Rapid ^{13}C tip angle / coil loading calibration for *in vivo* natural abundance ^{13}C studies in humans

P. E. Thelwall[1]

[1]Newcastle Magnetic Resonance Centre, Newcastle University, Newcastle upon Tyne, Tyne and Wear, United Kingdom

Introduction

^{13}C spectroscopy of the human body can provide insight into metabolic processes in a variety of organs and tissues (1,2). Natural abundance ^{13}C spectroscopy has been employed to measure the concentration of glycogen in tissues (2,3), allowing interrogation of energy storage and metabolism in normal subjects and with pathology. Surface coils are frequently used, but acquistion can be confounded by differences in subject morphology. The thickness of the subcutaneous fat layer alters coil loading and thus coil power requirements due to differences in conductivity between lipid and muscle. Secondly, the thickness of the subcutaneous fat layer determines the separation of coil from sample of interest in muscle ^{13}C spectroscopy studies. Previous studies have demonstrated how *in vivo* spectra can be quantified with the use of external standards and phantoms containing metabolite solutions of known concentration (1,3). Although the use of an external standard can account for differences in coil loading between subjects, it is preferable to employ optimised acquisitions (ie optimal tip angles) for each subject studied to attain maximal SNR in these signal-limited studies. Performing a ^{13}C tip angle calibration using endogenous signals is difficult due to the low natural abundance of ^{13}C, so we have developed a protocol employing a ^{13}C-containing external standard for this purpose. We used a short-T$_1$ ^{13}C external marker (located within the coil housing) comprising an aqueous solution of gadolinium-doped 2-^{13}C acetone, which provides a single resonance separate from the other signals of interest in an *in vivo* ^{13}C spectrum. Calibration spectra were acquired prior to each *in vivo* experiment, from which the coil power requirements could be determined. This measurement, coupled with imaging-based measurement of distance between the coil and tissue of interest, allowed the tip angle in muscle tissue to be accurately selected regardless of patient morphology.

We have applied this protocol to our measurements of muscle glycogen content using natural abundance ^{13}C spectroscopy. Data from these studies were quantified by comparison to spectra acquired from phantoms shaped to the organ of interest, filled with glycogen solutions of known concentration. ^{13}C pulse-acquire spectra with the same TR and ^{1}H-decoupler settings as used for *in vivo* studies were acquired from the phantoms at a range of flip angles and separations between coil and phantom. The magnitudes of glycogen C1-^{13}C signal from these datasets were entered into a lookup-table for tip angle and coil-sample separation, and the appropriate phantom data employed for comparision with an individual's *in vivo* data.

Methods

Data were acquired on a 3T Achieva scanner (Philips, Best, The Netherlands) equipped with a PulseTeq leg ^{13}C/^{1}H surface coil (6cm diameter ^{13}C coil). An external marker was placed within the coil housing, comprising a 5 mL mixture of 2-^{13}C acetone (170 mM) and GdCl$_3$ (25mM) in water. The resultant short-T$_1$ acetone ^{13}C resonance was used to perform a rapid (2.5 minute) ^{13}C tip angle calibration, which was acquired prior to all *in vivo* spectra. Four ^{13}C spectra centred on the ketone resonance of acetone were collected with sequential doubling of ^{13}C pulse power, corresponding to approximate tip angles of 25, 50, 100 and 200º (TR = 150ms, NS=256, SW=8kHz, TD=256). The variation in signal amplitude between the four spectra allowed the discrepancy between expected tip angle and real tip angle to be determined, by comparison of these spectra to previously acquired calibration spectra. ^{13}C acquisitions to measure glycogen content (TR = 200ms, SW = 8 kHz, NS = 3000, TD = 256, ^{1}H WALTZ decoupling) were preceded by ^{1}H scout images from which the distance between coil and muscle was determined. The optimal ^{13}C pulse power for maximal glycogen signal was determined from the ^{13}C tip angle calibration and the separation between coil and muscle. ^{13}C spectra were also acquired from leg-shaped phantoms filled with 100 mM glycogen solution for quantitation purposes. Scans were acquired over a range of tip angles at distances of 0 – 40 mm separation between coil and phantom to simulate the range of subcutaneous fat layer thicknesses observed *in vivo*. The magnitude of glycogen signals in the *in vivo* spectra were quantified by comparison to spectra from the phantoms acquired with a matching coil-tissue distance and ^{13}C tip angle. Spectra were analysed using jMRUI software.

Results and Discussion

Figure 1 shows images from the thigh of two volunteers with different thicknesses of subcutaneous fat. The ^{13}C coil loading varied significantly between the two volunteers, the separation between the coil surface and muscle was 4 mm (A) and 35 mm (B). Figure 2 shows rapid ^{13}C calibration scans acquired under high (A) and low (B) coil loading. The discrepancy between expected and actual tip was determined by comparison of spectra with a calibration chart (C), calculated as -15% and +20% relative to calibration for the high and low coil loadings respectively, representing a ~40% change in tip angle with the same input power between the two loading configurations. The pulse power employed for acquisition of *in vivo* ^{13}C spectra could then adjusted to account for these coil loading effects. Figure 3 shows a typical ^{13}C spectrum acquired from the thigh of an endurance-trained athlete, acquired in 10 minutes. The glycogen C1 resonance can be clearly seen at 100.5 ppm (red circle). Comparison to phantom data indicated that the muscle glycogen content was 127 mM, which is in agreement with literature values of fasting muscle glycogen concentration (3,4). Performing a ^{13}C tip angle calibration and measuring the separation between coil and tissue permits an accurate quantitation of glycogen content despite variable coil loading and thickness of subcutaneous fat layers between subjects.

References and Acknowledgements

References: (1) Gruetter *et al*. NMR Biomed 16:313-338 (2003). (2) Carey PE *et al*. Am J Physiol Endocrinol Metab 284:E688-694 (2003). (3) Taylor R *et al*. Magn Reson Med 27:13-20 (1992). (4) Burke *et al*. J. Appl. Physiol. 75:1019-1023 (1993).
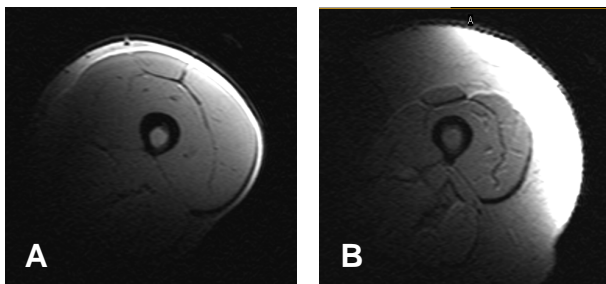
Figure 1 - Images of subjects with thin (A, 4 mm) and thick (B, 35 mm) subcutaneous fat layers between skin and muscle
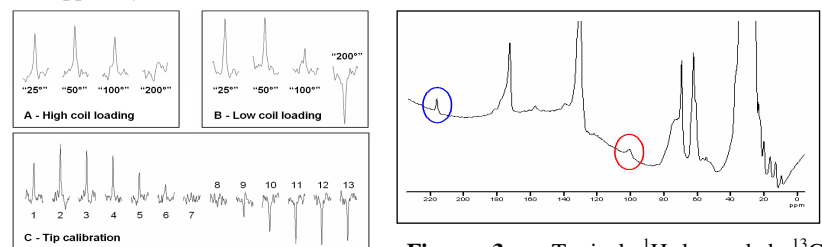
Figure 2 – Rapid ^{13}C tip angle calibration scans acquired under high (A) and low (B) coil loadings, and calibration data (C).
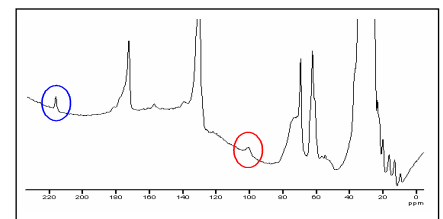
Figure 3 – Typical ^{1}H-decoupled ^{13}C spectrum acquired in 10 minutes from thigh muscle of a healthy volunteer. The acetone (216 ppm) glycogen (100.5 ppm) resonances can be clearly seen (circled).