

Magic Angle Spinning MRS to Evaluate Cardiac Preservation Prior to Transplantation

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Introduction

Heart transplantation has become a life-saving treatment for end-stage cardiac disease. Its application is limited by the availability of donor hearts and the brief ischemic tolerance of preserved myocardium. Standard cold storage limits preservation intervals to 4–6 hours. Recent advances suggest perfusion preservation with oxygenated storage solution may extend the storage interval and permit utilization of marginal donor hearts [1]. An inherent limitation of this technique when applied to long-term storage or use of suboptimal hearts is the difficulty in predicting post-transplant reperfusion function prior to implanting the organ. We previously utilized ¹H magnetic resonance spectroscopy (MRS) of cardiac extracts to determine important metabolic differences between hearts preserved under static and perfused conditions [2]. The tissue manipulation and time required to perform this analysis limit its clinical application. Magic angle spinning (MAS) MRS has been used to evaluate metabolism in intact tissue [3]. We hypothesized that ¹H MAS MRS could be used to rapidly determine the metabolic status of preserved myocardium. If successful, this would offer the potential to allow assessment of cardiac preservation success in a clinically relevant time interval. To test this possibility, we studied differences between standard static storage and perfusion storage in a large animal model of cardiac transplantation.

Methods

Hearts from adult mongrel dogs were harvested after cardioplegic arrest and stored at 4°C in Celsior organ preservation solution. Animals were randomized to either conventional static storage in an extracellular preservation solution (n=5) or perfusion preservation (n=6) in a device (Lifecradle®, Organ Transport Systems, Inc) that delivers the same solution to the coronaries at a continuous rate of 10 mL/100g/min. After 4 hours of hypothermic storage, small cylindrical left ventricular biopsies (approx 10 mg) were taken and immediately stored in liquid nitrogen at -80°C. This tissue was then placed in a 50 µl volume rotor with 5 µl of D₂O as a lock solvent. ¹H MAS was carried out with a gHx nanoprobe on a Varian 14.1T Vnmrs spectrometer. Spectra were acquired with the “presat” pulse sequence using a 1s delay, a 3.2s water presaturation pulse, and a 1.8s acquisition time for a T_r of 6s. The data were imported into the R statistical computing environment and evaluated with a peak-picking, partial least squares (PLS) algorithm to identify biomarkers that were indicative of the storage method.

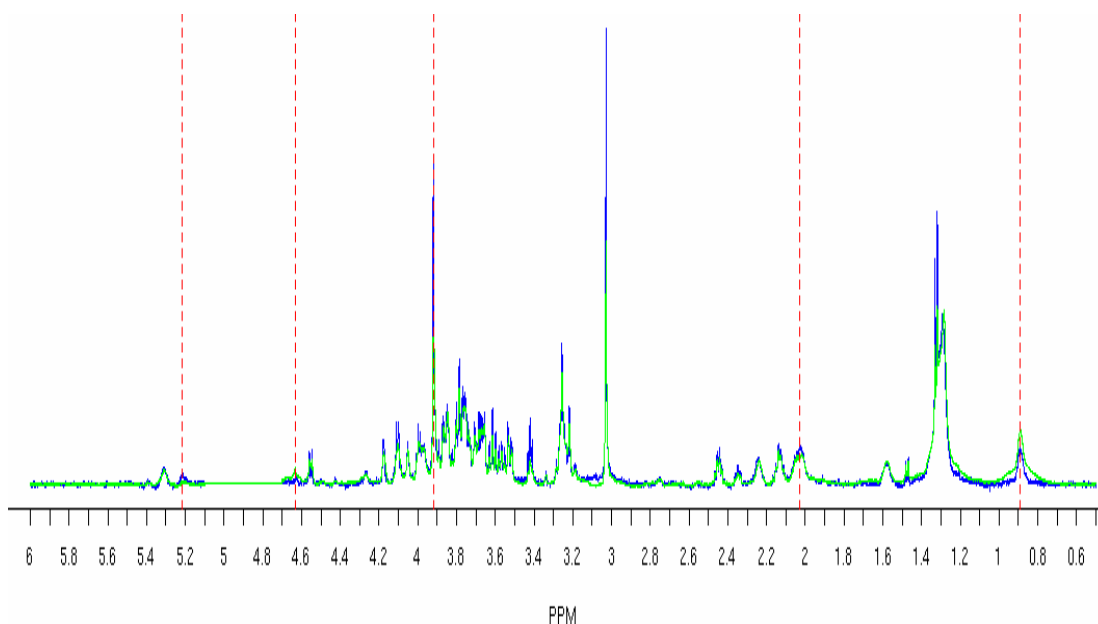


Figure 1. Average spectra for the static preservation (green) and perfusion preservation (blue) samples. Vertical bars mark the resonances chosen by the PLS algorithm for classification purposes.

Conclusions

These data demonstrate that ¹H MAS MRS can accurately identify metabolic differences that arise from varying cardiac preservation methods. Further studies will be required to test whether this technique can identify pre-implantation metabolic predictors of myocardial function after reperfusion under conditions where greater myocardial injury is more likely. The ability to use minute tissue samples and obtain metabolic data in short time intervals may make this technique an attractive option for cardiac transplantation, and may provide clinicians with important information on the status of a donor heart without deleterious extension of the ischemic interval.

References

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Results

Total analysis time for each sample was less than ten minutes. The peak-picking PLS algorithm identified 5 different resonances at .89 (triglyceride methyl's), 2.02 (protons alpha to a triglyceride double bond), 3.92 (creatine), 4.63 (unassigned), and 5.22ppm (glycerol protons) that were sufficient to categorize the spectra as either static storage or perfusion preservation with 100% efficiency. Of note, the algorithm selects the minimum number of resonances that can accurately classify the spectrum. Exclusion of a peak does not mean that significant differences are not present for that metabolite. Other compounds that are significantly different between the two preparations included lactate and myo-inositol.