CALIFORNIA STATE SCIENCE FAIR
2016 PROJECT SUMMARY

Name(s)
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Project Number
S0534

Project Title
Maintaining Viability in Cellular Therapies for Age-Related Macular Degeneration during Cryopreservation

Abstract

Objectives/Goals
Finding an effective combination of cryoprotective agent (trehalose) and cryopreservation method (vitrification) to maintain the highest percentage of viability in human embryonic stem cell derived retinal pigmented epithelium cells (hESC-RPE), so that a cellular therapy for dry age-related macular degeneration can be available to clinics worldwide.

Methods/Materials
I read hundreds of scientific journals and articles about stem cells and cryopreservation to gain background information. I Adapted and conducted two experiments based off of a published journal I found by Dr. Kuwayama, and tested for cell viability through an Alamar Blue assay. For the first experiment, I just substituted trehalose for the original, published cryoprotective agent and used a different cell type. I used samples exposed to trehalose and sucrose (from the original procedure) frozen through vitrification and a control sample not exposed to anything. For the second experiment, I added a 250mM trehalose pre incubation solution to the first experiment's procedure; samples were exposed to the pre solution for different amounts of time (control samples included).

Results
1) The cell viability in the first experiment was significantly higher for the trehalose samples than the samples with the original procedure by about 25% and proved to be contrary to my hypothesis.

2) The results were not as successful as the first experiment, but the difference between the most successful and worst samples was about 38%; my hypothesis was correct, but the overall procedure proved to be not effective with all of the percentages of viability under 50%.

Conclusions/Discussion
Although I am not able to conclude that a trehalose and vitrification combination is the most effective in maintaining viability in the cellular therapy, I can suggest that a pre incubation addition is in fact detrimental for the cells. Yes, there must be more experiments in place to confirm this "suggestion," but my experiment has found a starting place for other scientists.

Summary Statement
I proved that my first adapted version of a published procedure with trehalose could store human embryonic stem cell derived retinal pigmented epithelium more effectively without a pre incubation solution during cryopreservation.

Help Received
I did all of the research myself, including background research, and conducted each of the experiments myself. My mentor, Dr. Britney Pennington, demonstrated a few assays and supervised me in the stem cell lab. I used resources from Prof. Dennis Clegg's lab at UCSB and from CIRM.