Myocardial Ca2+ Influx Increases in Response to Hyperglycemia

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Introduction

One of the most common causes of morbidity and mortality in patients with diabetes mellitus include cardiovascular complications [1]. Streptozotocin injected rodents are a common model of diabetes mellitus and mimics some features of the human disease state. Indeed, altered L-type Ca^{2+} channel currents, sarcoplasmic reticulum Ca^{2+} uptake, Na^{+}/Ca^{2+} exchanger and sarcolemmal plasma membrane Ca^{2+}-ATPase pump may contribute to cardiac contractile deficits that have been reported in the rodent model. Manganese-enhanced MRI (MEMRI) has been shown to reflect the rate of calcium influx into the heart via voltage-gated channels [2]. Studies also indicate manganese ion (Mn^{2+}) is an intracellular contrast agent, and consequently, changes in signal enhancement due to Mn^{2+} infusion demonstrate intracellular calcium levels [2,3]. In the present work, these unique advantages offered by MEMRI were used to investigate the *in vivo* effects of hyperglycemia on myocardial Ca^{2+} influx in a mouse model of diabetes mellitus.

Methods

Streptozotocin induced diabetic and control C57/bl6 mice were used for this study. Blood glucose levels were determined utilizing a kit obtained from Sigma Aldrich (St. Louis, MO). All images were acquired on a 9.4 T Bruker magnet system. A 117.96 mM manganese chloride solution was prepared by dissolving solid $MnCl_2$ in stock saline. Manganese infusion was performed intravenously through the tail vein, using a syringe pump to deliver manganese chloride solution at a rate of 0.2 mL/hr for a total volume of 0.1 mL $MnCl_2$ and 0.05 mL saline. ECG electrodes were attached to the paws to gate signal acquisition with the end diastolic moment. Temperature was maintained at 37° C, and Isoflo kept at 2.5% during imaging. Images were acquired with Fast Low Angle SHot (FLASH) sequence (256x128, TR = 23.6 ms, TE = 1.4 ms, FA = 30 degrees, slice thickness = 1.0 mm, 3.0 cm FOV) for 80 repetitions over an experimental time course of approximately 90 minutes. Image analysis was carried out in Paravision v. 3.0.2. Signal intensities were calculated from a region of interest defined in the Left Ventricular Wall and normalized to an external water phantom. Signal intensities were then averaged for both pre-manganese and post-manganese steady states in each mouse, and the percent enhancement was calculated.

Results:

Our data indicate that on average, there was up to a 20% increase in signal intensity in control mice [Figure 1, blue squares]. In the diabetic mice with a blood glucose level ranging from 200 – 250 mg/dl, we observed that there was not a difference between this group of mice and control mice [Figure 1, red squares, blue arrow]. However, we also observed that as the blood glucose levels increased to > 400 mg/dl, the percent enhancement dramatically increased to 40-50% [Figure 1, red squares/red arrow].

Figure 1:

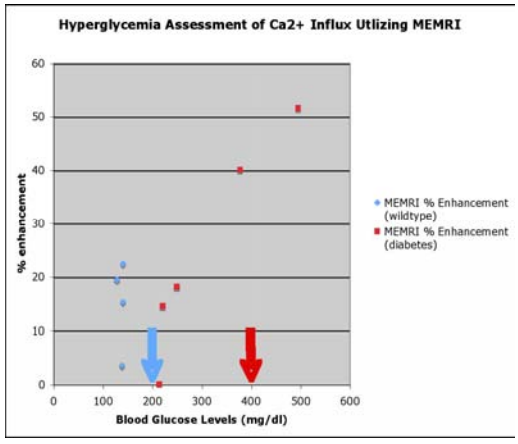
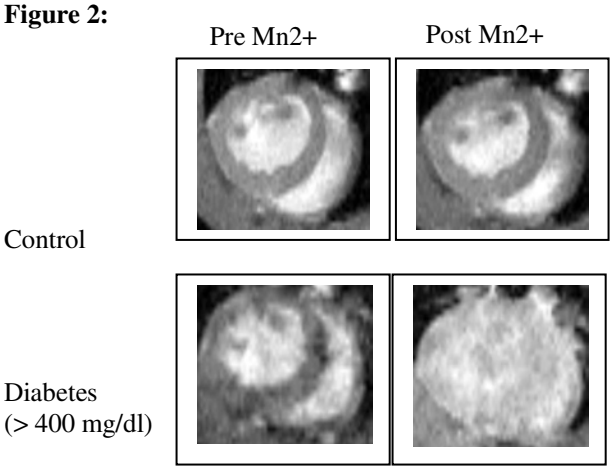


Figure 2:



Discussion: Our data demonstrate a relationship between hyperglycemia and increased Ca2+ influx in a mouse model of diabetes. At blood glucose levels less than 250 mg/dl, we did not observe any significant differences. However, at blood glucose levels above 400 mg/dl, we observed a magnified enhancement, indicative of increased Ca^{2+} influx in this group of mice. From these data, it is not clear, however, if the increased Ca^{2+} influx is occurring through L-type-Ca^{2+} channels or through the Na^{+}/Ca^{2+} exchanger. Additional experiments are underway to address this question.

References

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