Characterization of a Rodent Model of Metabolic Syndrome by MRS and MRI

D. H. Johnson[1], C. A. Flask[1,2], D. P. Wan[3], P. R. Ernsberger[3], and D. L. Wilson[1,2]

[1]Biomedical Engineering, Case Western Reserve University, Cleveland, OH, United States, [2]Department of Radiology, University Hospitals of Cleveland, Cleveland, OH, United States, [3]Department of Nutrition, Case Western Reserve University, Cleveland, OH, United States

Abstract

Fat distribution is characterized in a rat model of metabolic syndrome (diabetes, hypertension, obesity, and dyslipidemia) using MRI and localized ^{1}H MRS. Measurements include visceral and subcutaneous fat volumes and intramyocellular lipid, all in spontaneously hypertensive rats, which are lean, genetically obese, or dietary obese. Visceral adipose tissue makes up a fourfold larger proportion of body volume in the genetically obese rats relative to lean controls along with an elevenfold increase in IMCL, correlating with a tenfold elevation of insulin resistance. A sucrose-supplemented diet causes the lean controls to gain twice the visceral fat as compared to subcutaneous fat, an obesity phenotype indistinguishable by body weight.

Introduction

Magnetic resonance spectroscopy can measure muscle lipid content, which correlates with insulin resistance [1]. We have studied a specific rat model of metabolic syndrome, the Koletsky rat (SHR/SHROB), which provides insight into both dietary and hereditary obesity. The Spontaneously Hypertensive Rat (SHR) was crossed with a Wistar rat to yield SHROB, a rat predisposed to obesity and the comorbidities of metabolic syndrome. SHR is also susceptible to gaining weight on a high-sucrose diet, giving dietary obesity (SHR-DO). Previous research has established that SHR-DO has increased insulin resistance and hypertension as compared to SHROB, even when they weigh less. This suggests that dietary obesity may actually be worse than hereditary obesity [3, 4]. Elevated subcutaneous adipose tissue of SHROB is a unique feature of this rodent model, which may provide insights into human hereditary obesity.

Methods

Nine SHROB, six SHR-DO, and six non-obese SHR animals were chosen to cover a variety of ages and body weights for measurement of adipose tissue depots and total body volume and weight. Rats were anesthetized with up to 2.5% isoflurane and restrained within a phased-array receive coil. High resolution, T1-weighted coronal images were acquired with a spin echo sequence (TR/TE = 1240/13ms, resolution = 0.78x0.78x2mm, matrix = 256x128) on a clinical scanner. Each image was manually segmented by tracing the abdominal wall and applying a threshold to separate muscle and organs from adipose tissue. Intramyocellular lipid (IMCL) measurements were obtained for tibialis anterior and gastrocnemius lateralis muscles in SHROB (N=2, 113±6 days old) and SHR (N=2, 105±15 days old) on a Bruker Biospec 7T/30cm small animal MRI scanner. High-resolution T1 weighted images and MR ^{1}H spectra and were acquired using actively decoupled volume coils for both transmitting and receiving. Muscle fibers were aligned with the main magnetic field by carefully positioning the animal. Contrast between fat and muscle was maximized in T1 weighted images used for guidance with TR/TE = 1021/9.8 ms. Spectra were acquired with water suppression, TR 2000 ms, TE 21.5 ms, 2048 points, 512 averages, and at most a 1.5x1.5x7 mm voxel. Voxel placement minimized overlap with major vasculature and intramuscular fat deposits. The area under the IMCL aliphatic peak (1.3 ppm) was normalized to area under the total creatine peak (3.0 ppm) [2]. Insulin resistance was measured by blood chemistry in a separate cohort of SHROB and SHR.

Results

Differences in adipose tissue distribution are readily apparent by visual inspection of T1-weighted images (Fig 1). After image segmentation, subcutaneous adipose tissue normalized to body volume was significantly higher in SHROB (37.1%±7.4%, mean ± SD) than SHR-DO (6.8%±1.3%, P<0.001) or SHR (2.3%±0.9%, P<0.001, Fig 2 left). Visceral adipose tissue normalized to body volume was higher in SHROB (17.1%±2.3%) than SHR-DO (12.7%±2.8%, P<0.001) or SHR (3.8%±1.3%, P<0.001, Fig 2 right). SHR-DO visceral adipose tissue normalized to body volume was also significantly higher than SHR (P<0.005). Repositioning a single SHROB thrice did not affect subcutaneous or visceral adipose tissue normalized to body volume (P>0.75 and P>0.63, resp.). All statistical tests used Tukey's HSD. Repeated IMCL measurements in a SHROB were consistent in both tibialis anterior and gastrocnemius lateralis muscles (data not shown, P>0.50). SHROB gastrocnemius IMCL normalized to creatine (9.0±4.9) was 11-fold higher than SHR (0.8±0.3, P = 0.01, Fig 3). Insulin resistance was apparent in SHROBs, as fasting insulin was increased relative to SHR littermates (8.4 +/- 0.87 vs. 0.81 +/- 0.11 ng/ml, P<0.001 by t-test) while fasting glucose was unchanged (84 +/- 7 vs. 73 +/- 4, P>0.10). Glucose to insulin ratio, an index of insulin resistance, was 9-fold different between SHROB and SHR (10 +/- 1 vs. 90 +/- 5 mg/ng).

Discussion

Clear differences are apparent across the diverse cohort (ie, age, sex, and diet) of rats that were studied. Body weight can be used as a phenotype, but the ranking is almost invariably SHROB > SHR-DO > SHR. MRI and MRS provide new physiological information because body weight belies fat distribution. The dietary supplement of sucrose (SHR-DO vs. SHR) affects visceral much more than subcutaneous adipose tissue, which is consistent with literature reports regarding diet and muscle insulin resistance [5]. IMCL has been implicated in insulin resistance and aging [1]; the elevenfold elevation of SHROB IMCL observed here may explain the extreme (9-fold increase) insulin resistance of SHROBs, which becomes up to 18-fold in senescence. In conclusion, the phenotypes observed here may give new insights into human obesity due to the unique contrast of diet and heredity in SHR and SHROB.

References

[1] Krssak et al. Diabetologia 1999 Jan, 42(1):113-6. [2] Szczepaniak et al. Am J Physiol 1999 May, 276(5 Pt 1):E977-E989. [3] Ernsberger et al. Am J Hypertens. 1988;1:153S-157S. [4] Johnson et al. Obes. Res. 2005; 13:A113-A114. [5] Kim et al. Am J Physiol Regul Integr Comp Physiol 2000 279(6):R2057-65.

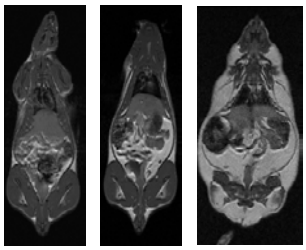

Figure 1. T1-weighted images of SHR, SHR-DO, and SHROB, left to right, resp.

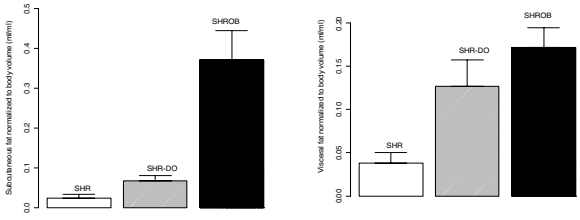

Figure 2. Subcutaneous and visceral fat normalized to total body volume.

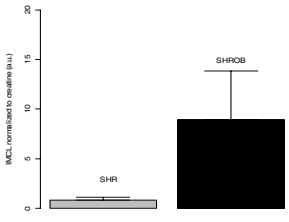

Figure 3. IMCL normalized to total creatine, SHR vs SHROB.