Diagnostic Accuracy of ^{1}H-MRS Using PRESS and MEGA-PRESS Techniques in the Preoperative Grading of Patients With Gliomas

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Background: Edited MRS technique such as MEshcher-GArwood Point RESolved Spectroscopy (MEGA-PRESS) can determine isocitrate dehydrogenase mutation (IDH) mutation status in patients with gliomas but its accuracy in assessing glioma grade has not yet been formally evaluated.
Purpose: To evaluate the diagnostic accuracy of metabolites such as lactate obtained from the PRESS and MEGA-PRESS sequences in the preoperative grading of glioma. To assess the prognostic value of those metabolite ratios in the overall survival of patients with gliomas.
Study Type: Prospective.
Subjects: Sixty-nine subjects with gliomas (16 grade 2, 21 grade 3, and 32 grade 4). Mean age was 50.5 ± 16.7 years; 38 were male and 31 were female.
Field Strength/Sequence: 3 T/MEGA-PRESS, PRESS.
Assessment: Single voxel PRESS and MEGA-PRESS spectra were obtained from tumors in patients undergoing preoperative MRI. Several tumor metabolites were measured from the PRESS, MEGA-PRESS edit-off, and difference spectra using LCModel (Linear Combination of Model Spectra) software. Diagnosis and glioma grading was done using the World Health Organization (WHO) 2016 classification. Overall survival was assessed.
Statistical Tests: Diagnostic accuracy was measured using receiver-operating characteristic (ROC) curve. Univariate and multivariate Cox proportional hazards modeling was used for the assessment of prognostic factors for time to death.
Results: In the differentiation between low- vs. high-grade gliomas, tCr/tCho ratios obtained from PRESS and MEGA-PRESS sequences had similar accuracies (area under the ROC curves [AUCs] = 0.71) while Lac/NAA from PRESS had a lower accuracy (AUC = 0.65). The presence of a detectable 2-hydroxyglutarate peak on the difference spectrum was a favorable prognostic factor in univariate analysis (hazard ratio = 0.25, 95% confidence interval: 0.08–0.83). No other metabolite was found to be a significant prognostic factor in univariate and multivariate analyses.
Data Conclusion: Edited MRS can be used to detect metabolites which can help in the preoperative grading of gliomas and in determination of the overall survival. A separate PRESS acquisition is needed for lactate quantification.
Plain Language Summary: Gliomas are brain tumors that vary in severity. This study explored the use of two advanced MR spectroscopy techniques (PRESS and MEGA-PRESS) in detecting tumor metabolites. The authors found that both techniques' choline/creatine ratio showed moderate accuracy in identifying high-grade gliomas. Lactate was better revealed

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with the PRESS technique and was associated with high-grade gliomas. They confirmed that the MEGA-PRESS technique allowed additional detection of 2-hydroxyglutarate in IDH-mutant gliomas, which was linked to better survival. These findings emphasize that advanced MR spectroscopy can extract metabolic information time-efficiently, which can be used to improve the preoperative diagnosis of patients with gliomas.
Level of Evidence: 1
Technical Efficacy: Stage 2

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H^1-magnetic resonance spectroscopy (MRS) helps in the noninvasive diagnosis of brain tumors by providing metabolite profiles.[1] Detected metabolites which are different between tumors compared to normal brain can serve as tumor biomarkers.[2] In the clinical setting, the (Point RESolved Spectroscopy) PRESS technique is most frequently used, which allows the measurement of choline containing compounds (tCho), N-acetyl aspartate (NAA), creatine and phosphocreatine (tCr), and myo-inositol (mIns). Increased tCho levels in tumors occur due to the increased cell proliferation and the breakdown of cell membranes.[3] Higher tCho levels are associated with a higher tumor grade.[1] Besides changes in the tCho content, nearly all tumors in the brain show decreased NAA levels, which results from the loss of normal neuronal tissue. The evaluation of tCho with NAA has shown to be a useful diagnostic feature to characterize brain tumors, since the ratio of tCho/NAA increased with the tumor grade.[3] Creatine is located in the mitochondria and the cytoplasm and serves as a marker for cellular energy metabolism. The level of tCr varies with tumor type and also with glioma grade.[3] A decrease in the tCr peak is related to an increase in the tumor metabolism, although the underlying biochemical processes leading to the change are not well understood.[3] Myo-inositol (mIns) is an osmolyte found in astrocytes and its concentration varies with tumor grade possibly due to disruption of the blood barrier in high-grade tumors.[4] Glycine (Gly) is an amino acid which is needed for cellular proliferation and its elevation has been associated with glioma aggressiveness.[5] Lactate is a product of anaerobic metabolism and is mainly found in high-grade gliomas.[3]

With the advent of higher field MRS, additional metabolites such as gamma-aminobutyric acid (GABA) and 2-hydroxyglutarate (2-HG) can also be detected in the brain.[6–8] As the signals from these metabolites overlap with coresonant signals from other metabolites in the spectrum, spectral editing methods have shown to be more reliable than classical localized spectroscopy methods.[7] One of the most commonly applied spectral editing sequences is the MEshcher-GArwood Point RESolved Spectroscopy (MEGA-PRESS) sequence, an extension of the PRESS sequence.[6] MEGA-PRESS enables the detection of J-coupled metabolites by applying a spectral selective pulse on one of the coupling resonances (edit-on) and repeating the scan by applying the spectral selective pulse on the opposite site of the center frequency, where no resonances of the metabolite are present (edit-off). The subtraction of the two spectra reveals the resonances of the edited metabolites, where overlapping noncoupled resonances are removed. MEGA-PRESS is used to detect GABA, but it was shown that it can be used to detect 2-HG in patients with IDH-mutant gliomas.[7,8]

For the grading of gliomas, it is hypothesized that the edit-off spectrum from the MEGA-PRESS sequence can provide metabolite measurements (such as tCho, tCr and NAA) similar to those obtained with the conventional PRESS sequence. However, a conventional PRESS sequence might still be needed to differentiate the lactate from the lipid peak. Therefore, it is unclear if the grading of gliomas can be determined from one MRS sequence only.

The aim of this study was to evaluate the diagnostic accuracy of standard metabolites obtained from the PRESS and the MEGA-PRESS sequences such as lactate in the preoperative grading of gliomas. The secondary aim was to assess the prognostic value of those metabolites in the overall survival of patients with newly diagnosed gliomas.

Materials and Methods

Subjects

This study was approved by the local ethics board (REB#20140754-01H). Written informed consent was obtained from each patient enrolled in this study. We prospectively recruited consecutive patients who presented with a newly suspected diagnosis of glioma between October 2015 and February 2022 at our hospital. Inclusion criteria were: adult patient (≥18 years of age) with first lifetime presentation of suspected glioma on initial scans. Exclusion criteria were pediatric patients (under 18 years of age), nonsurgical patients, prior temozolomide or brain radiation therapy, nonglioma tumors, patients with recurrent tumors, and inability to provide written informed consent. We recorded clinical variables such as the age, sex of patients, and type of surgery (biopsy vs. resection). The main outcome measure was overall survival. Follow-up clinical information was obtained primarily from chart review, telephone interview, and archived obituaries. The follow-up period was defined as the interval between the date of the preoperative MRI and the date of death or the date the patient was last known to be alive. Follow-up was halted on August 1, 2023.

Magnetic Resonance Imaging and Spectroscopy

MRI data were acquired as added to a clinical examination protocol using a 3 Tesla MR scanner (Trio, Siemens Healthineers®, Erlangen,

Germany) and a 32-channel receive-only head coil. The conventional MR images included an axial 2D T2-weighted (repetition time [TR] = 6700 msec, echo time [TE] = 97 msec, field-of-view [FOV] = 220 × 176 mm^2, acquisition matrix = 320 × 256, in-plane resolution = 0.69 × 0.69 mm^2, slice thickness = 3 mm), axial 2D spin-echo T1-weighted (TR = 280 msec, TE = 2.5 msec, FOV = 220 × 170 mm^2, acquisition matrix = 256 × 170, in-plane resolution = 0.86 × 1.00 mm^2, slice thickness = 3 mm), axial 2D fluid-attenuated inversion recovery (FLAIR) (TR = 9710 msec, TE = 93 msec, inversion time [TI] = 2580 msec, FOV = 220 × 175 mm^2, acquisition matrix = 256 × 163, in-plane resolution = 0.86 × 1.07 mm^2, slice thickness = 3 mm), coronal 2D FLAIR (TR = 9710 msec, TE = 90 msec, TI = 2580 msec, FOV = 210 × 190 mm^2, acquisition matrix = 256 × 186, in-plane resolution = 0.82 × 1.02 mm^2, slice thickness = 3 mm), and axial 3D gradient echo T1-weighted post contrast (TR = 8.48 msec, TE = 3.2 msec, flip angle = 12°, FOV = 256 × 256 × 192 mm^3, matrix = 256 × 256 × 192, resolution = 1 × 1 × 1 mm^3).

FLAIR images were used to locate the tumor within the brain and to plan the voxel position for the following spectroscopy experiments. The voxel was positioned by an MR physicist or a neuroradiologist to include the maximal amount of solid tumor tissue avoiding hemorrhagic or cystic areas.

Automated voxel selective shimming was performed using the vendor specific gradient-shimming routine for spectroscopy. The water resonance line width at full width at half maximum (FWHM) was checked after shimming, and if it exceeded 0.15 ppm (18 Hz), additional shimming was performed using manual shimming or the fast automatic shimming technique, by mapping along projections (FAST (EST)MAP), to improve local field homogeneity.[9] Standard point resolved spectroscopy (PRESS) spectra were acquired with chemical selective water suppression using the following parameters: TR = 2 seconds, TE = 135 msec, receiver bandwidth = 1.5 kHz, sampling points = 1024, averages = 128, duration = 4:24 minutes. The MEGA-PRESS was applied to acquire edit-on and edit-off spectra from the same voxel based on the method published by Choi et al.[8] For spectral editing, a 20 msec Gaussian 180° pulse was applied at 1.9 ppm for the edit-on condition and interleaved at 7.5 ppm for the edit-off condition to edit for 2-HG at 4.02 ppm in the difference spectra. Acquisition parameters were: TR = 2 seconds, TE = 60 msec, receiver bandwidth = 2.5 kHz, sampling points = 2048, scan time = 4:24 minutes for the first group of patients (64 pairs of scan) and 8:48 minutes for the second group of patients (128 pairs of scans). Following a midterm analysis, the scan time for the second group of patients was increased to improve the signal-to-noise ratio (SNR).

Data Processing and Analysis

MR SPECTROSCOPY. PRESS, MEGA-PRESS difference and edit-off spectra were analyzed with LCModel 6.3-1 L.[10] The data was filtered with 1 Hz (exponential line broadening) and metabolites were fitted using the simulated basis sets and a spectral window from 0.2 to 4.2 ppm. Simulated basis spectra were generated for both sequences using the simulation tool within the Vespa (Versatile Simulation, Pulses and Analysis) spectroscopy package.[11] Metabolites included: 2-HG = (2-hydroxyglutarate), NAA = (N-acetylaspartate + N-acetylaspartylglutamate), tCr = (creatine + phosphocreatine), tCho = (glycerophosphorylcholine + phosphocholine), Glx = (glutamate [Glu] + glutamine [Gln]), glutathione (GSH), myo-inositol, glycine, scyllo-inositol, aspartate, cystathione, taurine, gamma-aminobutyric acid (GABA), aspartate, alanine and lactate. The subtracted spectrum of the edited MRS acquisition was fitted for 2-HG, cystathione, Glx, GSH, GABA, and NAA. Metabolite concentrations were expressed as institutional units (IU) since no water scaling was performed. Cramer-Rao lower bounds (CRLB) were provided by the LCModel as an expression of the uncertainty of the estimate for each metabolite.

For quality control, an MR physicist (G.M., 15 years of experience) first inspected the PRESS and edit-off spectra from the LCModel. Patients with poor quality spectrum spectra (SNR < 8; FWHM > 0.15 ppm) and with spectral artifacts (such as out-of-volume lipid contamination and baseline distortion) were removed from further analysis.[12,13] FWHM was measured from the highest metabolite signal following baseline subtraction. SNR was calculated as the ratio between the highest metabolite signal intensity over twice the root mean square of the residuals.[10] The MR physicist then inspected the difference spectra from the MEGA-PRESS acquisition and excluded additional patients with artifacts.

PATHOLOGICAL ANALYSIS AND CLINICAL OUTCOME. Following surgery, the diagnosis of glioma was confirmed and graded using WHO 2016 classification by two neuropathologists (J.W. and G.J., 25 and 29 years of experience, respectively).[14] IDH1 mutation status was assessed by immunohistochemistry using the H09 clone (Dianova) generated against the R132H mutant IDH1. Bound antibody was revealed using the Leica Bond III automated immunostaining platform. All patients with grade 2 or 3 gliomas and a negative immunohistochemistry for the IDH R132H mutation underwent further molecular testing for the detection of non-canonical IDH1 or IDH2 gene mutations using next-generation sequencing on the MassArray iPLEX platform (Agena Bioscience®, San Diego, CA, USA).

Statistical Analysis

Differences between the median metabolite ratios among gliomas of various grades were assessed with Kruskal–Wallis test. Following a Holms-Bonferroni correction, an adjusted *P*-value of <0.05 was considered as significant. Receiver-operating curve (ROC) analysis was performed to measure the diagnostic accuracy of different metabolite ratios in the diagnosis of high-grade vs. low-grade gliomas. Noninferiority analysis of the area under the ROC curves (AUC) for metabolite ratios obtained from the edit-off and PRESS spectra was performed. Noninferiority of edit-off is inferred if there is no worse than 0.10 reduction in AUC from PRESS. Clinical variables (such as age and type of surgery), IDH mutation status and metabolite ratios were assessed as prognostic factors for time to death using a univariate Cox proportional hazards modeling. Variables that resulted in a probability <0.05 on univariate analysis were entered sequentially into a multivariate analysis in a forward stepwise regression. After a variable is entered in the model, variables that became nonsignificant ($P > 0.2$) are removed. Statistical analysis was performed with MedCalc Statistical Software version 18.9 (MedCalc Software bvba, Ostend, Belgium; http://www.medcalc.org; 2018).

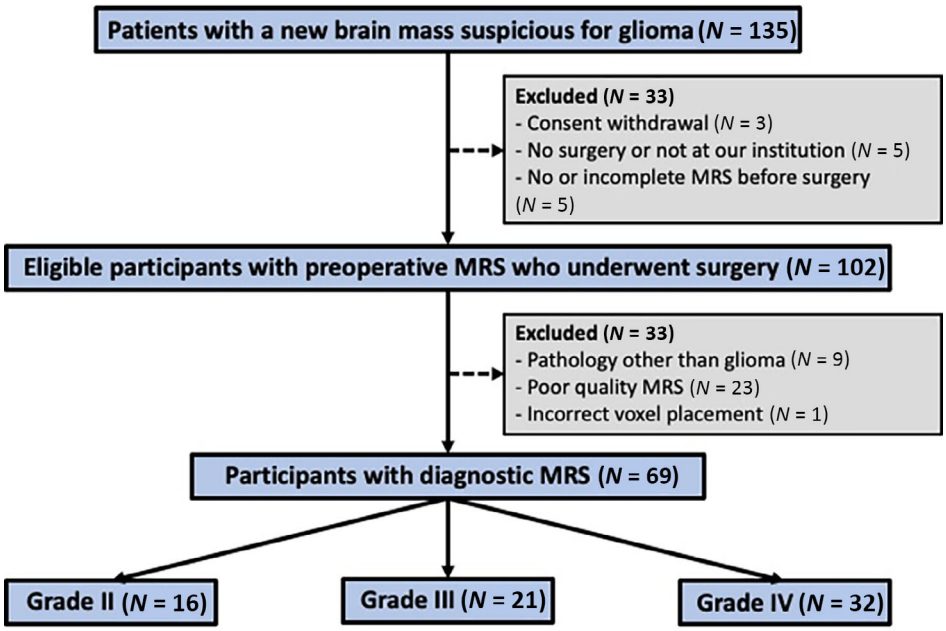

FIGURE 1: Flow chart of prospective patients who were enrolled in the study.

TABLE 1. Sample Demographic Characteristics: Clinical and Pathological Data of Patients

	Grade 2 ($N = 16$)	Grade 3 ($N = 21$)	Grade 4 ($N = 32$)
Mean age (years) ± SD	42.2 ± 12.9	41.8 ± 13.5	59.7 ± 15.7
Gender			
Male	11	12	15
Female	5	9	17
Astrocytoma	10	14	32
Oligodendroglioma	6	7	0
IDH-mutant	11	15	1
Resection	16	19	25
Biopsy	0	2	7

Results

Subjects

There were 135 patients prospectively enrolled in this study. There were 66 patients excluded for the following reasons: withdrawal of consent ($N = 3$), no pathological diagnosis ($N = 15$), diagnosis other than glioma ($N = 9$), no or incomplete MRS acquisition ($N = 15$), incorrect voxel placement ($N = 1$), poor quality MRS ($N = 23$) (Fig. 1). The clinical and pathological data of the patients are summarized in Table 1. There were 38 (55%) patients with a voxel volume of 8 cm^3, 8 (12%) patients with a voxel size less than 8 cm^3 (range 1.2–7.5 cm^3) and 23 (33%) patients with a voxel size greater than 8 cm^3 (range 10–36 cm^3).

MR Spectroscopy

A neuroradiologist and a MR physicist inspected MR spectra from edit off, difference and PRESS spectra. On the difference spectrum, four patients were excluded due to the presence of artifacts on visual inspection. The average SNR was 9.97 ± 8.05 for the difference spectra. The median 2-HG concentration was 1.33 IU (interquartile range [IQR] 0–2.34 IU) for IDH-mutant gliomas compared to 0 IU (IQR 0–0.37 IU) IDH-wildtype gliomas. The median CRLB value was 40% (IQR 23–999) for IDH-mutant gliomas compared to 999% (IQR 137–999) for IDH-wild type gliomas. Using a predefined CRLB less or equal to 30% for 2-HG,[15,16] MEGA-PRESS had sensitivity of 36% and

TABLE 2. Data Quality Values: Mean Full-Width at Half Maximum (FWHM) and Signal-to-Noise Ratios (SNR) With Their Standard Deviations; Sequence Specific Mean Metabolite Concentration in Institutional Units (IU); and CRLB% Obtained From the PRESS and MEGA-PRESS Edit-Off Spectra

	FWHM (ppm)	SNR	Mean Metabolite Concentration (IU) and CRLB (%)									
			Lac	CRLB (%)	NAA	CRLB (%)	tCho	CRLB (%)	tCr	CRLB (%)	Ins + Gly	CRLB (%)
PRESS ($N = 69$)	0.06 (0.02)	25.5 (10.6)	104	16	170	11	157	1.6	253	2.5	150	8.7
Edit off, all ($N = 69$)	0.06* (0.02)	26.8* (11.3)	4.9	25	8.3	5.4	6.8	1.8	11.9	3.0	17.0	8.5
Edit off, FID = 64 ($N = 25$)	0.07 (0.03)	26.5 (12.0)	5.3	27	7.5	6.0	6.0	1.8	10.1	3.3	12.5	11.8
Edit off, FID = 128 ($N = 44$)	0.06* (0.02)	27.0* (11.0)	4.6	24	8.9	5.0	7.2	1.8	12.9	2.8	19.5	6.6

The absolute values (IU) between PRESS and MPRESS are different due to vendor specific internal scaling of the dicom spectra. Mean metabolite concentration was calculated from patients with mean metabolite concentration greater than 0 and CRLB <999%.
*One patient was excluded from this group because the SNR was 260 due to an unsuppressed lipid peak.

specificity of 90% in the identification of the IDH mutation status in gliomas.

The tCr, tCho and NAA peaks could be identified on the edit-off and PRESS spectra in all 69 patients included in this study. The average FWHM was 0.06 ± 0.02 ppm for both the edit-off and PRESS spectra (Table 2). The average SNRs were 26.8 ± 11.3 for the edit-off spectra (after exclusion of a patient with a large lipid peak and a SNR of 260) and 25.5 ± 10.6 for PRESS spectra. The median CRLB% for lactate were 25% for edit-off spectra and 16% for PRESS spectra. From consensus interpretation of the edit-off spectrum, the lactate 1.33 ppm peak was not detected in nine patients (four patients with CRLB% >100% and five patients with CRLB% <100%). In the remaining patients, the lactate peak was seen with no or minimal overlapping resonances in 22 patients and was overlapping with a large lipid peak in 38 patients. On the PRESS spectrum, the lactate resonance was not detected in five patients (two patients with CRLB% >100%, three patients with CRLB% <100%). The lactate resonance was visible as an inverted peak without co-resonant lipids in 38 patients and was overlapping with lipid signal in 26 patients. Representative MRS spectra for gliomas of various grades are shown in Fig. 2. On the edit-off spectrum, the combined mIns + Gly resonance was detected in all patients with varying contributions from mIns and/or Gly. On the PRESS spectrum, the combined mIns + Gly resonance was nearly exclusively due to Gly as detected by the LC model.

From the edit-off spectrum from MEGA-PRESS, the median tCr/tCho ratios were 2.16 (IQR 1.92–2.5) for grade 2, 2.04 (IQR 1.54–2.60) for grade 3, and 1.49 (IQR 1.05–1.99) for grade 4 gliomas. From PRESS, the median tCr/tCho ratios were 1.87 (IQR 1.62–2.30) for grade 2, 1.71 (IQR 1.43–1.98), and 1.37 (IQR 0.92–1.73) for grade 4 gliomas. The PRESS Lac/tCr ratios were 0.15 (IQR 0.11–0.69) for grade 2, 0.17 (0.11–0.37) for grade 3, and 0.56 (0.23–1.18) for grade 4 gliomas. Following Holms-Bonferroni correction for multiple comparisons, the tCr/tCho ratio from the edit-off spectrum and the Lac/tCr from conventional PRESS sequence were different between various tumor grades. The tCr/tCho from PRESS was not statistically different between tumor grades ($P = 0.01$). Similarly, from the PRESS and edit off spectra, there was a trend of higher mIns + Gly/tCho ratios in low-grade vs. high-grade gliomas but this did not reach difference following correction for multiple comparisons ($P = 0.03$ for edit off and $P = 0.04$ for PRESS; Table 3).

In the differentiation between low-grade (grade 2) and high-grade (grade 3 and 4) gliomas, the diagnostic accuracies of tCr/tCho obtained from PRESS and MEGA-PRESS sequences were similar (AUCs = 0.71) (Table 4, Fig. 3). The tCr/Cho from MEGA-PRESS is noninferior to PRESS at the 0.10 level (no worse than 0.10 reduction in AUC from PRESS). The Lac/NAA ratio from the PRESS sequence can also differentiate between low- and high-grade gliomas

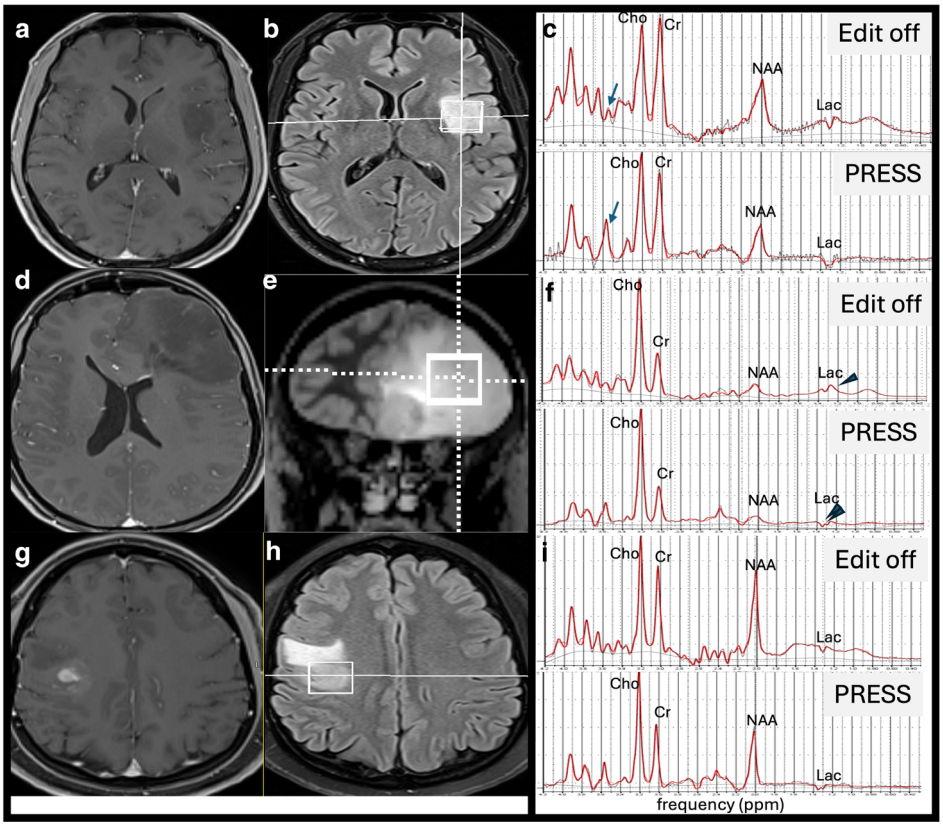

FIGURE 2: Representative conventional MR images and spectra for gliomas of various grades. (a) Axial T1-weighted and (b) axial FLAIR images demonstrate a nonenhancing hyperintense T2 mass, which was correctly interpreted as low-grade astrocytoma. (c) tCr/tCho ratios were high on both spectra. The mIns + Gly peak can be identified at 3.55 ppm (arrow) on the edit-off spectrum and on the regular PRESS spectrum. (d) Axial T1-weighted and (e) coronal FLAIR images show a nonenhancing tumor, which was erroneously interpreted as a low-grade astrocytoma. High tCho and low tCr peaks are seen on both the edit-off and regular PRESS spectra (f). The inverted lactate peak can be clearly identified as an inverted doublet peak (double arrowhead) on the regular PRESS spectrum but appears as a M-shaped peak with positive and negative components on the edit-off spectrum (arrowhead) due to the phase evolution at TE = 60 msec, which makes it more difficult to separate it from overlapping lipids. Difference spectrum reveals 2-HG peak (not shown). Pathology revealed an anaplastic oligodendroglioma. (g) Axial T1-weighted and (h) axial FLAIR images show an enhancing tumor, in keeping with a high-grade astrocytoma. High tCho and low tCr peaks are seen on both the edit-off and regular PRESS (i) spectra. Pathology confirmed the presence of a glioblastoma.

(AUC = 0.65), but not the Lac/NAA from the edit-off sequence (AUC = 0.58). For tCr/tCho obtained from PRESS spectrum, using a pre-defined cut off ≤1.5,[17] the sensitivity was 43% (95% confidence interval [CI]: 30–58), specificity was 81% (95% CI: 54–96), positive predictive value was 89% (95% CI: 73–96) and negative predictive value was 30% (95% CI: 24–38). Using a tCr/tCho ≤1.5 cutoff from the edit-off sequence, sensitivity was 40% (95% CI: 27–54), specificity was 88% (95% CI: 62–98), positive predictive value was 91% (95% CI: 73–98), and negative predictive value was 30% (95% CI: 25–37).

Survival Analysis

Forty-two deaths occurred during the study period. The median follow-up for those still alive was 1261 days (95% CI: 1037–1713). The median survival time for all patients was 777 days (95% CI: 643–995). Clinical variables such as younger age and surgical resection instead of biopsy were found to be significant positive prognostic factors for the overall survival of patients with gliomas with hazard ratios (HRs) of 1.06 (95% CI: 1.03–1.08) and 0.28 (95% CI: 0.14–0.57), respectively (Table 5). A positive 2-HG peak detected on the difference spectrum using a CRLB% ≤30% was found to be a favorable prognostic factor (HR = 0.25, 95% CI: 0.08–0.83) in univariate analysis but not in multivariate analysis. None of the other metabolite ratios (tCr/tCho, mIns + Gly/tCho, and Lac/NAA) was found to be a significant factor in univariate or multivariate analysis.

Discussion

Spectral editing sequences such as MEGA-PRESS typically take advantage of scalar coupling for the optimal detection of 2-HG.[8] Prior studies have reported that MEGA-PRESS technique has high specificity in the detection of 2-HG in patients with IDH-mutant tumors.[7,8] However, the difference spectrum cannot provide quantification of choline and creatine, which are subtracted out. The spectrum acquired

TABLE 3. Difference in Metabolite Ratios Among Various Grades of Gliomas

	Grade 2 ($N = 16$)		Grade 3 ($N = 21$)		Grade 4 ($N = 32$)		
Metabolite Ratios	**Median**	**IQR**	**Median**	**IQR**	**Median**	**IQR**	***P***
Edit-off mIns + Gly/tCho	2.62	2.23–4.00	2.84	2.00–3.53	2.01	1.15–3.06	0.031
Edit-off Lac/NAA	0.32	0.19–0.66	0.23	0.16–0.53	0.64	0.14–2.01	0.16
Edit-off Lac/tCr	0.19	0.13–0.74	0.16	0.09–0.24	0.56	0.10–1.46	0.06
Edit-off NAA/tCho	1.23	0.85–1.83	0.92	0.45–1.23	0.87	0.37–1.50	0.12
Edit-off tCr/tCho	2.16	1.92–2.51	2.04	1.54–2.60	1.49	1.05–1.99	0.0024*
PRESS mIns + Gly/tCho	1.00	0.94–1.44	1.01	0.84–1.27	0.75	0.66–1.07	0.041
PRESS Lac/NAA	0.25	0.14–0.79	0.36	0.17–0.71	0.73	0.25–3.34	0.02
PRESS Lac/tCr	0.15	0.11–0.68	0.17	0.11–0.37	0.56	0.23–1.18	0.0027*
PRESS NAA/tCho	1.44	0.94–1.76	1.12	0.52–1.47	1.02	0.36–1.38	0.01
PRESS tCr/tCho	1.87	1.62–2.30	1.71	1.43–1.98	1.37	0.92–1.73	0.01

CI = confidence interval; Lac = lactate; NAA = N-acetyl aspartate; tCr = creatine and phosphocreatine; tCho = choline containing compounds; mIns = myo-inositol.
*Adjusted *P*-value <0.05 is considered statistically significant following Holms-Bonferroni correction.

TABLE 4. Diagnostic Accuracy of Various Metabolite Rations in Differentiating Low-Grade (Grade 2) From High-Grade Gliomas (Grade 3 and 4)

Variable	**AUC**	**95% CI**	***P***
Edit-off mIns + Gly/tCho	0.62	0.47–0.76	0.12
Edit-off Lac/NAA	0.54	0.41–0.66	0.6
Edit-off Lac/tCr	0.51	0.39–0.63	0.14
Edit-off NAA/tCho	0.67	0.55–0.78	0.01
Edit-off tCr/tCho	0.71	0.58–0.81	0.002
PRESS mIns + Gly/tCho	0.66	0.52–0.80	0.03
PRESS Lac/NAA	0.65	0.53–0.76	0.04
PRESS Lac/tCr	0.63	0.51–0.75	0.09
PRESS NAA/tCho	0.69	0.57–0.80	0.04
PRESS tCr/tCho	0.71	0.59–0.81	0.001

AUC = area under the curve; CI = confidence interval; Lac = lactate; NAA = N-acetyl aspartate; tCr = creatine and phosphocreatine; tCho = choline containing compounds; tCr = creatine and phosphocreatine; mIns = myo-inositol.

without the editing pulse (edit-off spectrum) can be used for more comprehensive metabolite profiling. Using the tCr/tCho and NAA/tCho ratios, we have found that the diagnostic accuracies of the PRESS and MEGA-PRESS sequences were similar in the differentiation between low- and high-grade gliomas. The areas-under-the curve for tCr/tCho and NAA/tCho from both sequences were slightly lower in our study than values reported in a prior meta-analysis study,[18] which could be due to a higher proportion of anaplastic astrocytomas relative to glioblastomas in our study.

The tCr/tCho ratios for gliomas obtained from the edit-off spectrum are higher compared to those derived from the

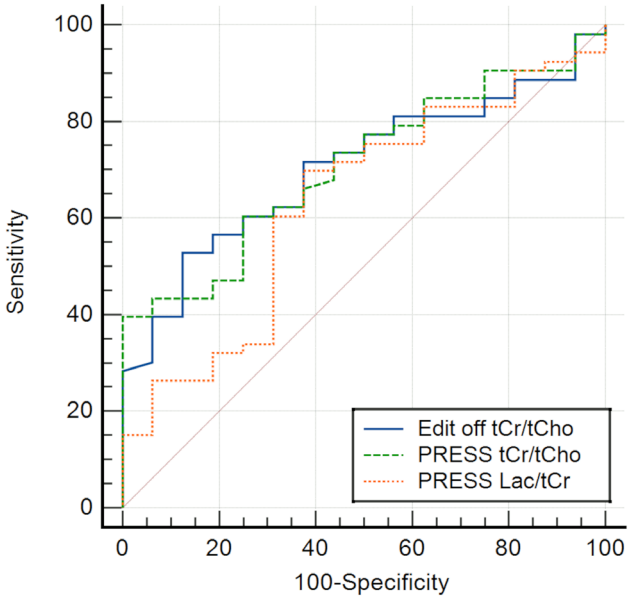

FIGURE 3: Accuracy of different metabolite ratios in differentiating between gliomas. The tCr/tCho ratios from edit-off and PRESS spectra have similar diagnostic accuracies (AUC = 0.71) in differentiating low-grade from high-grade gliomas.

TABLE 5. Univariate and Multivariate Analysis Using Different Metabolite Ratios and Clinical Variables as Prognostic Factors in the Overall Survival of Patients Presenting With Newly Diagnosed Gliomas

Variables	Hazard Ratio (95% CI)	*P*
Edit-off tCr/tCho	0.79 (0.53–1.18)	0.25
PRESS tCr/tCho	0.85 (0.53–1.35)	0.49
PRESS Lac/NAA	1.00 (0.99–1.00)	0.43
Positive 2-HG[a]	0.25 (0.08–0.83)	0.02
IDH-mutation	0.12 (0.05–0.30)	<0.0001
Age	1.06 (1.03–1.08)	<0.0001
Resection vs. biopsy	0.28 (0.14–0.57)	0.004
Model (IDH mutation, age, type of surgery)		
IDH-mutation	0.20 (0.08–0.51)	0.0007
Age	1.04 (1.02–1.06)	0.0024
Resection vs. biopsy	0.33 (0.15–0.76)	0.0092

CI = confidence interval; 2-HG = 2-hydroxyglutarate.
[a]Positive 2-HG was defined as CRLB% ≤ 30% on difference spectrum.

PRESS spectrum. This could be due to the longer TE used for PRESS acquisition and the longer T2 relaxation time of choline relative to creatine in normal white matter and in gliomas.[19] In comparison to T2 uncorrected metabolite concentration, there is a higher degree of correlation between T2 corrected metabolite concentrations from the edit-off spectrum and concentrations from the regular PRESS.[20] However, T2 correction was not performed in this study because our primary objective was not to assess reproducibility but to evaluate diagnostic accuracy in identifying high-grade gliomas. The T2 relaxation times for the major metabolites (tCho, tCr, tNAA, and Lac) have been reported to be similar between low-grade gliomas and high-grade gliomas.[21] Thus, T2 correction is not expected to improve the diagnostic accuracy of each metabolite ratio in differentiating between low-grade and high-grade gliomas (Fig. 3).

Lactate was more frequently and more reliably detected by the LCModel analysis using the PRESS sequence with the longer TE of 135 msec. This could be due to the increased signal decay of co-resonant lipid signals at the longer TE compared to the overlapping metabolite signal. Further, the lactate resonance shows a strong phase dispersion at TE of 60 msec, compared to a clear phase inversion at TE = 135 msec, which possibly affects the LCModel analysis in this spectral range, especially when co-resonant signals from lipids or macromolecules are present. In our study, the Lac/NAA ratio from PRESS was found to be moderately accurate in differentiating low- from high-grade gliomas. This result is in line with prior studies which have reported the lipid + lactate/Cr ratio to be significantly different between low- and high-grade gliomas.[18]

In this study, the ratio of mIns + Gly over Cho tends to be higher in low-grade vs. high-grade gliomas. Myoinositol is an osmolyte produced by healthy neuroglial cells while glycine is associated with tumor proliferation and aggressiveness.[4,5] Thus, competing effects of decreased mIns and increased Gly could explain why the combined mIns + Gly peak did not differentiate between low- vs. high-grade gliomas. Separate identification of Gly from mIns has been previously reported using an optimized PRESS sequence of 97 msec.[5] In our study, we used a longer TE (60 msec) in the MEGA-PRESS sequence because this TE range enables the detection of 2-HG using spectral editing. We believe that detection of mIns and Gly might be possible on the edit-off spectrum using LCModel for fitting the full metabolite spectrum but further research is needed. Shyu et al reported the detection of mIns in normal brain with a good spectral quality, although the CRLB% was almost double for edit-off vs. a short TE PRESS sequence.[22] At longer TEs such as 135 msec used in our PRESS sequence, the fast mIns signal decay due to T2 impacts its detection, with presumably Gly contributing to the combined mIns + Gly peak.

Limitations

This study was performed in one single center on one single 3 T scanner, which limits the reproducibility of our results. Our study was also limited by the lack of direct spatial correlation between the preoperative MRS voxel placed in the tumor and the tumor sample resected for histological and molecular analysis. In order to maximize the SNR for the MEGA-PRESS sequence, we used a single voxel MRS which might encompass areas of different tumor grades. In the future, spatial 2D or 3D spectral-editing sequences might become available for clinical use, which would improve the spatial resolution and allow more direct correlation between tumor metabolites and histological analysis in heterogeneous tumors. Another limitation of our study was the use of the WHO 2016 CNS classification instead of the most recent 2021 classification. Our study began before the molecular markers such as TERT promoter mutation and CDNK2A/B became available at our institution. Thus, some gliomas classified as low-grade in our study might be reclassified as molecular glioblastomas with the current classification.

Conclusion

Edited MRS can detect several metabolites which are useful for the preoperative glioma subtyping. While the difference spectrum can reveal the presence of 2-HG in IDH-mutant tumors, the edit-off spectrum of the MEGA-PRESS can provide metabolite ratios such as tCr/tCho, which can help in the differentiation between low-grade and high-grade gliomas, similar to regular PRESS acquisition. However, the PRESS acquisition allows more accurate identification of the lactate peak which is higher in high-grade gliomas.

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