Detection of Elevated Glutathione in Meningiomas by Quantitative *in vivo* ^{1}H MRS

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Abstract

Glutathione has a major role in removing toxic free radicals from tissue, but its presence also hinders the effectiveness of many anti-cancer therapies. Careful analysis of brain tumor ^{1}H spectra has shown the presence of elevated glutathione in meningiomas compared to normal white matter and low-grade gliomas. The ability to quantify glutathione *in vivo* may aid selection of treatment therapies.

Introduction

LCModel uses a linear combination of *in vitro* metabolite solution spectra (the "basis-set") to model *in vivo* ^{1}H-MRS spectra and determine individual metabolite concentrations[1]. When fitting short echo time spectra of meningiomas using the standard LCModel (containing 13 individual metabolite spectra), we consistently observed underestimation at ~2.9 and 3.78 ppm and overestimation at ~2.36 and 3.4 ppm (see Figure 1). This suggests that the basis set does not include all NMR visible metabolites that contribute to meningioma spectra. This study proposes that glutathione (GSH) makes a substantial and quantifiable contribution to short echo meningioma spectra, as well as in other tissue.

Methods

From an ongoing study of brain tumors by ^{1}H-MRS, we analysed 6 each of spectra from meningiomas, astrocytomas (grade II) and normal white matter in volunteers. Data (*in vivo* and metabolite solution spectra) were acquired using a 1.5T Signa Horizon, (GE Medical Systems, Milwaukee, WI) with standard quadrature head coil and the fully automated PROBE acquisition sequence using short echo time STEAM (TE=30ms, TR=2020ms).

Reduced (GSH) and oxidised (GSSG) glutathione (SIGMA) solutions were prepared in deoxygenated distilled water with TSP (Fluka) and Sodium formate (SIGMA) added to provide a chemical shift reference and scaling for concentration determination by LCModel. The solution pH was adjusted to 7.2 using solid NaOH (BDH).

In vivo spectra were analysed using LCModel (Version 5.2-3R) both with and without GSH or GSSG added to the basis set, and with synthesised spectral peaks to represent the macromolecules and lipid resonances at ca. 2.85, 2.25, 2.05, 1.3 and 0.9 ppm[2, 3].

Results

Adding the GSH spectrum to the LCModel basis set, significantly reduced the residual component (top panels Fig. 1) of the fit to meningioma spectra between 2.2 and 3.9 ppm to almost the noise level, and the baseline (lower continuous line) is also reduced. GSSG alone did not produce such a good fit. The estimated *in vivo* GSH concentrations (using the unsuppressed water as reference signal) are given in Table 1 and compared to literature values of GSH in normal brain and tumour as measured by histofluorescence[4].

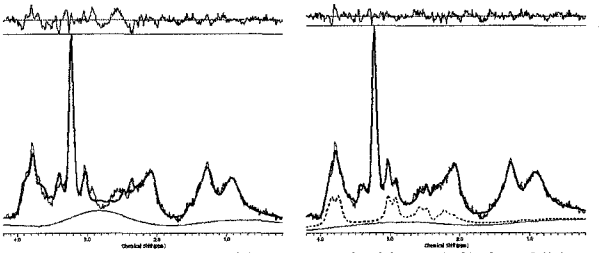

Figure 1: LCModel fits, with (right) and without (left) the addition of GSH in the basis set, of a meningioma spectrum. The dotted line (right) shows the contribution from GSH in the fit.

	LCModel	Histofluorescence
	GSH (mM)	GSH (mM)
Meningioma	3.3 ± 1.5* (n=6)	2.0 ± 0.8
Normal	1.2 ± 0.15 (n=6)	1.4 ± 0.34
Astrocytoma (II)	1.0 ± 0.26 (n=6)	1.1 ± 0.03

Table 1: Comparison of GSH concentrations as measured by LCModel in *in vivo* ^{1}H-MRS and histofluorescence techniques on brain tissue samples[4]. *Mann Whitney, $p<0.005$ compared to normal and astrocytoma II.

Discussion

Adding GSH to the LCModel basis set significantly improved the fitting of the meningioma and astrocytoma II spectra. The spectral components from glutamate and glutamine (Glx), and macromolecules, were not sufficient to correctly fit the 2.0–2.5 ppm region. The MRS quantified GSH is comparable to that measured by histofluorescence[4], reflecting significantly higher levels in meningiomas than in astrocytoma II and normal tissue, and supports our assignment. The total Glx in meningiomas was also found to be about twice that of normal white matter, consistent with MAS MRS data from biopsies[5], which also supports our analysis. Glutathione is an important antioxidant present in reduced and oxidised forms in tissue. Including both in the basis set gave a GSH/GSSG ratio of 4.0 ± 1.2 in meningiomas. As GSH levels are reportedly more than 50 times higher than GSSG in normal parietal white matter[6] this result seemed unlikely, and probably due to low spectral resolution at 1.5T *in vivo*. In conclusion we believe we have demonstrated the ability of MRS to detect an important biochemical that has not previously been quantified *in vivo* except at 7T in normal brain tissue[7].

References

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