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Two-parameter model of cell membrane response to cryoprotectant agents

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Cryobiology is a widely applicable topic of biological science. From thoroughbred horse gene preservation to organ transplants, the applications of cryobiology, specifically cryopreservation, are broad. The two major problems encountered during cryopreservation are the following: intracellular ice formation and solute damage (Mazur et al, 1972, Exp Cell Res 71: 343-355). These are somewhat alleviated with cooling rate variation and the use of cryoprotectants (CPAs) such as DMSO, glycerol, and ethylene glycol. Scientists desire standardized protocols to optimize the process of adding and removing CPAs and applying optimal cooling and warming rates. Part of this requires understanding cells and/or tissues membrane permeability to water and the chosen CPA (Gao and Critser, 2004, Biomaterials and Biomedical Engineering). There are several mathematical models of the volume change process including the "two-parameter" model which is used exclusively for this project (Benson et al., 2004). The two differential equations governing the cell volume behavior are the following:

To empirically find the unknown parameters L_p and P_s , water permeability and solute permeability respectively, a culture counter was used to gather cell volume change data. Cells were placed in a media then CPA was added to cells. The culture counter measures a voltage change which is directly related to the cells' volume change. The culture counter is a "dirty" way to gather data and the data sets acquired have substantial amounts of digital noise. The first step in analyzing the data was to filter the data to eliminate the majority of the noise and extract a relatively clean data set. This was done by isolating small bins of data, finding the average value, and removing the outlier datum. Next, a curve was fit to the data set to find the two unknown parameters. This was done with the Runge-Kutta numerical method for solving ordinary differential equations. The data clean up and curve fitting required a computer function and was written using the program MATLAB. The function can be applied to most sets of data to find the parameters for various cells and CPAs. These results will be used in optimizing data analysis for solute and solvent mass transport studies and will facilitate improved cryopreservation methods.