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Associations of age, gender and body mass with ^1H MR-observed brain metabolites and tissue distributions

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Abstract

Recent reports have indicated that a measure of adiposity, the body mass index (BMI), is associated with MR-observed brain metabolite concentrations and tissue volume measures. In addition to indicating possible associations between brain metabolism, BMI and cognitive function, the inclusion of BMI as an additional subject selection criterion could potentially improve the detection of metabolic and structural differences between subjects and study groups. In this study, a retrospective analysis of 140 volumetric MRSI datasets was carried out to investigate the value of including BMI in the subject selection relative to age and gender. The findings replicate earlier reports of strong associations of *N*-acetylaspartate, creatine, choline and gray matter with age and gender, with additional observations of slightly increased spectral linewidth with age and in female relative to male subjects. Associations of metabolite levels, linewidth and gray matter volume with BMI were also observed, although only in some regions. Using voxel-based analyses, it was also observed that the patterns of the relative changes of metabolites with BMI matched those of linewidth with BMI or weight, and that residual magnetic field inhomogeneity and measures of spectral quality were influenced by body weight. It is concluded that, although associations of metabolite levels and tissue distributions with BMI occur, these may be attributable to issues associated with data acquisition and analysis; however, an organic origin for these findings cannot be specifically excluded. There is, however, sufficient evidence to warrant the inclusion of body weight as a subject selection parameter, secondary to age, and as a factor in data analysis for MRS studies of some brain regions.

Keywords

brain; ^1H MRSI; body mass index; magnetic field inhomogeneity

INTRODUCTION

In vivo ^1H MRS measurements in the brain enable the monitoring of metabolites that are indicators of altered tissue metabolism with disease or injury. In many cases, these changes are diffuse and subtle, and the detection of altered concentrations is greatly facilitated by comparison against normative data from a control subject group. As metabolite levels vary across brain regions, between gray (GM) and white (WM) matter and as a function of age, these factors are routinely taken into account in the analysis of individual subject or group

differences. Recent studies by Gazdzinski *et al.* (1–3) have indicated that there are also associations between brain metabolite concentrations and adiposity, as indicated by the body mass index (BMI). Findings based on a group of 50 subjects, aged 33–50 years (1), included decreased *N*-acetylaspartate (NAA) in GM and WM and decreased total choline (Cho) in frontal WM with increasing BMI. A study of 54 male subjects (3), aged 28–66 years, found decreased NAA, Cho, creatine (Cre) and myo-inositol in the frontal lobe, subcortical nuclei and cerebellar vermis. A study of 23 subjects, aged 62–76 years (2), found lower NAA/Cre and NAA/Cho ratios in the anterior cingulate cortex with increasing BMI. These findings indicate that it may be of value to include BMI as a variable in the comparison of MRS results between subjects, thereby refining the normative metabolite values based on individual subject characteristics.

As metabolite concentrations differ in GM and WM, the relative tissue content should also be taken into account for MRS analyses. Differences in GM and WM tissue distributions are known to occur with age and gender (4–6), and it has also been reported that BMI is associated with total brain volume (TBV) measures (7–10). For comparisons of relative brain tissue volumes between groups, it is therefore similarly of interest to determine whether BMI should be included as a subject selection criterion.

In this study, a retrospective analysis of a normative brain metabolite image database was carried out to examine whether the previously reported associations of MR measures were also applicable for volumetric MRSI studies carried out at 3 T. The existing MRSI data were acquired for the purpose of mapping brain metabolite distributions (11) and to provide control data for the evaluation of MRSI results from individual subjects as part of on-going clinical MRS research studies (12–14). A previous analysis of this normal subject MRSI data (11) found significant differences in brain metabolites with brain region, tissue type, age and gender. For this study, an analysis similar to that used by Gazdzinski *et al.* (1–3) was applied to examine the association of metabolite levels in GM and WM over lobar regions with BMI or body weight. This analysis also included measures of spectral quality, as well as 52 additional datasets collected since the previous report.

METHODS

MRI and MRSI studies of the brain were obtained from an existing database of 140 studies carried out at 3 T (Siemens Trio, Erlangen, Germany) with an eight-channel phased-array head coil. Of these, 88 were included in the previously reported analysis (11). These studies were of healthy subjects carried out under four research protocols, each approved by the institutional human subjects research review committee and with informed consent obtained from all participants. Participants were screened to exclude any history of brain disease or injury, substance abuse or psychiatric condition. Eight elderly subjects were screened using neuropsychological test procedures and verified to be cognitively normal, whereas self-reporting procedures were used for all other subjects. BMI was calculated as the weight (kg) divided by the height (m) squared. Subjects were typically weighed before the study and height was obtained by self-report. Individual metabolite images for three studies showed evidence of incorrect intensity scaling of unknown origin, and were therefore removed from the individual metabolite analyses, i.e. $n = 137$, but were retained for the metabolite ratio analyses.

MRSI data were acquired and processed using previously described methods (11,15). A volumetric spin echo acquisition was used, with echo planar readout, frequency-selective water suppression, lipid inversion nulling, 73° excitation and TE/TR/TI = 70/1710/198 ms. Data were sampled with $50 \times 50 \times 18$ points over a field of view of $280 \times 280 \times 180\text{mm}^3$ and with selection of a slab of 135mm covering the cerebrum. The MRSI acquisition

included a second dataset that was obtained in an interleaved manner without water suppression and using 20° excitation and gradient echo observation, and which provided a water reference signal with identical spatial parameters to the metabolite MRSI. The water reference MRSI was used for several processing functions, including measurement and correction of the resonance frequency offset Δf at each voxel location, correction of lineshape distortions and to provide the signal reference for the normalization of metabolite values. Each study also included a T_1 -weighted MRI (MPRAGE, magnetization prepared rapid gradient echo) at an isotropic resolution of 1mm (TR/TE/TI = 2150/4.4/1100 ms; flip angle, 8°), which was employed for tissue segmentation using FSL/FAST (16) and spatial registration.

Metabolite and water reference images were reconstructed in a fully automated manner using the MIDAS (Metabolic Imaging Data Analysis System) package (11,15). This included spatial smoothing and interpolation of the metabolite images to $64 \times 64 \times 32$ points, resulting in a voxel volume of approximately 1 mL. Spectra in voxels for which the water signal intensity linewidth was 15 Hz or less were analyzed using an automated parametric spectral analysis procedure (17) to determine volumetric maps of NAA, Cre and Cho. The spectral model used a Gaussian lineshape with a common linewidth for all resonances, based on the assumption that the *in vivo* lineshape is dominated by local magnetic susceptibility effects. The resulting images of the individual metabolites were signal intensity normalized to institutional units (i.u.) based on the tissue water signal derived from the water reference MRSI. For this procedure, each voxel of the water MRSI signal was first scaled to 100% water equivalence using information on the MRSI voxel tissue content, which was derived from the segmentation maps, and estimates for the tissue T_1 values (18,19) and water content (20). The T_1 values used were as follows: GM, 1350 ms; WM, 840 ms; cerebrospinal fluid (CSF), 4300 ms; the tissue water density factors were as follows: GM, 0.797; WM, 0.703; CSF, 0.98. The resulting 100% water-equivalent image was then smoothed to diminish the effect of local variations in T_1 and water density to provide the image used for metabolite signal normalization. Although variations in tissue water T_1 values and water density with age have been shown (20–22), the relative age-dependent changes appear to be smaller than variances across subjects, and have not been well characterized at 3 T; therefore, these values were assumed to be constant for all subjects. Metabolite ratio images were also generated for NAA/Cre and Cho/Cre.

Additional measures obtained included the Cramer–Rao bounds (CRBs) for each of the spectral model parameters and an estimate of the signal-to-noise ratio (SNR) derived from the peak value of the NAA resonance relative to the standard deviation of the noise. This latter measurement selected spectra with fitted linewidths within a narrower range of 6–10 Hz, and used a noise measurement obtained from a portion of the spectrum in the range of 0 to –0.4 ppm and a linear baseline estimate in the NAA resonance region to correct for the possible influence of lipid resonances.

Nonlinear registration to a standard spatial reference (23) at an isotropic resolution of 2mm was applied to the individual metabolite maps, the metabolite ratio maps and maps of the relative GM, WM and CSF volume contribution to each MRSI voxel. Using a brain atlas aligned to the spatial reference, all voxels within each of the left and right frontal, temporal, occipital and parietal lobes, and the cerebellum, were selected, and a linear regression analysis was carried out for each region against the relative WM voxel fraction, to obtain the metabolite measures corresponding to 100% GM and 100% WM. The CSF-corrected metabolite values were used for the individual metabolite images. Voxels were excluded from the analysis with a linewidth of greater than 12 Hz or a CSF volume fraction greater than 0.3. Outlying values were also removed if not within three times the standard deviation from the mean value, based on all voxels within the selected region. In addition, mean

values from all voxels in the cerebellum were obtained, without discrimination of GM and WM.

A multiple linear regression analysis was used to test associations of the metabolite parameters in each brain region for GM and WM with gender, age, BMI and interactions between gender and age, and gender and BMI. Initially, the interaction between age and BMI was also included, but this interaction caused co-linearity problems and was therefore eliminated. A backward elimination procedure was used to eliminate from the model interactions that were not significant at the 0.05 level; the interaction with the largest p value was removed first. Gender, age and BMI were always retained in the model in that order. Significance tests of the model parameters were based on sequential (type 1) sums of squares, so that the significance test for each successive parameter in the model indicates the significance of the additional variance explained after the preceding parameters were entered. As previous studies have shown that both age and gender are associated with brain metabolite concentrations (11), the variation caused by these variables was accounted for first to determine the additional effect of BMI. Using this approach, any effect of BMI in common with gender and age was removed, yielding an estimate of the independent effect of BMI. The R^2 values for the full model and the contribution of each parameter were calculated. An initial analysis performed for right and left for each brain lobe separately found significant differences in average metabolite levels, in agreement with a previous report (11); however, the regression of the metabolite parameters on gender, age and BMI was very similar, and therefore the results are presented for the average of the two sides. To evaluate the relative influence of adiposity to body weight alone, the analyses were repeated using age, gender and subject weight. SAS 9.2 (SAS Institute, Cary, NC, USA) was used for all analyses. The analysis results are presented for the associations with BMI to be consistent with previous reports, but the findings for the associations with weight are also summarized. All p values are presented without correction for multiple comparisons, and a value of $p < 0.05$ was used to determine statistical significance.

To test whether the sequential analysis approach could influence the significance of BMI or weight, secondary analyses were run using type 3 analysis, which does not depend on the order in which the terms for the model are specified, and, in addition, the sequential analysis was repeated using BMI as the first parameter. To also test for only the most significant findings, the results of both the sequential and type 3 analyses were re-examined with correction for multiple comparisons applied for all tests within each brain region (i.e. six parameters and for GM and WM), leading to a p value of <0.004 for significance.

To test for the possibility of spatially localized changes in metabolite parameters with any of the subject variables, a voxelbased linear regression analysis was performed. This procedure first applied Gaussian smoothing (full width at half-maximum, 8 mm) to the metabolite images, followed by the selection of data from all subjects and for each voxel location in turn, and then application of a linear regression analysis of the metabolite parameter against age, BMI or weight. The same voxel selection criteria as described previously were applied and, in addition, a minimum of 20 valid data points were required for the regression analysis to be applied. The resulting parameter maps were displayed using MRICro (www.mricro.com).

To further investigate and visualize the impact of BMI, age and gender on the spatial distributions of parameters that have an impact on the spectroscopic measurement, including Δf , spectral linewidth, CRBs and SNR, images of the mean values from different subgroups of subjects were compared. These included groups of subjects corresponding to the lowest and highest BMI values for a similar age range, female and male subjects for a similar age and BMI, and young and elderly subject groups for a similar BMI range. The subject

selections used are provided with the discussion of each test result. To compute the Δf , CRB and SNR maps, all voxels within the brain, as defined by the tissue volume fraction from the sum of the tissue segmentation maps being >0.4 , were used, whereas the spectral linewidth maps were calculated only for all voxels for which the linewidth of the water MRSI resonance was ≤ 15 Hz. Maps of the linear regression of Δf with BMI were also generated using data from all subjects, with smoothing of 8mm full width at half-maximum applied.

RESULTS

Figure 1 shows the age and BMI (kg/m^2) for the selected subject group, which consisted of 63 males and 77 females, aged 18–84 years (median, 36 years). The BMI values were in the normal range ($18.5 \leq \text{BMI} < 25$) for 75 subjects, 49 were overweight ($25 \leq \text{BMI} < 30$) and 16 were obese ($30 \leq \text{BMI}$). The median BMI was 25.2 for male subjects and 23.9 for female subjects. Linear regression analysis for the whole group indicated increased BMI with age ($1.12 \text{ kg}/\text{m}^2/\text{decade}$, $p < 0.0001$) and weight with age ($2.2 \text{ kg}/\text{decade}$, $p = 0.02$), which is consistent with previous studies (24).

The results of the linear regression analysis for age, gender and BMI are shown in Tables 1–3. The findings from the metabolite ratio measures generally followed the observations from the individual metabolites; therefore, the major findings are only summarized for the primary metabolite measures unless additional findings were found in the ratio measures.

Significant changes in metabolite concentrations with age were consistently found, with decreased NAA and increased Cre and Cho, in agreement with a previous analysis (11). Average regional linewidths also increased with age, except for frontal lobe GM. This finding is consistent with reports of increased brain iron concentration (25) and decreased water T_2^* (26) in several brain regions with age.

In males relative to females, all individual metabolite measures were decreased in frontal and temporal WM, and temporal and occipital GM, but increased in occipital WM. In the metabolite ratio measures, the gender differences remained in both the type 1 and type 3 analyses for decreased NAA/Cre in parietal GM and decreased Cho/Cre in temporal GM. Spectral linewidths were also decreased slightly (by ~ 0.1 Hz on average) in most regions in males relative to females, reaching significance in frontal, temporal and occipital WM, although with a significant increase of 1 Hz in the cerebellum for males.

Significant associations of individual metabolite levels with BMI were found, with decreased NAA and Cre in occipital WM and cerebellum, with males having a significantly greater decrease than females in occipital WM. Increased Cho/Cre with BMI was also found in parietal GM ($p = 0.039$). Higher BMI was also associated with increased linewidth, which was highly significant in frontal and parietal regions, although decreased in the cerebellum for male subjects only.

Tables presenting the results of the linear regression analysis for age, gender and body weight are provided in Supporting information. The regions and strengths of the associations were very similar to those presented in Tables 1–3. For example, Cho/Cre was significantly increased with weight in parietal and frontal GM ($p = 0.011$ and $p = 0.025$), and the regions with significant associations of increased linewidth with weight were the same as the corresponding associations with BMI, suggesting that the observed associations are not specifically associated with subject adiposity. The only additional finding in the analysis using weight was an indication of increased Cho in frontal GM ($p = 0.046$, $R^2 = 0.026$).

When accounting for multiple comparisons using a p value of <0.004 , significant associations with BMI remained for decreased NAA and Cre in occipital WM and linewidth

in the parietal lobe. Significant associations with body weight also remained for these same parameters, in addition to NAA/Cre in occipital WM and frontal and parietal GM.

The p values resulting from the type 3 sum of squares analysis, which indicates the significance of each variable after all variables have been entered in the model, resulted in no changes in the significance for BMI in any of the regions, and the association of Cho with weight in frontal GM changing to nonsignificant. Other differences included three instances in which the p value for gender became greater than 0.05, namely Cre and Cho in frontal WM and linewidth in occipital WM. Examination of the type 1 analyses that used BMI as the first variable in the sequential regression analysis indicated that, in those cases in which metabolite measures were significantly associated with both age and BMI, the R^2 values for age were larger than or equal to those for BMI in 18 cases, although not for Cho in frontal GM and NAA in occipital WM and cerebellum. This result supports the type 1 analysis procedure used for the results shown in Tables 1–3.

The R^2 values indicated that the impact of BMI or weight on the metabolite parameters in the cerebrum was small relative to age. For example, for NAA, which had the smallest change with age, age accounted for 8.2% of the variance in WM when averaged over all lobes, relative to 2.1% for BMI. However, the coefficient of determination for BMI relative to age was slightly larger in the cerebellum for Cre.

Figure 2 shows representative plots of the distributions of mean NAA, Cre and linewidth in occipital WM as a function of age, BMI and weight, together with the R^2 and p values from the multiple linear regression analysis (Table 2). Also shown in Fig. 2i, j are the mean SNR and CRB for the NAA area parameter for occipital WM.

The multiple linear regression analysis confirmed the finding indicated in Fig. 2g of significantly decreased SNR as a function of BMI, as well as age, in all regions (data not shown), with $p < 0.001$ for both age and BMI. These findings are consistent with the increased spectral linewidth with BMI or weight, and with age (Table 1), as the SNR measurement is based on the peak amplitude of the NAA resonance, rather than the area. However, decreased SNR was also found in all regions for male relative to female subjects ($p \leq 0.03$), even though a slightly narrower mean linewidth was found in males. Averaged over the whole cerebrum, SNR was 7% lower in males (calculated for an age of 20 years and BMI = 20 kg/m²). The R^2 values for SNR and age were, on average, 2.9 times greater than for BMI and 7.8 times greater than for gender, indicating that the age-related line broadening had the strongest influence. The R^2 values for SNR and BMI, and SNR and weight, were very similar in all regions.

The regression analysis of the mean regional NAA CRB values showed significant increases in all regions with increasing age and BMI ($p < 0.001$ in all cases), as illustrated, for example, in Fig. 2h, and increased values in males relative to females in frontal WM and temporal GM. Although this is consistent with the previous findings of corresponding decreases in SNR, the relative strengths of the associations, as indicated by the R^2 values for age and BMI, were opposite to those found for SNR, with BMI having a greater influence by a factor of 1.8 on average. This finding suggests that, although the decreased SNR was partly responsible for the increased CRB values, there are additional factors increasing the uncertainty of the spectral fitting that are associated with increasing BMI.

Figure 3 shows selected results from the voxel-based regression analyses. Figure 3b and 3c show the intercept, calculated for an age of 20 years, and the slope of the regression analysis for Cre as a function of age. This latter image has been scaled and displayed using a color scale covering a range of -5% to +5% per decade, relative to the value at 20 years. This result indicates an overall increase in Cre with age, without strong regional differences,

although with some indication of localized increases in the upper slices. Corresponding results for NAA and Cho (not shown) showed decreased and increased signal, respectively, with age, without localized differences. Figure 3d and 3g show the axial and coronal maps of the association of Cre with BMI, indicating decreasing signal with increased BMI in inferior regions and increasing signal in superior regions. The corresponding results for NAA and Cho (not shown) show similar patterns. Figure 3e and 3h show the results for the association of spectral linewidth with BMI, which reveal similar patterns, with decreasing linewidth with increasing BMI in inferior regions, and increasing linewidth with BMI particularly in parietal–occipital regions. Also shown in Fig. 3i is the result for linewidth as a function of subject weight. In contrast with the changes with age (Fig. 3c), these maps clearly indicate regional variations, with increases at the top of the head and decreases in the cerebellum and base of the cerebrum. As indicated in Fig. 3h and 3i, these regional variations are very similar for the association with BMI or weight.

Figure 4 shows the results for image-based comparison between subgroups. Figure 4b and 4c show images of the mean values of Δf and spectral linewidth for 14 subjects in low- and high-BMI subject groups. The low-BMI group corresponded to a median BMI value of 20.4 kg/m² (range, 17.9–21.5 kg/m²; median weight, 56.7 kg) and median age of 28.8 years (range, 23.5–59.4 years), and the high-BMI group to a median BMI of 31.5 kg/m² (range, 29.3–46.0 kg/m²; median weight, 95.3 kg) and median age of 39.8 years (range, 24.4–52.3 years). The Δf maps are displayed using a color scale, with blue indicating decreased values (lower B_0) and red–white showing increased values (higher B_0). These maps indicate increased resonance frequency for the low-BMI group (Fig. 3a) in central areas of the lower slices and superior–frontal regions. The patterns of the field variations shown in Fig. 4b and 4c suggest the presence of macroscopic magnetic field inhomogeneity terms of greater than second order, for which shim corrections are not available on the majority of clinical MR instruments. Manual inspection of the individual subject B_0 maps indicated Δf values at edge voxels, particularly in the posterior temporal lobe, on the order of 25–30 Hz. The range of the mean group Δf values was on the order of ± 18 Hz (± 0.14 ppm), and differences between the low- and high-BMI groups ranged from 20 Hz in the cerebellum to ≤ 10 Hz in the cerebrum.

Figure 4d shows a map of the slope of the regression of the B_0 value against BMI using all subjects, which reflects the differences between Fig. 4b and 4c. Strong changes are seen in the cerebellum and orbital–frontal regions and smaller changes in the central and superior brain regions. The corresponding map generated using body weight as the regression variable was indistinguishable from that shown in Fig. 4d, again suggesting that this finding is not specifically associated with adiposity. Maps of the spectral linewidth (Fig. 4e, f) also show different distributions between the low- and high-BMI groups, with relatively narrower linewidths in the superior regions of the low-BMI group, although broader linewidths in the region of the superior frontal cortex and cerebellum, which is consistent with the findings of more rapid Δf changes in these regions of the low-BMI group. Although spectral linewidth plays a role in the accuracy of the spectral analysis, it should be noted that the more important consideration of lineshape distortions cannot be assessed directly from the results shown in Fig. 4. Visual analysis of the water MRSI lineshape confirmed the presence of non-Gaussian lineshapes, although with considerable variability between subjects.

Figure 4g and 4h show mean CRB maps for the NAA area from the low- and high-BMI subject groups. These maps indicate excellent performance of spectral fitting in both groups (below 8% in most brain regions), but also indicate relatively increased CRB values (i.e. increased uncertainty in the spectral fitting) for the high-BMI group, as well as a trend to increased values in anterior regions relative to posterior. These findings were similar to the

SNR distributions (data not shown), which reflect the increased coil loading for heavier subjects and the sensitivity distributions of the phased-array detection coil. SNR values in the low- and high-BMI groups ranged from approximately 15 and 14, respectively, in frontal regions, to 40 and 30, respectively, in occipital regions.

The previous findings indicate that the accuracy of spectral fitting is associated with the weight (or BMI) of the subject. To test whether the metabolite differences between male and female subjects could also be a result of similar effects, maps of the mean Δf , linewidth, SNR and NAA CRB for age- and BMI-matched groups of female and male subjects were examined. Data were selected for 15 female and 19 male subjects, with median ages of 32.9 and 32.6 years, respectively, and a median BMI value of 26.5 in both groups. Figure 4i and 4j show B_0 maps from these two groups, which reveal no strong differences in the field distributions. The maps for the mean linewidth, NAA CRB and SNR (data not shown) similarly indicate no significant differences in the parameter distributions; although the multiple regression analyses revealed decreased SNR in males, varying regionally in the range of -3.6% to -12% (in temporal GM) and reaching significance in all regions, and marginally higher CRB values for males in frontal WM (by 0.18%) and cerebellum (0.47%). These findings indicate that the observed gender-related metabolite differences are not influenced by differences in spectral quality.

The associations of regional tissue volumes relative to TBV as a function of gender, age and BMI are shown in Table 4. The corresponding tables using age, gender and body weight are provided in Supporting information. The relative GM fraction was reduced significantly and the CSF fraction was increased with age in all lobes, in agreement with previous findings (4–6). When averaged over the cerebrum, GM was reduced at a rate of 3%/decade, as indicated in Fig. 5a. The GM fraction was also greater in females in the frontal lobe. WM increased with age in temporal and occipital lobes. The average value over the cerebrum showed no significant trend, as illustrated in Fig. 5b, although there is some suggestion of values peaking around middle age, as has been indicated in previous studies (4,5). Significant associations of relative tissue fractions with BMI were found for the temporal lobe, with decreased GM and increased WM, and increased CSF fraction in temporal and occipital lobes. The associations with weight were significant only for temporal WM ($p = 0.047$). None of the associations for either BMI or body weight were significant with Bonferroni correction for $p < 0.004$. Plots of the changes in total tissue volume relative to TBV are shown in Fig. 5d–f, and indicate similar trends with increasing BMI as for increasing age.

DISCUSSION

The most significant findings of lobar analysis were changes in metabolite concentrations for NAA, Cre and Cho, metabolite spectral linewidths and relative GM content with age. Although changes in these same parameters with BMI were also found, the findings were inconsistent. A comparison of the significant associations between metabolite concentrations and BMI in this study with the reports of Gazdzinski *et al.* (1–3) indicates no commonality in the locations of the findings, and, although Gazdzinski *et al.* (1) detected decreased Cho in frontal WM, this study found increased Cho in parietal and frontal GM. In two regions in which significant associations of NAA and Cre with BMI were found, it is notable that all metabolite values decreased with increasing BMI, which is inconsistent with known mechanisms of metabolic alterations of brain tissue (27), and lends support to a measurement-associated cause. Additional findings were that the associations of metabolite parameters with BMI were equally seen with body weight, indicating that these signal variations were not specifically associated with adiposity, and that, if the analysis included correction for multiple comparisons, the only significant and consistent associations found

were for increased linewidth with increasing BMI or body weight in frontal and parietal lobes.

A second major finding of this study was that both the regional and image-based regression analyses demonstrated strong associations between either BMI or body weight and factors associated with the quality of spectroscopic measurements, namely B_0 inhomogeneity, spectral linewidth, SNR and CRB. Although parametric spectral analysis should be relatively insensitive to linewidth, the spectral model used in this study assumed a Gaussian lineshape, and therefore the result is sensitive to deviations from this lineshape. A Gaussian model was chosen under the assumption that the *in vivo* MRSI resonance is dominated by local susceptibility broadening, and a fixed lineshape model has been found to be most robust for the range of SNRs encountered with the MRSI acquisition used. However, the example plots shown in Fig. 2 demonstrate that the interaction of linewidth alone with the spectral fitting algorithm is not responsible for the changes in metabolite values, as both decreased and increased metabolite values (e.g. NAA and Cre, respectively) are observed with increasing linewidth. Furthermore, the decreases in NAA and Cre with BMI are also seen in the absence of a significant change in linewidth (Fig. 2a, c, e).

The presence of lineshape distortions will have a direct impact on spectral quantification; however, the presence of systematic subject-related differences can only be indirectly inferred from the observations of differences in the B_0 homogeneity as a function of BMI (Fig. 4). Additional causes of lineshape distortion arise from temporal changes in the magnetic field. It is known that even small changes in body distribution remote from the region under study result in measurable changes in resonance frequency, as can be seen, for example, from the effect of respiration on measurements in the brain (28,29). It is speculated that different weights and body shapes, and possibly different amounts of adipose tissue around the neck and head, lead to both different residual B_0 field distributions within the brain and different time-dependent B_0 inhomogeneities resulting from physiological motion.

The highly significant decrease in SNR with body weight is not unexpected, although it may not be widely appreciated that this finding also applies to multichannel head-only receive coils and can result in differences in SNR by a factor of two across the adult weight range, which has a direct impact on spectral quantification. Smaller decreases in SNR also occur with age, reflecting the corresponding increase in linewidth; however, it is notable that the increase in CRB values with BMI or weight, relative to that with age, is greater than would be expected on the basis of the corresponding associations with SNR. This finding indicates that additional factors affect the quality of the spectral fitting, for which the contributions of increased linewidth and lineshape distortions are considered to be likely.

Although it may be expected that changes in SNR, linewidth and lineshape would result in similar errors for all resonances, or that, perhaps, decreased SNR may result in increased variance without a change in mean values, these model-fitting interactions are complex and also affected by local baseline variations and differences in overlapping resonances, potentially leading to both over- and under-fitting of different resonances in the same spectrum (30,31).

In the light of the above findings, it is concluded that the associations seen between brain metabolite signals and BMI, and reported by Gazdzinski *et al.* (1–3), apply equally to body weight, and that methodological factors affecting the quality of the spectroscopic measurement contribute to these associations. However, this study does not exclude the possibility of the role of biological differences, or demonstrate directly the cause of the observed associations.

Differences in the acquisition methods from the previous reports of Gazdzinski *et al.* (1–3) should be noted. These include TE (70 ms compared with 25 and 15 ms), field strength (3 T compared with 1.5 and 4 T) and lipid inversion nulling (TI = 198 ms compared with TI = 165 ms and no inversion pulse). The signal normalization for the first study by Gazdzinski *et al.* (1) used a scaling relative to CSF water obtained in a separate MRI measurement, and also corrected for coil loading, whereas this study used scaling to an interleaved water MRSI measurement. Although both methods apply a normalization based on the CSF signal, the interleaved measurement of this study does not require correction for coil loading factors that are dependent on subject weight. The multi-voxel analysis method used in this study was very similar to that employed in the first study by Gazdzinski *et al.* (1), although with a larger brain volume. Integration over lobar volumes benefits the analysis, and the excellent intra-subject reproducibility of this method has been demonstrated relative to single-voxel measurements (32). This study also used a whole-brain MRSI acquisition, which necessarily involves some compromise of the magnetic field homogeneity in some brain regions, and the echo planar acquisition results in temporal changes in field distributions (28) that are not fully corrected by the processing method used in this study.

There are additional differences between this and previous studies in the subject selections. This study used a larger group of subjects that had an identical acquisition protocol, with a wider age range (18–84 years) and a lower mean age (38 years). Two of the earlier reports (1,3) largely included middle-aged subjects (mean ages of 42 and 51 years), and the second study (2) included more subjects over the age of 59 years (23 compared with 11 in this study).

This study also indicated differences in individual metabolites with gender, with all metabolites being decreased significantly in male relative to female subjects in frontal and temporal WM and males having increased NAA and Cre in occipital WM. However, in the absence of the associations of any metabolite ratio measure remaining significant at the $p < 0.004$ level, it is considered likely that these findings are influenced by differences in the metabolite signal normalization to tissue water, which could, for example, be caused by differences in water content and water relaxation with gender (20,33), or by B_1 variations. Differences in tissue water content could similarly influence the associations of metabolite values with BMI, although associations of brain water with BMI have not been reported.

An additional finding of this study was an association of decreased GM and increased WM with BMI in the temporal lobe. Several studies have reported BMI to be associated with decreased TBV in middle-aged and elderly subjects (7,9,10), although reports of regional variations and for younger subjects are mixed. Ward *et al.* (34) showed that decreased high-density lipoprotein was associated with decreased temporal lobe GM volume in a subject group with an average age of 58 years, which is consistent with the finding of this study in the light of the negative association of BMI with high-density lipoprotein (35). In a large study performed at 0.5 T with a wide age range of subjects, Taki *et al.* (36) found both increases and decreases in GM volumes, although in men only. Other reports performed at higher field strengths (1,8,9,37,38) have consistently found decreased GM volumes, although both increased (37), decreased (9) and unchanged (8) WM volumes have been reported. In the light of the differences in B_0 inhomogeneity distributions between low- and high-BMI groups (Fig. 3), notably along the frontal cortex and inferior portions of the temporal lobes, and the expectation of image distortions in conventional T_1 -weighted images as a result of local susceptibility-induced B_0 distortions (39), it is possible that tissue volume measurements could reflect systematic differences in B_0 inhomogeneities with body weight. The data in Fig. 4d indicate local B_0 differences between low- and high-BMI subjects on the order of 10 Hz in the frontal lobe to 20 Hz in the brainstem and cerebellum. These values are small relative to the bandwidth of the MPRAGE acquisition used, which was 240 Hz/pixel,

suggesting that systematic differences in the spatial distortions are not significant; however, the B_0 measurement in this study is the average value over a volume of approximately 1 mL, and greater differences will exist on a smaller spatial scale. For studies that seek to characterize changes in cortical thickness on the order of 5% (0.1mm) (40), the magnitude of the spatial distortions considered here could become significant when using standard T_1 -weighted imaging sequences (39).

The limitations of this and previous studies include the use of BMI as a measure. Although BMI can serve as a simple and indirect means of estimation of adiposity for a comparable subject group, it is limited in accuracy across different races, age groups and degree of muscularity (24), and does not account for the lifetime variability of weight. The studies of Gazdzinski *et al.* (1–3) have also not reported analyses using body weight alone. The reported associations of BMI with cognitive function should also be qualified, as this has been reported for elderly (41), but not middle-aged (7), subjects, suggesting that the mechanisms of cognitive decline reflect long-term health issues. On the assumption that changes in cognitive function will be associated with altered brain metabolites (12,42), a strong association between brain metabolites and BMI may not be anticipated for young to middle-aged healthy subjects, who represent the majority of subjects included in this study.

Further limitations of this study are common to those discussed by Gazdzinski *et al.* (1), and include the derivation of BMI from the self-reporting of height by the study participants and the fact that covariates reportedly associated with metabolite concentrations, such as smoking, alcohol consumption (43) and hypertension, were not controlled for. The metabolite signal normalization method also assumed the same tissue water content for all GM and WM across different brain regions, and the same values for all subjects, and, as such, did not account for possible differences in water or metabolite relaxation rates between subjects.

CONCLUSIONS

A primary finding of this study was that methodological factors dependent on body weight can have an impact on the accuracy of metabolite concentration and volume measurements in the brain. This finding indicates that body weight should be taken into account when making comparisons of MRS and MRI volumetric measures between subject groups, or that weight should be included as a subject inclusion parameter in the selection of comparative data. Although this study confirms that significant associations of metabolite values with BMI can occur in some brain regions, the coefficient of variation was always smaller than that for age, with the exception of Cre in the cerebellum. Therefore, accounting for metabolite variations with age is of greater importance for all brain regions and for all metabolite measures.

Although this study supports the observation of an association between ^1H MRS measurements of metabolite concentrations with BMI, it does not reproduce earlier findings. However, this study also does not specifically exclude an organic origin for the observed metabolite findings or identify the specific cause of the observed associations. The observed associations of B_0 , linewidth, SNR and CRB with BMI and body weight suggest that the MRS measurements were influenced by these factors, as well as by undetected B_0 inhomogeneity-related lineshape distortions and possible differences in tissue water concentration and relaxation rates, and B_1 inhomogeneity.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Abbreviations used

BMI	body mass index
Cho	choline
CRB	Cramer–Rao bound
Cre	creatine
CSF	cerebrospinal fluid
GM	gray matter
i.u.	institutional unit
NAA	N-acetylaspartate
MIDAS	Metabolic Imaging Data Analysis System
MPRAGE	magnetization prepared rapid gradient echo
SNR	signal-to-noise ratio
TBV	total brain volume
WM	white matter.

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Figure 1.
Age and body mass index (BMI) distribution of the subject group for males (triangles) and females (circles), with the regression line generated for all subjects.

Figure 2.

Representative plots showing the distributions of the mean parameter values in occipital white matter (WM) across all subjects. Examples are shown for *N*-acetylaspartate (NAA) (a–c) and creatine (Cre) (d–f) as a function of age, body mass index (BMI) and body weight. (g, h) Distributions of linewidth for age and BMI. (i, j) Distributions of the mean signal-to-noise ratio (SNR) (for the peak NAA value) (i) and the NAA Cramer–Rao bound (CRB) (j) as a function of BMI.

Figure 3.

Example axial and coronal images generated from voxel-based linear regression analyses. (a) Axial slices from the spatial reference MRI at a slice spacing of 8 mm. (b) Creatine (Cre) intercept (for age of 20 years). (c) Change in Cre as a function of age (scale: $\pm 5\%$ /decade). (d) Change in Cre as a function of body mass index (BMI) (scale: ± 14 i.u./kg/m²). (e) Change in spectral linewidth with BMI (scale: ± 0.09 Hz/kg/m²). (f) Coronal images from the reference MRI corresponding to Fig. 2g–i at a spacing of 14 mm. (g) Change in Cre with BMI (scale: ± 14 i.u./kg/m²). (h) Change in spectral linewidth with BMI (scale: ± 0.09 Hz/kg/m²). (i) Change in spectral linewidth with body weight (scale: ± 0.025 Hz/kg).

Figure 4.

Parameter maps generated for different subject groups, as detailed in the text. (a) Reference MRI at a slice spacing of 10 mm. Mean B_0 for the low-body mass index (low-BMI) (b) and high-BMI (c) groups. (d) Slope of the regression of B_0 against BMI for all subjects. Mean spectral linewidth for the low-BMI (e) and high-BMI (f) groups. *N*-Acetylaspartate Cramér-Rao bound (NAA CRB) for the low-BMI (g) and high-BMI (h) groups. Mean B_0 for female (i) and male (j) subject groups (with same scale as in c).

Figure 5. Plots of total cerebral tissue fractions relative to total brain volume (TBV) as a function of age (a–c) and body mass index (BMI) (d–f) for gray matter (GM) (a, d), white matter (WM) (b, e) and cerebrospinal fluid (CSF) (c, f). The results of a linear regression for each are also shown.

Table 1

Slope $\beta \pm$ standard error (SE), R^2 and p values from linear model analysis using body mass index (BMI) for each metabolite parameter in frontal and temporal lobes. $p < 0.05$ shown in bold

Parameter	Frontal gray matter			Frontal white matter			Temporal gray matter			Temporal white matter		
	$\beta \pm$ SE	R^2	p	$\beta \pm$ SE	R^2	p	$\beta \pm$ SE	R^2	p	$\beta \pm$ SE	R^2	p
NAA												
Overall	2550 $\pm$ 70	0.11	0.002	2817 $\pm$ 93	0.09	0.006	2857 $\pm$ 117	0.03	0.250	2930 $\pm$ 101	0.15	<0.001
Intercept												
Male	-20.1 $\pm$ 24.5	0.00	0.857	-71.1 $\pm$ 32.9	0.03	0.040	-79.2 $\pm$ 41	0.03	0.047	-105.5 $\pm$ 35.7	0.06	0.004
Age	-3.66 $\pm$ 0.91	0.10	<0.001	-2.7 $\pm$ 1.2	0.05	0.007	-0.05 $\pm$ 1.52	0.00	0.887	-3.47 $\pm$ 1.32	0.07	0.001
BMI	3.62 $\pm$ 2.82	0.01	0.201	-4.15 $\pm$ 3.77	0.01	0.273	-1.52 $\pm$ 4.71	0.00	0.747	-6.79 $\pm$ 4.09	0.02	0.100
Cre												
Overall		0.23	<0.001		0.15	<0.001		0.17	<0.001		0.16	<0.001
Intercept	1589 $\pm$ 59			1507 $\pm$ 53			1777 $\pm$ 104			1530 $\pm$ 69		
Male	25.66 $\pm$ 20.93	0.00	0.561	-25.9 $\pm$ 18.6	0.03	0.044	-9.2 $\pm$ 36.5	0.01	0.358	-60.7 $\pm$ 24.5	0.06	0.002
Age	4.5 $\pm$ 0.78	0.23	<0.001	2.92 $\pm$ 0.69	0.12	<0.001	6.72 $\pm$ 1.35	0.16	<0.001	3.51 $\pm$ 0.91	0.09	<0.001
BMI	0.86 $\pm$ 2.4	0.00	0.721	-1.58 $\pm$ 2.13	0.00	0.460	-1.8 $\pm$ 4.19	0.00	0.668	-3.36 $\pm$ 2.81	0.01	0.234
Cho												
Overall		0.16	<0.001		0.22	<0.001		0.04	0.134		0.12	0.001
Intercept	310.6 $\pm$ 27.5			395 $\pm$ 26			316 $\pm$ 28.9			436.5 $\pm$ 29.2		
Male	-50.6 $\pm$ 52.2	0.00	0.410	-13.9 $\pm$ 9.1	0.03	0.041	8.1 $\pm$ 10.1	0.00	0.413	-18.14 $\pm$ 10.23	0.03	0.028
Age	1.46 $\pm$ 0.41	0.07	0.001	1.69 $\pm$ 0.34	0.19	<0.001	0.48 $\pm$ 0.38	0.02	0.073	1.27 $\pm$ 0.38	0.08	0.001
BMI	0.43 $\pm$ 1.1	0.03	0.052	0.87 $\pm$ 1.04	0.00	0.408	1.54 $\pm$ 1.16	0.01	0.189	-0.01 $\pm$ 1.17	0.00	0.994
Male $\times$ age	-1.55 $\pm$ 0.6	0.03	0.050									
Male $\times$ BMI	4.52 $\pm$ 2.09	0.03	0.032									
NAA/Cre												
Overall		0.47	<0.001		0.28	<0.001		0.32	<0.001		0.31	<0.001
Intercept	1.598 $\pm$ 0.041			1.881 $\pm$ 0.057			1.578 $\pm$ 0.05			1.971 $\pm$ 0.067		
Male	-0.037 $\pm$ 0.014	0.01	0.204	-0.015 $\pm$ 0.02	0.00	0.927	-0.036 $\pm$ 0.017	0.01	0.214	-0.006 $\pm$ 0.024	0.00	0.626
Age	-0.006 $\pm$ 0.001	0.46	<0.001	-0.0048 $\pm$ 0.0007	0.27	<0.001	-0.005 $\pm$ 0.001	0.31	<0.001	-0.0062 $\pm$ 0.0009	0.31	<0.001
BMI	0.001 $\pm$ 0.002	0.00	0.443	-0.001 $\pm$ 0.002	0.00	0.580	0 $\pm$ 0.002	0.00	0.893	-0.001 $\pm$ 0.003	0.00	0.603

	Frontal gray matter		Frontal white matter		Temporal gray matter		Temporal white matter	
Cho/Cre								
Overall		0.03	0.250	0.11	0.002	0.11	0.002	0.01
Intercept	0.195 ± 0.011			0.266 ± 0.013		0.172 ± 0.011		0.288 ± 0.016
Male	0.001 ± 0.004	0.00	0.530	-0.003 ± 0.0047	0.00	0.416	0.04	0.020
Age	0 ± 0.0001	0.00	0.928	0.001 ± 0.0002	0.09	<0.001	0.03	0.027
BMI	0.001 ± 0.0005	0.03	0.055	0.001 ± 0.0005	0.01	0.173	0.04	0.021
Linewidth								
Overall		0.06	0.044		0.42	<0.001	0.14	<0.001
Intercept	7.6 ± 0.38			7.4 ± 0.19		9.1 ± 0.34		7.8 ± 0.37
Male	0.018 ± 0.135	0.00	0.563	-0.152 ± 0.07	0.03	0.007	0.04	0.013
Age	-0.003 ± 0.005	0.00	0.674	0.02 ± 0.003	0.35	<0.001	0.10	<0.001
BMI	0.043 ± 0.016	0.05	0.006	0.021 ± 0.008	0.03	0.010	0.00	0.388
							0.012 ± 0.014	0.011 ± 0.015
								0.06
								0.12
								0.00
								0.455

Cho, choline; Cre, creatine; NAA, *N*-acetylaspartate.

Table 2

Slope $\beta \pm$ standard error (SE), R^2 and p values from linear model analysis using body mass index (BMI) for each metabolite parameter in parietal and occipital lobes. p 0.05 shown in bold

Parameter	Parietal gray matter			Parietal white matter			Occipital gray matter			Occipital white matter		
	$\beta \pm$ SE	R^2	p	$\beta \pm$ SE	R^2	p	$\beta \pm$ SE	R^2	p	$\beta \pm$ SE	R^2	p
NAA												
Overall	2616 $\pm$ 94	0.03	0.266	2793 $\pm$ 91	0.10	0.004	2768 $\pm$ 102	0.09	0.006	3142 $\pm$ 114	0.26	<0.001
Intercept												
Male	-65 $\pm$ 33.1	0.03	0.055	-59 $\pm$ 31.9	0.01	0.153	-119.5 $\pm$ 35.8	0.07	0.002	465.1 $\pm$ 218.2	0.04	0.007
Age	-0.03 $\pm$ 1.23	0.00	0.894	-3.94 $\pm$ 1.18	0.08	0.001	-2.23 $\pm$ 1.33	0.02	0.096	-4.05 $\pm$ 1.27	0.12	<0.001
BMI	1.79 $\pm$ 3.8	0.00	0.638	1.19 $\pm$ 3.66	0.00	0.744	1.23 $\pm$ 4.11	0.00	0.766	-6.88 $\pm$ 4.5	0.06	0.002
Male $\times$ BMI										-21.46 $\pm$ 8.31		0.011
Cre												
Overall	1592 $\pm$ 74	0.28	<0.001	1426 $\pm$ 45	0.25	<0.001	1858 $\pm$ 95	0.11	0.002	1672 $\pm$ 77	0.23	<0.001
Intercept												
Male	13.2 $\pm$ 25.9	0.00	0.799	-20.1 $\pm$ 15.8	0.02	0.057	-67.7 $\pm$ 33.4	0.04	0.014	156.3 $\pm$ 72.6	0.03	0.030
Age	6.4 $\pm$ 0.96	0.28	<0.001	3.35 $\pm$ 0.58	0.23	<0.001	3.75 $\pm$ 1.24	0.06	0.003	7.02 $\pm$ 1.27	0.11	<0.001
BMI	1.01 $\pm$ 2.98	0.00	0.734	1.21 $\pm$ 1.81	0.00	0.506	-1.75 $\pm$ 3.84	0.00	0.649	-9.53 $\pm$ 2.9	0.06	0.003
Male $\times$ age										-4.84 $\pm$ 1.78		0.007
Cho												
Overall	237.6 $\pm$ 23.1	0.19	<0.001	385.1 $\pm$ 25.1	0.16	<0.001	302.9 $\pm$ 26.8	0.02	0.356	342.5 $\pm$ 24.3	0.12	0.001
Intercept												
Male	-86.4 $\pm$ 44.4	0.01	0.238	-10.8 $\pm$ 8.8	0.02	0.112	-14.7 $\pm$ 9.4	0.02	0.089	1.89 $\pm$ 8.53	0.00	0.736
Age	0.89 $\pm$ 0.26	0.12	<0.001	1.29 $\pm$ 0.33	0.13	<0.001	0.13 $\pm$ 0.35	0.00	0.822	1.28 $\pm$ 0.32	0.11	<0.001
BMI	0.53 $\pm$ 0.92	0.02	0.060	1.13 $\pm$ 1.01	0.01	0.264	-0.58 $\pm$ 1.08	0.00	0.595	-0.5 $\pm$ 0.98	0.00	0.612
Male $\times$ BMI	3.7 $\pm$ 1.69	0.03	0.031									
NAA/Cre												
Overall	1.601 $\pm$ 0.04	0.41	<0.001	1.969 $\pm$ 0.055	0.39	<0.001	1.527 $\pm$ 0.058	0.20	<0.001	1.91 $\pm$ 0.064	0.34	<0.001
Intercept												
Male	-0.045 $\pm$ 0.014	0.02	0.033	-0.019 $\pm$ 0.019	0.00	0.934	-0.017 $\pm$ 0.02	0.00	0.904	-0.025 $\pm$ 0.022	0.00	0.824
Age	-0.005 $\pm$ 0.001	0.39	<0.001	-0.0062 $\pm$ 0.0007	0.39	<0.001	-0.004 $\pm$ 0.001	0.20	<0.001	-0.007 $\pm$ 0.001	0.34	<0.001
BMI	0.001 $\pm$ 0.002	0.00	0.727	-0.001 $\pm$ 0.002	0.00	0.557	0.001 $\pm$ 0.002	0.00	0.615	0 $\pm$ 0.003	0.00	0.939

	Parietal gray matter		Parietal white matter		Occipital gray matter		Occipital white matter	
Cho/Cre								
Overall		0.05	0.096	0.02	0.514	0.03	0.207	0.05
Intercept	0.136 ± 0.009			0.277 ± 0.014		0.159 ± 0.009		0.194 ± 0.014
Male	0.003 ± 0.003	0.01	0.219	-0.003 ± 0.005	0.00	0.551	0.899	0.005 ± 0.005
Age	0 ± 0.0001	0.00	0.446	0 ± 0.0002	0.01	0.196	0.039	0 ± 0.0002
BMI	0.001 ± 0.0004	0.03	0.039	0 ± 0.0006	0.00	0.615	0.604	0.001 ± 0.0006
Linewidth								
Overall		0.14	<0.001	0.43	<0.001	0.09	0.005	0.17
Intercept	5.8 ± 0.5			6.0 ± 0.25		7.8 ± 0.38		8.3 ± 0.33
Male	0.093 ± 0.181	0.00	0.489	-0.098 ± 0.087	0.01	0.158	0.237	-0.222 ± 0.115
Age	0.015 ± 0.007	0.08	0.001	0.023 ± 0.003	0.35	<0.001	0.002	0.017 ± 0.004
BMI	0.066 ± 0.021	0.06	0.002	0.041 ± 0.01	0.07	<0.001	0.124	0.014 ± 0.013
								0.01
								0.311

Cho, choline; Cre, creatine; NAA, N-acetylaspartate.

Table 3

Slope $\beta \pm$ standard error (SE), R^2 and p values from linear model analysis using body mass index (BMI) for each metabolite parameter in the cerebellum. $p < 0.05$ shown in bold

Cerebellum			
Parameter	$\beta \pm$ SE	R^2	p
NAA			
Overall		0.14	<0.001
Intercept	3179 $\pm$ 106		
Male	-59.2 $\pm$ 37.2	0.02	0.107
Age	-3.51 $\pm$ 1.38	0.08	0.001
BMI	-10.48 $\pm$ 4.26	0.04	0.015
Cre			
Overall		0.08	0.009
Intercept	2974 $\pm$ 178.7		
Male	-11.4 $\pm$ 62.6	0.01	0.388
Age	6.32 $\pm$ 2.32	0.03	0.055
BMI	-19.75 $\pm$ 7.19	0.05	0.007
Cho			
Overall		0.01	0.738
Intercept	664 $\pm$ 59.3		
Male	11.2 $\pm$ 20.8	0.00	0.727
Age	0.43 $\pm$ 0.77	0.00	0.819
BMI	-2.49 $\pm$ 2.38	0.01	0.299
NAA/Cre			
Overall		0.15	<0.001
Intercept	1.108 $\pm$ 0.066		
Male	-0.035 $\pm$ 0.023	0.00	0.423
Age	-0.0041 $\pm$ 0.0009	0.13	<0.001
BMI	0.004 $\pm$ 0.003	0.01	0.159
Cho/Cre			
Overall		0.02	0.406
Intercept	0.219 $\pm$ 0.016		
Male	0.004 $\pm$ 0.006	0.01	0.346
Age	-0.0003 $\pm$ 0.0002	0.01	0.332
BMI	0.0007 $\pm$ 0.0006	0.01	0.299
Linewidth			
Overall		0.19	<0.001
Intercept	10.1 $\pm$ 0.39		
Male	0.96 $\pm$ 0.753	0.15	<0.001
Age	0.003 $\pm$ 0.004	0.00	0.966
BMI	-0.005 $\pm$ 0.015	0.01	0.146
Male $\times$ BMI	-0.058 $\pm$ 0.029	0.02	0.045

Cho, choline; Cre, creatine; NAA, *N*-acetylaspartate.

Table 4

Slope $\beta \pm$ standard error (SE), R^2 and p values from the linear model analysis for relative tissue content in the four lobes. $p < 0.05$ shown in bold

Parameter	Gray matter			White matter			Cerebrospinal fluid		
	$\beta \pm$ SE	R^2	p	$\beta \pm$ SE	R^2	p	$\beta \pm$ SE	R^2	p
Frontal									
Overall		0.56	<0.001		0.04	0.158		0.59	<0.001
Intercept	0.52 $\pm$ 0.01			0.44 $\pm$ 0.01			0.03 $\pm$ 0.01		
Male	-0.012 $\pm$ 0.004	0.00	0.583	0.002 $\pm$ 0.004	0.00	0.583	0.01 $\pm$ 0.0032	0.01	0.086
Age	-0.002 $\pm$ 0.0001	0.03	0.047	0.0002 $\pm$ 0.0001	0.03	0.047	0.002 $\pm$ 0.0001	0.58	<0.001
BMI	0 $\pm$ 0	0.01	0.332	0 $\pm$ 0.0005	0.01	0.332	0 $\pm$ 0.0004	0.00	0.947
Temporal									
Overall		0.60	<0.001		0.09	0.004		0.55	<0.001
Intercept	0.62 $\pm$ 0.01			0.32 $\pm$ 0.01			0.05 $\pm$ 0.01		
Male	-0.009 $\pm$ 0.004	0.00	0.802	-0.002 $\pm$ 0.0041	0.00	0.802	-0.032 $\pm$ 0.0219	0.01	0.074
Age	-0.002 $\pm$ 0.0001	0.05	0.008	0.0003 $\pm$ 0.0002	0.05	0.008	0.001 $\pm$ 0.0001	0.53	<0.001
BMI	-0.001 $\pm$ 0	0.04	0.012	0.001 $\pm$ 0.0005	0.04	0.012	0 $\pm$ 0.0004	0.00	0.944
Male $\times$ BMI							0.002 $\pm$ 0.0008	0.01	0.048
Parietal									
Overall		0.42	<0.001		0.02	0.486		0.43	<0.001
Intercept	0.48 $\pm$ 0.01			0.45 $\pm$ 0.01			0.05 $\pm$ 0.01		
Male	-0.009 $\pm$ 0.005	0.00	0.829	-0.002 $\pm$ 0.0046	0.00	0.829	0.011 $\pm$ 0.0041	0.01	0.101
Age	-0.001 $\pm$ 0.0002	0.00	0.497	0 $\pm$ 0.0002	0.00	0.497	0.001 $\pm$ 0.0002	0.42	<0.001
BMI	-0.001 $\pm$ 0.001	0.01	0.165	0.001 $\pm$ 0.0005	0.01	0.165	0 $\pm$ 0.0005	0.00	0.932
Occipital									
Overall		0.34	<0.001		0.13	<0.001		0.23	<0.001
Intercept	0.54 $\pm$ 0.01			0.4 $\pm$ 0.02			0.06 $\pm$ 0.01		
Male	-0.004 $\pm$ 0.005	0.00	0.622	-0.001 $\pm$ 0.0055	0.00	0.622	-0.042 $\pm$ 0.0211	0.01	0.155
Age	-0.001 $\pm$ 0.0002	0.11	<0.001	0.001 $\pm$ 0.0002	0.11	<0.001	0.001 $\pm$ 0.0001	0.18	<0.001
BMI	-0.001 $\pm$ 0.001	0.01	0.192	0.001 $\pm$ 0.0006	0.01	0.192	0 $\pm$ 0.0004	0.01	0.130
Male $\times$ BMI							0.002 $\pm$ 0.0008	0.03	0.023

BMI, body mass index.