-localization of pigment, GFP, RPE65 and rhodopsin within the same single cells.
Scale bars: (**C,D**) 100μm, (**E**) 25μm, (**F**) 10μm. Bars represent mean±SEM. (adapted from ref. 5)

OBJECTIVES

- The safety and tolerability of OpRegen is being evaluated in a dose escalating Phase I/IIa clinical study in patients with advanced dry AMD accompanied by GA (NCT02286089).
- Safety & imaging data from the first 3 cohorts (n=9 subjects), who received a subretinal transplant of 50k-200k cells in suspension, with up to 2 years follow up in some, are reported in this poster.

METHODS

- Trial is planned for 24 patients, ≥50 years, with advanced dry AMD and GA
  - Cohorts 1-2: Three patients each; Cohort 3: six patients (all 3 Cohorts with BCVA ≤ 20/200)
  - Cohort 4: Twelve (12) patients, BCVA ≤ 20/64
  - Doses have ranged from $50x10^3$-$200x10^3$ in 50-100μl of balanced salt solution (BSS)
  - Staggering intervals within and between cohorts initially with periodic Data and Safety Monitoring Board (DSMB) review and approval before proceeding to next cohort.
- Transplantation is performed by subretinal injection following conventional 23 or 25G vitrectomy.
- Systemic immunosuppression is administered 1 week prior to transplantation until 1 year after.
- Systemic and ocular safety is closely monitored. Retinal function & structure are assessed using various techniques including BCVA, and color, OCT and fundus autofluorescence (FAF) imaging.

RESULTS

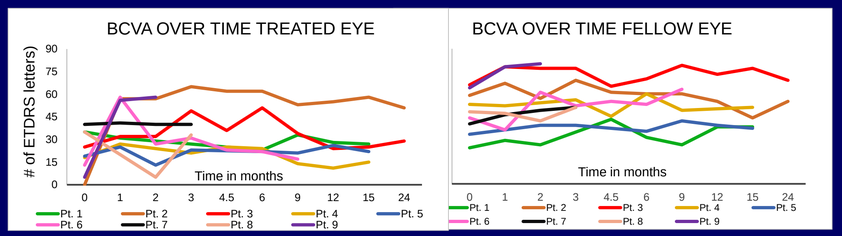


Figure 2. Best corrected visual acuity (BCVA). Best corrected visual acuity (BCVA) did not decrease in treated eye. Improvement noted in treated eye of Patient 2 is most probably due to clearing of vitreous and post capsule opacity during surgery. BCVA in fellow eye has remained largely stable.

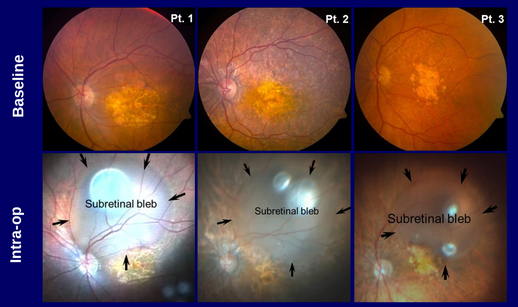


Figure 3. Color Fundus Imaging – Cohort 1
- Pre-operative photos show large areas of GA.
- Intra-operative images show location of the subretinal blebs formed following injection of the cell suspension.

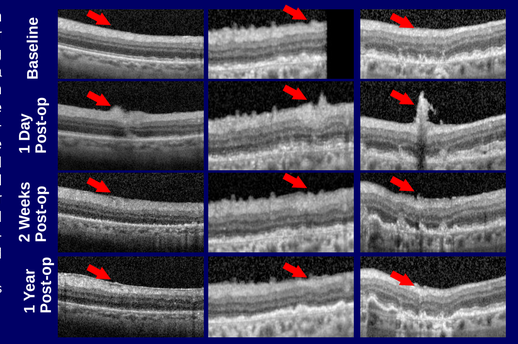


Figure 4. Healing of injection site – Cohort 1
Subretinal fluid absorbed rapidly and OCT images show that the site of retinal penetration by the cannula (arrows) heals within 2 weeks. ERMs developed in some cases, likely due to incomplete vitrectomy or potential efflux of RPE cells. Red arrow indicates location of the injection.

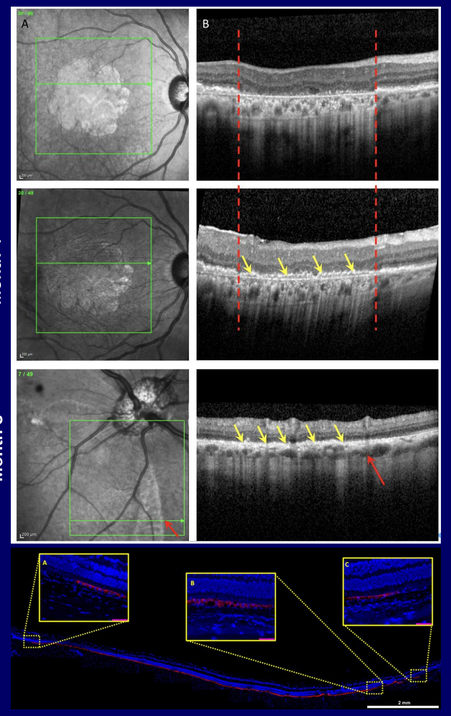


Figure 6. Comparison of OpRegen transplantation in human subject and minipig eye. In-vivo OCT images in a human and in a minipig following transplantation demonstrated similar findings, namely irregular subretinal hyper-reflectance (column B, yellow arrows) which is especially prominent in the human subject in an area of GA (delineated by dashed red lines). In the pig eye, this irregular hyper-reflectance abruptly stops at the border of the surgical bleb (red arrow). It was later possible to show by IHC in the pig eye that the transplanted cells survived and formed a subretinal layer with an intact layer of photoreceptors above them (lowest panel, paraffin-embedded pig eye sections stained with a human-specific marker, anti-Tra-1-85 (red); DAPI nuclear counterstain (blue). The results in the pig eye support the possibility that hyper-reflectance on OCT imaging correlates with presence of the transplanted cells.

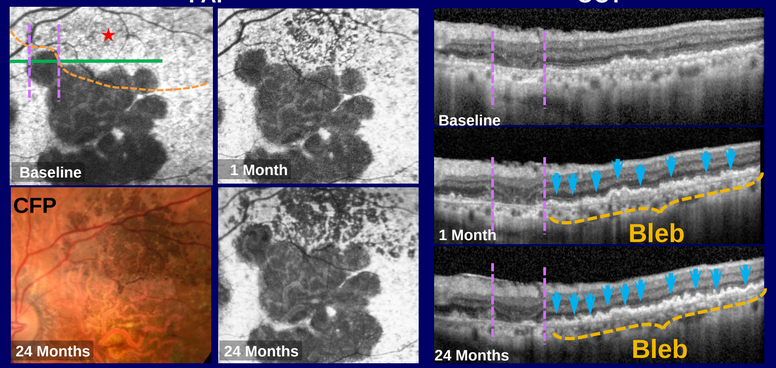
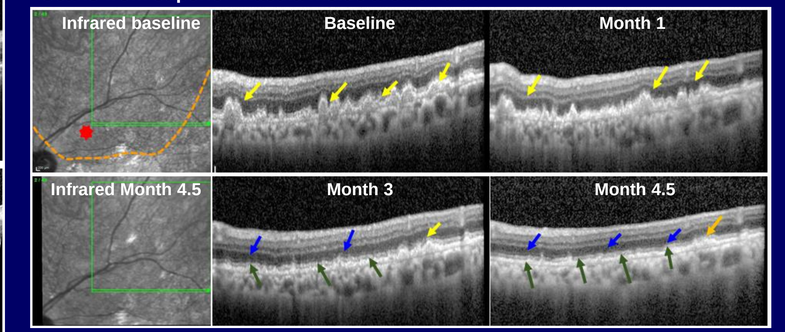


Figure 5. FAF, CFP, and OCT images demonstrate potential signs of long-term engraftment and survival of transplanted cells. Note subretinal pigmentation on color fundus photos and hypo/hyperflourescent spots on FAF imaging in area of transplant persisting to two years. On OCT, irregular subretinal hyper-reflectance is visible as soon as Month 1 and throughout follow-up. Of note, systemic immune suppression was halted at 1 year. These findings may suggest that immune rejection is limited or not present.

Scans from superior macula inside area of bleb



Scans from inferior macula outside area of bleb

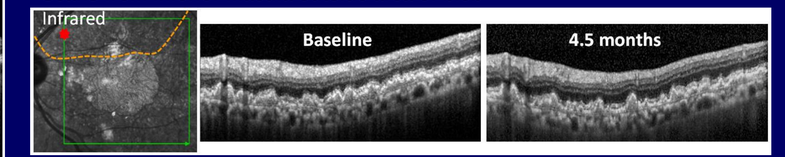


Figure 7. Changes in drusen appearance in area of transplant. Note reduction and change in the appearance of soft and confluent drusen (yellow arrows) over the span of a few months post-transplantation in a treated area (first two rows). The outer nuclear (photoreceptor) layer (blue arrows) and ellipsoid zone (green arrows) assume a more regular appearance with only few cuticular-like drusen (orange arrow) remaining. In an untreated area (lower row) the drusen persist. These findings are early and need to be further followed.

SYSTEMIC AND OCULAR SAFETY OBSERVATIONS IN PATIENTS TREATED TO DATE (N=9):

- No treatment-related systemic SAEs to date
- Four unrelated SAEs occurred in three subjects
- No unexpected ocular AEs have been observed
- Expected AEs included surgery-related conjunctival hemorrhage, worsening of cataract and epiretinal membrane formation (ERM)
- New or worsening ERM was reported in 7/9 subjects, 4 of which were considered mild and 3 were thicker. In 2 patients no ERM-related changes were present. None required intervention to date.
- A small PED developed in one patient during F/U, also in fellow untreated eye without sub- or intra-retinal fluid.

CONCLUSIONS

- Following subretinal transplantation of Opregen (hESC-RPE) in suspension there is rapid healing of the injection sites, and visual acuity remained stable throughout the study with follow-up of up to 24 months
- Subretinal pigmentation in the treated area is observed, which has remained stable for 2 years in some subjects
- There are additional signs suggesting potential RPE engraftment in the area of implantation, particularly subretinal hyperreflective areas seen on OCT in both pigs and human subjects; in pigs this correlated with presence of transplanted cells on histology
- New or worsening ERMs that did not require surgical removal were observed in 7/9 patients
- Subretinal transplantation of OpRegen appears well tolerated to date and there are potential early signs indicative of improved retinal structure in the treated areas in some cases, which will need additional follow-up and observation.
- Cohort 4 will be launched soon, treating patients with better vision (<20/64) and smaller areas of GA

REFERENCES: 1) L. Cruz et al. Nature Biotech. 2018;36: p328–37. 2) A. Kashani et al. Science Transl Med. 2018;10(435). 3) W. Song et al. Stem Cell Reports. 2015;4(5): p860–72. 4) S. Schwartz et al. Lancet. 2015;385(9967):p509–16. 5) M Idelson et al. Cell Stem Cell. 2009;5(4): p396-408. 6) T. McGill et al. Transl Vis Sci Technol. 2017;6(3):17.

Disclosures: Eyal Banin, Cell Cure Neurosciences, Consultant, Patent; Yitzchak Hemo, Cell Cure Neurosciences, Patent; Tareq Jaouni, None; Devora Marks-Ohana, None; Maria Gurevich, Cell Cure Neurosciences, Employment; Oscar Cuzzani, BioTime, Inc., Employment; David Boyer, Retina Vitreous Associates Medical Group, Employment, BioTime Inc., Consultant, Investigator; Benjamin Reubinoff, Cell Cure Neuroscience, Financial Support, Consultant, Patent.

These studies are supported by Cell Cure Neurosciences Ltd, Jerusalem, Israel and BioTime Inc., Alameda, CA, USA. We wish to thank all patients who are participating in this study and all investigators and nursing staff at participating centers.