TWO LIPID POOLS DETECTED IN THE MYOCARDIUM OF RATS FED A HIGH FAT DIET

Vijayasarathi Nagarajan[1], Philip William Kuchel[1], and S. Sendhil Velan[1]
[1]*Laboratory of Molecular Imaging, Singapore Bioimaging Consortium, Singapore, Singapore, Singapore*

Introduction: Excess free fatty acids are deposited in liver, skeletal muscle, abdomen, and heart. Myocardial triglyceride stores are increased in obesity, impaired glucose tolerance, and Type 2 diabetes (1,2). In particular the intramyocellular lipid (IMCL) present in skeletal muscle has been well correlated with insulin resistance and diabetes (3). Evaluation of lipid accumulation in the myocardium by magnetic resonance imaging (MRI) and spectroscopy (MRS) has significantly improved due to the current generation of MR technology in RF coils and pulse sequences. Recently the presence of two lipid pools, including IMCL and extramyocellular lipid (EMCL) was demonstrated in patients with muscular dystrophy (4,5). In the present study, we investigated myocardial lipids in rats fed a high fat diet and a control diet, with assessment of left ventricular function.

Methods: Control group (n = 5) received placebo diet (Clea, Japan, CE2) from 4 weeks of age. The high-fat diet group (n = 5) was fed with a high calorie diet (Research Diet, D12079B) from 4 to 15 weeks of age to induce obesity and insulin resistance. All experiments were approved by the local Biological Resource Center. At the age of 15 weeks rats were subjected to MRS and MR imaging. During this period body weight, food intake, blood glucose, triglyceride, cholesterol, and insulin levels were measured. Estimations of myocardial fat and left ventricular function were performed by MRI/MRS techniques. All the measurements were performed on a 7 Tesla Bruker ClinScan using a four-channel cardiac coil. CINE imaging was performed with a FOV of 50 mm and a matrix size of 256. Cardiac- and respiratory-gated long- and short-axis images of the heart were acquired for functional evaluation and MRS localization. The left ventricular function was calculated by using a custom developed MATLAB program. Localized PRESS experiments were performed on the septum of the myocardium with a voxel size of $3 \times 3 \times 3$ mm^3 and TR = 4.0 s, TE = 13 ms, SW = 3500 Hz. The resonances from the methyl or methylene ^{1}H atoms of (phospho)creatine (Cr), trimethylamine (TMA), IMCL and EMCL were analyzed by jMRUI software using the AMARES algorithm (6).

Results: Figure 1 shows the frames from cine MRI images (sagittal, coronal and transverse) for localization of the MRS voxel. Figure 2a (upper trace) shows the MR spectrum obtained from the myocardium of a rat fed with the control diet. All spectra from these animals displayed only one set of lipid resonances including CH_3 (0.85 ppm), and $[CH_2]n$ at (1.32 ppm). Figure 2b shows the localized MR spectrum from the myocardium of a rat fed with the high fat diet. The myocardial spectra from these animals shows two sets of lipid resonances including CH_3 from IMCL at 0.85 ppm, CH_3 from EMCL at 1.05 ppm, $[CH_2]n$ from IMCL at 1.3 ppm, $[CH_2]n$ from EMCL at 1.5 ppm. The estimation of fat content (%) relative to the unsuppressed water signal is shown in Figure 3. The IMCL and EMCL of rats fed with the high fat diet (IMCL and EMCL) were significantly higher than the IMCL in control rats. The left ventricular mass was significantly higher ($P < 0.005$), and the ejection fraction was significantly lower ($P < 0.005$) in rats receiving the high fat diet, compared to rats on the control diet. The triglyceride content in blood plasma was three fold higher in rats fed with the high fat diet.

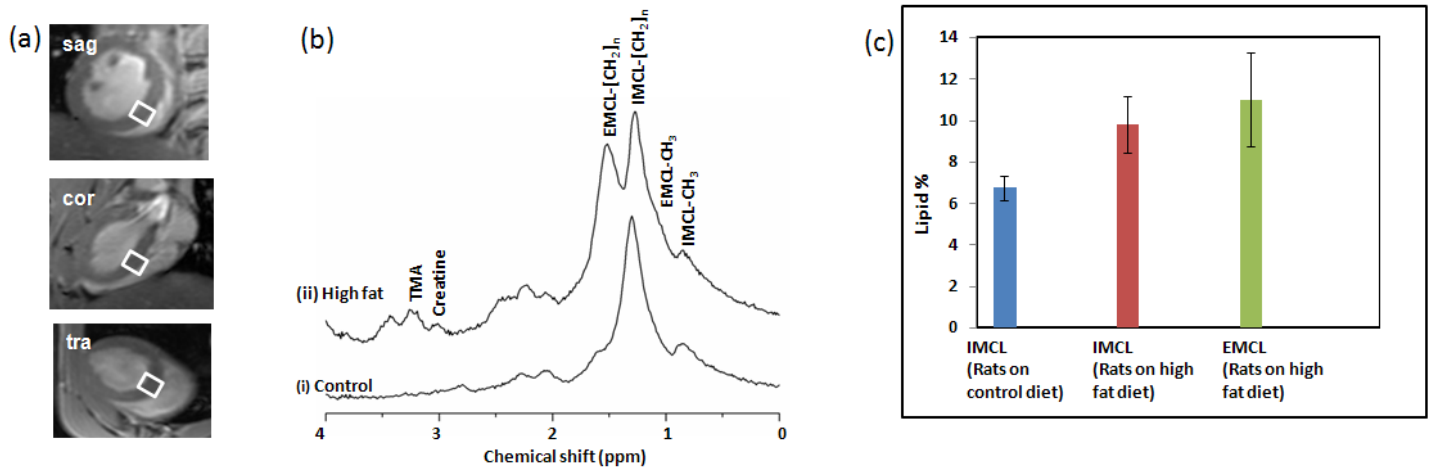

Figure 1. (a) MRI cine images (sagittal, coronal and transverse). (b) Localized MRS spectra from myocardium of rats under the control diet (i) and high fat diet (ii). (c) Lipid (%) in rats under control and high fat diet.

Discussion: We assessed the relative amounts of myocardial lipid in rats fed with a control diet and a high fat diet. We observed two distinct lipid pools in the myocardium of rats fed with the high fat diet. The detection of two lipid pools in skeletal muscle is made possible by differences in the bulk susceptibility between lipid droplets and the surrounding medium, and the differences in the geometry of the droplets in the two compartments (3) whereas, the observation of two sets of lipid resonances in myocardium was only recently described (4), in patients with muscular dystrophy. In the present work we report the first observation, to our knowledge, of two lipid pools in rat hearts fed with a high fat diet. The infiltration of fat as elongated cylinders between the muscle fibers results in separate MR resonances from the IMCL and EMCL. Rats receiving the high fat diet showed significant increases in the IMCL and a reduction in the ejection fraction of the heart.

Conclusion: We evaluated the myocardial fat content in rats fed with a placebo and a high fat diet. We demonstrated the presence of two lipid pools in the myocardium of the rats with obesity induced by the high fat diet. The left ventricular function was significantly reduced in rats fed with the high fat diet. The intensity of the IMCL resonance may be a potential marker of myocardial steatosis and could potentially be an early marker of Type 2 diabetes mellitus.

References

[1] Hansen, P. A, J. Biol. Chem (1998), **273**, 26157–26163.
[2] Park, S. Y et al. Diabetes (2005), **54**, 3530–3540.
[3] Boesch, C et al, Proc Nutri Soc (1999) **58**, 841-850**.**
[4] Rial. B et al, Proc. Intl. Soc. Mag. Reson **.** Med (2011)**, 19**,1360.
[5] Doblas. S et al, Proc ISMRM (2010), 173, 2628.
[6] Vanhamme. L et al, J Magn Reson (1997). **129** (1), 35.