Repeatability of measured lactate and other metabolites in patients with astrocytoma

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Introduction

Tumours can contain large amounts of lactate, and an early drop in the lactate signal may be a sensitive measure of response to chemotherapy (1). However, measurement of lactate in tumours is challenging due to overlapping lipid peaks, whose unusual mobility makes them visible even at the relatively long TE of 144 ms used to identify inverted lactate. Subtraction-based spectral editing using BASING pulses has been used to discriminate lactate from lipids (2,3). We have implemented lactate editing at 3T and assessed its repeatability in phantoms and in human brain tumours *in vivo* to evaluate it as a possible biomarker in drug treatment.

Methods

Two PRESS-localized spectra were acquired (TE/TR 144/2500ms, 32 averages), with BASING pulses applied at frequency offsets toggled between -40 Hz ('editing on', inverting the lactate peak at 4.1ppm) and +30 Hz ('editing off'). Unsuppressed water signal (4 averages without BASING pulses) was collected as a reference. Nine patients with newly-diagnosed primary astrocytoma were studied with informed consent prior to biopsy: 7 Grade IV glioblastoma, 2 Grade III astrocytoma. Single voxels were placed in the area of maximal enhancement on fast spoiled gradient echo (FSPGR) images following Gadovist injection and in normal-appearing contralateral white matter. After a break of 5-10 minutes the examination was repeated, using automated repositioning of voxel positions based on registration of the FSPGR images (4). Repeatability was also assessed in the GE MRS Head sphere containing 5mM lactate, 12.5 mM NAA, 10 mM creatine, and 3 mM choline, using the same voxel size (7.9cc) but only 2 averages at each frequency to more closely approximate SNR *in vivo*.

Spectra were analyzed using LCModel 6 with water-scaling after pre-processing in SAGE7.0 to optimally reconstruct the multi-coil data using the unsuppressed water, perform the subtraction and addition, and extract matched spectra with and without water suppression. For each metabolite the within-subject differences (scan 2 – scan 1) were calculated as were the corresponding means and standard errors (SD/sqrt(n)) of the differences. Also 95% confidence intervals were calculated (for the differences *in vivo* and for the means *in vitro*) referencing a t-distribution with (n-1) degrees of freedom. The coefficient of variation (CoV) was calculated *in vitro* as (100%*SD/mean).

Results and Discussion

Due to time constraints, the contralateral voxel could not be acquired in 1 patient. Difference spectra typically showed clear inverted lactate (Fig. 1). Fitting of the difference spectra was improved by constraining the phase and by choosing CSF as the spectral template: otherwise LCModel would distort the spectrum in the attempt to maximize any small residual peaks of NAA, creatine and choline.

In phantoms, a coefficient of variation of 11% was achieved for Lac with only 2 averages at each frequency (Table 1). This gives a similar SNR to 32 averages *in vivo*: typically SNR was >5 for Lac, and >10 for Cho, Cr and NAA, which showed CoV of 2-4%.

Table 2 summarizes mean metabolite concentration (mM), minimum and maximum concentrations, mean difference (scan2 – scan 1, mM), and 95% confidence interval for the true mean difference, in both tumour and contralateral tissue. In patients, the automated voxel repositioning provided good consistency between scans 1 and 2 both in the voxel location and in the spectral patterns observed. It was estimated that the mean serial change in lactate concentration could be as large as 3.1 mM. For NAA, Cr and Cho, the corresponding values were 2.6, 0.9, and 0.4 mM. For lipid content the mean serial change could be as large as 7.6 mM. However, all of the 95% CIs included zero so a smaller or zero mean change cannot be ruled out On average, metabolite concentrations tended to be lower on scan 2 than scan 1, possibly because of the different time since Gd contrast injection (approximately 10 minutes for scan 1 and 50 minutes for scan 2). This difference did not reach significance (α=.05, two-sided) for any metabolite on paired t-tests.

The expected pattern of difference in metabolite levels was seen in tumour compared with normal-appearing tissue: NAA and Cr were much lower; lactate and lipid were highly elevated. Choline was higher in tumour than contralateral tissue in some patients but lower in others, perhaps due to the unavoidable inclusion of some necrosis in the voxel, since choline is thought to arise principally from viable proliferating cells. The inter-subject variability was extensive, with particularly marked differences between the 2 patients with Grade III astrocytoma and the 7 with Grade IV glioblastoma. Further study is underway to correlate the content of lactate and other metabolite with the tumour grade and also with image-based estimates of the volumes of T_2 hyperintensity, contrast enhancement, and necrosis. Spectroscopic imaging data has also been acquired and analysis is in progress.

Conclusion

This is to our knowledge the first study to estimate the repeatability of lactate and other brain metabolites in human tumours *in vivo*. The lactate editing procedure is practical in the clinical setting and tolerated by patients. This may provide a useful means of simultaneously monitoring lactate, choline and lipids *in vivo*, all of which are of interest in tumour progression and response to treatment.

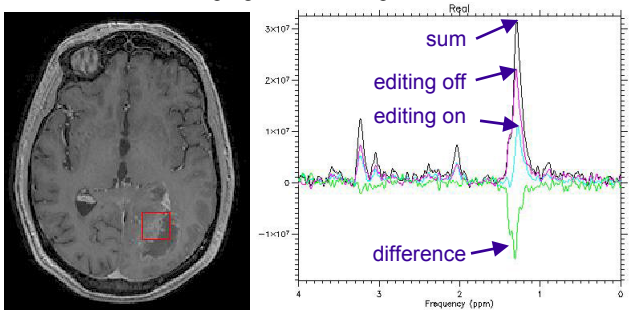

Figure 1: Lactate editing in Grade IV Glioblastoma *in vivo*.

References

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Table 1: Mean metabolite concentrations (mM ± SD), coefficient of variation (CoV), and 95% confidence interval for GE MRS head sphere *in vitro* (n=5).

	Mean ± SD	CoV	95% CI
Lactate	5.3 ± 0.6	11%	(4.6, 6.1)
Choline	1.29 ± .05	4%	(1.22, 1.35)
NAA	7.36 ± 0.17	2%	(7.15, 7.57)
Creatine	4.78 ± 0.14	3%	(4.61, 4.96)

Table 2: Mean metabolite concentration (mM), minimum and maximum concentrations, mean difference (scan2 – scan 1, mM), and 95% confidence interval for the true mean difference, in both tumour and normal-appearing contralateral tissue *in vivo*.

		Lactate	Choline	NAA	Creatine	Lipid
Tumour (n=9)	Mean Conc. (mM)	11.5	3.1	1.8	2.7	33.5
	Range (min, max)	1.8, 30	1.1, 7.1	0.8, 5.6	1.1, 5.7	0.3, 68
	Mean Difference	0.9	-0.08	-0.3	-0.13	-2.2
	Range of Diff.	-2.2, 7.2	-0.56, 0.37	-1.04, 0.32	-1.62, 1.3	-17, 7.5
	95% CI Diff.	(-1.4, 3.1)	(-0.3, 0.1)	(-0.6, 0)	(-0.7, 0.4)	(-7.6, 3.2)
Contralateral (n=8)	Mean Conc. (mM)	1.1	3.5	21.6	11.7	1.3
	Range (min, max)	0, 3.0	2.2, 4.8	14.9, 29.2	8.3, 14.7	0, 4.5
	Mean Difference	-0.8	-0.06	-0.2	-0.03	-1.0
	Range of Diff.	-2.6, 2.1	-0.79, 0.38	-1.8, 6.3	-2.4, 1.0	-9.1, 4.5
	95% CI Diff.	(-2.1, 0.4)	(-0.4, 0.2)	(-2.6, 2.1)	(-0.9, 0.9)	(-4.2, 2.2)