The findings have important implications for the advancement of cryopreservation techniques for cardiac allografts, which may be an important step in tackling the organ shortage crisis. Moving forward, further research should focus on optimizing cryopreservation protocols, exploring species-specific variations, and evaluating CPA performance in both animal and human trials.
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EXPANDING THE HORIZON: USING FATTY ACIDS AND MANNITOL AS ALTERNATIVE FUELS AND ONCOTIC AGENTS FOR NORMOTHERMIC EX VIVO HEART PERFUSIONS IN ADULT RATS

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Normothermic machine perfusion (NMP) is an advanced technique for ex vivo organ perfusion using a nutrient- and oxygen-rich perfusate at 37°C. The main objectives are to reduce injury, increase preservation duration, assess graft viability, and facilitate transplantation. The perfusate, enriched with glucose as a vital energy source, is crucial in meeting the metabolic demands of specialized organs like the heart in rodent models. Additionally, oncotic agents like bovine serum albumin (BSA) help maintain osmotic pressure and reduces fluid build-up (edema). While glucose and BSA are commonly used in rodent heart perfusions, there is growing interest in exploring alternative fuel options such as fatty acids, which are natural fuels for the heart, and alternate oncotic agents like Mannitol which recently have shown to have some cardioprotective properties. This preliminary study used a fatty acid (Oleic acid)/Mannitol perfusate and compared it with Glucose/BSA (control group) in adult rats (n=3) to assess the cardiac physiology using Langendorff perfusion. Major markers for heart injury (Troponin), heart parameters (vascular resistance, edema, oxygen uptake rate (OUR) and heart rate) during perfusion were analysed for each group. Interestingly, in Oleic acid-Mannitol group, OUR rate was lower during 4 hr perfusion compared with control group, which was expected as fatty acids are more energy dense fuel compared to glucose and yield more energy using lower oxygen consumption. Additionally, the addition of Mannitol, which is less viscous compared to BSA, significantly reduced vascular resistance. Moreover, troponin levels were lower in Oleic acid-Mannitol group (although not significantly different), while edema levels were comparable between both groups. These findings suggest the potential for improving traditional cocktails in rodent heart perfusions to enhance preservation and transplantation outcomes in clinical settings. Further research is needed to comprehend the full benefits and implications of these alternative fuel options and oncotic agents.
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RESONANCE RAMAN SPECTROSCOPY DETECTS LEAKY MITOCHONDRION IN RAT LIVERS WITH ISCHEMIA-REPERFUSION INJURY

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Ischemia-Reperfusion Injury (IRI) is a critical problem in organ transplantation that hinders clinical translation of novel technologies aiming to extend organ preservation time without continuous oxygenation. Resonance Raman (RR) spectroscopy is an innovative approach to quantitatively assess mitochondrial redox state via rapid organ surface measurements. Since mitochondrial damage contributes greatly to the development of IRI, RR reduced mitochondrial ratio (3RMR), a readout of RR spectroscopy on percentage of mitochondria in reduced state, can be a powerful parameter in organ assessment predictive of transplant success/outcome. In this study, 3RMR surface readings were performed on rat livers during three hours of subnormothermic machine perfusion (SNMP) after fresh procurement, as well as one hour and three hours of warm ischemic time (storage in 37oC saline solution) to evaluate mitochondria health following ischemia-reperfusion stress. Preliminary data confirmed that rat livers with 3 hours of ischemia exhibited classic injury parameters such as absent bile production, elevated lactate level, excessive tissue edema, and significantly lower oxygen consumption during SNMP in comparison to healthy fresh livers (n=4 per group, $p<0.05$). Interestingly, RR results showed that injured livers had lower percentage of reduced mitochondria (3RMR < 10%) than healthy livers (3RMR ~ 15-20%), despite complex IV being fully oxidized at maximum capacity of direct oxygen consumption in all study groups. Further RR spectroscopy analysis suggested that complex III of injured livers significantly leaked electrons/protons during reperfusion with less than 8% in a reduced state after 180 minutes of SNMP. Meanwhile, healthy livers had 30% reduced complex III which remained relatively unchanged through SNMP. These observations agreed with published literature that organs which developed IRI post-transplant commonly had leaky mitochondria resulting in ROS production, oxidation stress, and, finally, cell death. In other words, RR spectroscopy enables quantitative mitochondrial assessment beneficial for detection of organ injury following warm ischemic stress.
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Conflict of Interest: SN Tessier a co-inventor on a pending patent with Pendar Technologies.

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DEVELOPMENT OF A LEVAN-BASED FORMULATION FOR EFFICIENT SPERM CRYOPRESERVATION

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Cryopreservation is widely used in scientific, commercial, and therapeutic applications to store cells, tissues, and organs. While sperm cryopreservation is a common practice in assisted reproductive technology (ART), the process can damage the sperm membrane, leading to poor motility and irregular morphology. To address the issue, this study explored the use of Levan polysaccharide to minimize sperm loss before and after preservation. Levan is an uncommon non-structural polysaccharide found in most microorganisms and a few varieties of plants, which consists of residues of β-D-fructofuranose connected by β-2,6 glycosidic bonds and occasional β-(2,1) fructosyl-fructose branching. Levan's biocompatibility, biodegradability, and biological activity make it a good candidate for biomedical use. The study investigated the cryoprotective capabilities of fructan formulations on human sperm samples by firstly evaluating the effect of four