Effects of Resistance Trainging and β-hydroxy-β-methylbutyrate (HMB) on Muscle Fiber CSA and Lean Body Mass in Aged Rats: A DTI and DEXA Study

I. S. Masad[1,2], Y-M. Park[3], S-R. Lee[3], J. M. Wilson[3], P. C. Henning[3], B. H. Arjmandi[3], S. C. Grant[1,2], and J-S. Kim[3]
[1]Department of Chemical & Biomedical Engineering, The Florida State University, Tallahassee, FL, United States, [2]National High Magnetic field Laboratory, Tallahassee, FL, United States, [3]Department of Nutrition, Food & Exercise Sciences, The Florida State University, Tallahassee, FL, United States

Introduction:

Aging mammalian skeletal muscles exhibit sacropenia, which includes loss of muscle mass and strength. Estimates indicate that approximately 45% of older Americans are sarcopenic [1]. The healthcare cost of sarcopenic patients in the United States of America was estimated at $18.5 billion in 2000 [2]. Diffusion tensor imaging has shown high accuracy and sensitivity to study muscle architecture [3, 4] and microstructure [5, 6]. In this study, the effects of β-hydroxy-β-methylbutyrate (HMB), a dietary supplement known to promote muscle strength and lean body mass (LBM) [7, 8], were investigated in a pre-clinical model of aged rats during resistance training (RT).

Methods:

Experimental setup: Sixteen 19-month-old Sprague-Dawley female rats were divided randomly into three groups: BL (Baseline), HMB (0.46 g/kg b.w./d) and Non-HMB. HMB and Non-HMB groups underwent intense resistance training (weighted ladder climbing) every third day for 10 weeks. Animals were perfusion fixed using 4% paraformadehyde (PFA) and a trans-cardial procedure, after which the gastrocnemius and soleus muscles were harvested and directly immersed in 4% paraformadehyde. The fixed muscle tissues were washed with phosphate buffered saline (1xPBS) at least one day prior imaging, and immersed in fresh 1xPBS for scanning.

Imaging protocol: DTI images (7-noncollinear gradient directions) were acquired using a widebore 11.75-T vertical magnet with a Bruker Avance console and Micro2.5 gradients. Using a 15-mm birdcage coil, spin-echo (SE) DTI scans were acquired with b values of 0, 500 and 1000 s/mm^2 at in-plane resolution of 50×50 μm^2, with a slice thickness of 500 μm. The DTI acquisition parameters were as following: TE = 20.5 ms, TR = 2.75 s, Δ = 12.7 ms and δ = 2.1 ms. Also, a high resolution (40 μm^3) 3D gradient-recalled echo (GRE) image was acquired (Fig.1) (TE/TR= 10/150 ms) for anatomical and volumetric measurements. *In vivo* and prior to sacrifice, pre- and post-RT LBM was assessed by dual energy X-ray absorptiometry (DEXA).

Data Analysis: After acquisition, the images were processed by MedINRIA software to calculate the diffusion tensor, providing the parameters fraction anisotropy (FA), apparent diffusion coefficient (ADC) and eigenvalues (λ_1, λ_2 and λ_3). A region of interest (ROI) was chosen in the widest region of the soleus muscle for processing as shown in Fig. 2. Tukey's HSD test was used to determine if there were any statistical differences between groups. The statistics was performed using SPSS 17.

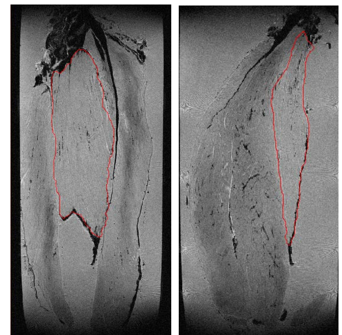

Figure 1: Coronal (left) and sagittal (right) views of a 3D GRE image of the rat skeletal muscle (soleus marked in red)

Results and Discussion:

Diffusion tensor imaging of the soleus muscle showed no change in the principal eigenvalue (λ_1) while λ_2 and λ_3 increased (+17% and +20%, respectively) significantly ($p<0.05$) in both groups after RT, indicating a significant increase in cross sectional area (CSA) of the muscle fiber (data is shown in Fig. 3 below). The increased CSA also was evident in the decreased FA (-30%) and increased ADC (+13%). The findings suggest that the RT muscles are either swollen or contain hypertrophic myofibers due to resistance training. As assessed by DEXA, RT also improved LBM (+21%, $p<0.05$) in both HMB and Non-HMB groups. However, no between-group differences (HMB v. non-HMB) were identified for all variables tested.

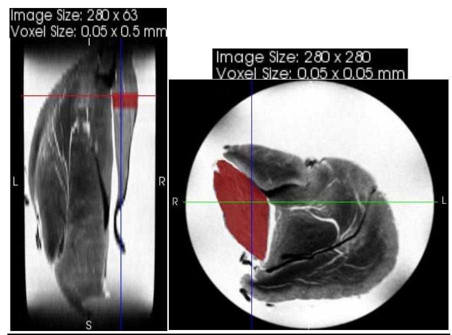

Figure 2: Sagittal (left) and axial (right) B0 images of soleus muscle with the processed ROI drawn in red

Conclusions:

Results indicated that HMB failed to facilitate the changes in LBM and soleus CSA during high intensity RT. However, an RT-induced increase in the CSA of the soleus muscle was identified in addition to associated changes in FA and ADC when compared to the baseline group, which indicates that at least RT played a role in muscular maintenance with age. Further corroborative studies are underway to determine the effect of age on muscle fibers and impact of HMB on sedentary conditions. Future analysis also will evaluate the effects of HMB and RT on the gastrocnemius muscle.

Acknowledgments:

All MR data were collected at the Dept. of Chemical & Biomedical Engineering, FAMU-FSU College of Engineering; DEXA data were collected at the Dept. of Nutrition, Food & Exercise Sciences, College of Human Sciences (CHS), The Florida State University, Funding provided by: NSF (IOS-0718499 to SCG), FSU (CRC-MDS to SCG/BHA/JSK, CRC-PG and CHS RIAP to JSK) and NHMFL (NSF DMR-0084173).

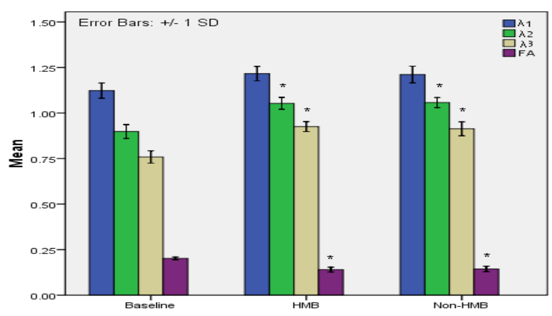

Figure 3: Comparison soleus muscle DTI data between experimental groups. * indicates a significant difference from the baseline group ($p < 0.05$)

References:

[1] Melton et al.; J Am Geriatr Soc 2000; 48: 625 -630
[2] Janssen et al.; J Am Geriatr Soc 2004, 52:80-85
[3] Damon et al.; Magn Reson Med 2002; 48: 97-104
[4] Heemskerk et al.; Magn Reson Med 2005; 53: 1333-40
[5] Galban et al.; NMR Biomed 2005; 18: 489-98
[6] Heemskerk et al.; Magn Reson Med 2006; 56: 272-81
[7] Knitter et al.; J. Appl. Physiol. 2000; 89: 1340–1344
[8] Panton et al., Nutrition 2000; 9: 734–739