Inverse correlation between IMCL content of the human calf muscle and *local* glycogen synthesis rate

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Introduction

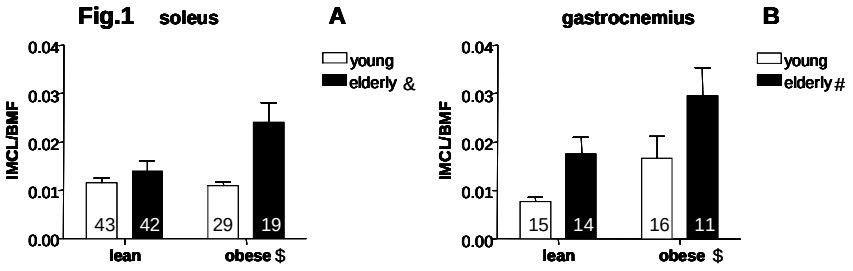
Intramyocellular lipid (IMCL) content of skeletal muscle, as measured with ^{1}H MRS, is inversely correlated with insulin sensitivity as determined by whole body glucose uptake [1]. The latter, however, does not necessarily represent the actual glucose uptake in the corresponding skeletal muscle [2]. In this study, we examined whether IMCL content in human calf muscle correlated with local glucose uptake assessed by measurement of glycogen synthesis rate within the same muscle compartment.

Methods

Subjects: We studied twenty subjects belonging to four subgroups of five persons each (mean±SD): young lean (3F/2M, age: 22.2±5.5 yrs, BMI: 22.5±1.4 kg/m^2), elderly lean (3F/2M, age: 57.4±3.9 yrs, BMI: 22.8±3.4 kg/m^2), young obese (4F/1M, age: 24.6±2.2 yrs, BMI: 30.8±3.0 kg/m^2) and elderly obese (4F/1M, age: 56.8±5.1 yrs, BMI: 36.2±3.8 kg/m^2). Approval of the local ethics committee was obtained and the volunteers gave written informed consent.

Methods: After a standardized fasting period IMCL content in the soleus and gastrocnemius muscle was determined in a 10-mm transversal slice positioned 15 cm below the knee by ^{1}H MR spectroscopic imaging using a standard extremity coil. ^{1}H MRSI was performed with a STEAM sequence (TR/TE/TM 1000/20/30 ms), CHESS water suppression, 32x32 steps of phase encoding and a FoV of 160x160mm^2 resulting in nominal voxels of 5x5x10mm^3. Then, local glycogen synthesis rate in the calf muscle was measured by ^{13}C MRS using a 13-cm concentric ^{13}C surface coil with a circularly polarized ^{1}H coil of 2 x 15 cm in diameter for decoupling and shimming [3] during a 120-min. euglycemic hyperinsulinemic clamp with 20% w/v 30% ^{13}C-1-labeled glucose infusion. Unlocalized ^{13}C MR spectra were obtained in 15-min blocks consisting of 5000 scans using an adiabatic 2560-µs excitation pulse and a repetition time of 180 ms. During the first 60 ms of the acquisition period continuous wave decoupling at 26W was applied. The combined MR examination was performed on a clinical 1.5T whole body MR system (Siemens, Erlangen, Germany).

Data processing and statistical analysis: After application of a Hanning filter and Fourier transformation in the spatial domain, signals of IMCL and EMCL in the ^{1}H MRSI spectra were fitted in the time domain using the MRUI software package [4]. Only spectra with both signals clearly separated by 0.2 ppm were included in the further analyses. IMCL signal integrals were quantified relative to the signal intensity of bone marrow fat (BMF) and expressed as IMCL/BMF [5]. Quantification of the ^{13}C-1-glycogen signal at the start of the clamp was performed by calculating the contribution of signal distribution over the skeletal muscle to the final spectrum from MR images and an RF B1 fieldmap [6] using a reference phantom. Increments in muscle glycogen concentration were calculated from the change in ^{13}C-1-glycogen signal integral after the start of ^{13}C-1-glucose infusion and the plasma ^{13}C-1-glucose fractional enrichment [2]. The rate of muscle glycogen synthesis (µmol/kg muscle/min) was calculated from the slope of a least-squares linear fit to the glycogen concentration curve. Statistical analyses were performed using Graphpad Prism (San Diego CA, USA). Differences between groups were analyzed using a two-way ANOVA with factors for age and obesity. If no interaction was present, Bonferroni post-testing was applied. Differences between muscles within a group were statistically analyzed using a Student's t-test. Correlations were calculated using Spearman's rank correlation. Data are expressed as mean±SEM unless otherwise indicated.



Results

Relative IMCL contents in soleus and gastrocnemius muscles are presented for each of the four groups in Figure 1 with the total number of spectra selected per group indicated in the bars. Data on IMCL content could be determined for all volunteers except for one young obese and one elderly obese subject. For the soleus muscle, significant higher values for elderly (&, P<0.0001) and obese ($, P<0.01) were found, with a strong interaction (P<0.01). For the gastrocnemius muscle, the relative IMCL values are also higher for elderly (#, P<0.01) and obese ($, P<0.01), but without interaction and no significant differences between subgroups. Only for the young lean group differences between the two muscles were observed with a significantly higher relative IMCL content in the soleus than in the gastrocnemius muscle (P < 0.05). Local glycogen synthesis rates (Fig. 2) were significantly lowered for obese ($, P<0.01), and the rates obtained for elderly lean and elderly obese were also significantly different (**, P<0.01). Figure 3 shows that the average relative IMCL content for soleus and gastrocnemius together and the local glycogen synthesis rate in the calf muscle are inversely correlated (r_s = -0.50, P<0.05).

Discussion and conclusion

Although an inverse correlation between IMCL content and insulin resistance on the whole body level has been previously reported [1], this relationship has never been assessed within a single muscle compartment. As nearly all signal in the ^{13}C MR spectra of this study originated from the soleus (~30%) and gastrocnemius muscle (70%), the inverse correlation between average IMCL content in soleus and gastrocnemius muscle and the local glycogen synthesis rate (Fig. 3) shows that this relationship also exists within the same muscle compartment.

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