Complete Summary

"The ability to replace organs and tissues on demand could save or improve millions of lives each year globally and create public health benefits on par with curing cancer" - Organ Preservation Alliance (Nat Biotech 2017: 35: 530-42).

Storage at subzero temperatures has long been pursued as a strategy to improve the availability of tissues and organs for transplantation. The successful cryopreservation of multicellular systems and complex vascularized tissues remains a formidable challenge. Liver cryopreservation is particularly challenging due to the delicate hepatic microstructure, with an anatomy comprised of multiple cell types each with their own optimal cryopreservation conditions. However, the need for advancing liver preservation is great as more than 321 Canadians (CORR, 2017) and 14,000 Americans (UNOS, 2018) are waiting for a liver to become available.

The most significant cause of decreased viability and impaired function during tissue and organ cryopreservation is physical and chemical damage resulting from ice crystal formation and growth. Ice recrystallization is the process that occurs during freezing and thawing where large ice crystals grow larger at the expense of smaller ones and is a major cause of cellular damage and mechanical disruption to tissue structures that reduces post-thaw viability and potency. Our group has identified carbohydratebased libraries of small molecule ice recrystallization inhibitors (IRIs) that we have used successfully to improve the cryopreservation of cell-based therapeutics. By preventing or limiting extra- and intracellular ice recrystallization, the physical and chemical stresses encountered during freezing and thawing can be significantly altered allowing for improved cell recovery and viability. This approach mimics strategies used in nature by freeze tolerant organisms that survive the winter by controlling ice formation and growth. We hypothesize that reducing ice recrystallization using novel small molecule ice recrystallization inhibitors will reduce cryoinjury and enhance post-thaw viability and function of cryopreserved whole liver.

To exploit the potential of this emerging class of cryoprotective agents, we will employ a comprehensive strategy to: evaluate the effectiveness of IRIs to control intra- and extracellular ice recrystallization and modulate cryoinjury in cell model systems (SA1); assess and model permeation, distribution and interstitial ice control characteristics of IRIs in liver tissue sections (SA2); and evaluate the permeation of IRIs and their use to control intravascular, interstitial and intracellular ice crystal damage in partially frozen rat liver (SA3).

A major strength of this proposal is its interdisciplinary research team comprised of a cryobiologist with expertise in ice formation, a biomathematician building cell-based models of cryoprotocol damage in tissues and organs, a synthetic organic chemist with extensive expertise in cryobiology, and a transplant surgeon with expertise in ex vivo perfusion and microvascular surgery. New knowledge generated in these objectives will provide insights into the general mechanism of action of IRIs and how they improve postthaw recovery of cell, tissue and organ systems. In addition, the development of novel methods for hepatocyte and liver tissue and organ cryopreservation will greatly improve the banking of products for biomedical research, ex vivo bioartificial devices, drug screening and transplantation.

Lay Title and Lay Abstract

Lay Title:
Storage of liver at low temperatures using synthetic ice blockers

Lay Abstract:
As stated by the Organ Preservation Alliance in their recent call to action, “the ability to replace organs and tissues on demand could save or improve millions of lives each year globally and create public health benefits on par with curing cancer” (Nat Biotech 2017: 35: 530-42). Storage at subzero temperatures has long been pursued as a strategy to improve the availability of the liver for transplantation. However, the successful cryopreservation of liver remains a formidable challenge as ice crystal formation and growth can cause cell damage and disrupt important structures within the tissue or organ resulting in poor post-thaw function. Our group has developed synthetic, small molecule ice recrystallization inhibitors which function to control both extra- and intracellular ice crystal growth and can be used to improve the recovery of many different cell-based therapeutic products. As this approach of controlling ice crystals is similar to strategies used in nature by freeze tolerant organisms to survive winter, we are interested in testing if these ice control agents can protect liver tissue and whole liver from freezing and thawing damage. Through this work, we will develop new insights into how these ice control agents function to protect cells, tissues and organs from freezing injury and we will explore their use in the low temperature preservation of cell-based products that are needed in biomedical research, drug testing, and transplantation.