# Proton MR Spectroscopy of Substantia Nigra in the Human Brain at 4 Tesla: Measurement of High GABA Concentrations

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## Introduction

Parkinson's disease (PD) is characterized by loss of dopaminergic neurons in the substantia nigra (SN), but its etiology remains poorly understood (1). *In vivo* $^1$H MRS may provide a noninvasive alternative to culture, animal model and post-mortem studies that are more commonly utilized to understand the pathogenesis of PD. However, due to its small size, the MRS investigation of the SN in the human brain is difficult (2). Previous studies primarily employed 6-8ml voxels which implicate substantial partial volume effects. In addition, they were performed at 1.5 Tesla and therefore only provided data on NAA, creatine, choline and their respective ratios (2). The aim of the current study was to determine the feasibility of expanding the information content of $^1$H MR spectra from smaller volumes at 4 Tesla to neurotransmitters, such as GABA and glutamate, and the antioxidant glutathione.

## Methods and Subjects

All studies were performed on a 4 Tesla / 90 cm magnet (Oxford/Varian). A TEM volume coil (3) was used as the NMR transceiver. STEAM combined with OVS and VAPOR water suppression (4) (TE=5ms, TM=42ms, TR=4.5s, NEX=400) was used to obtain single voxel spectra from SN unilaterally in 8 healthy volunteers (4 males : 4 females, average age±SD: 51.4±12.6 years). The voxel size was 2.2ml (n=4) or 3.4ml (n=4). Preliminary data was also acquired from 9 mild-moderate PD patients (4 males : 5 females, 57.7±10.7 years) from 2.2 ml voxels encompassing the SN contralateral to the more affected side. Quantitation of metabolites was done with LCModel (5) using the water signal as an internal reference. In one healthy subject GABA editing was performed using macromolecule suppressed MEGA-PRESS in a 5.3 ml voxel that encompassed the SN bilaterally according to previously described procedures adapted to 4T (6).

## Results and Discussion

Despite the broad linewidths of $^1$H MR spectra (Fig.1) due to the high iron content in the SN several metabolites including GABA, glutamate, glutathione and *myo*-Inositol were quantified using the LCModel method in all subjects with reasonable reliability as judged from the average Cramer-Rao lower bounds (CRLB, Table 1). This was not unexpected for these resonances, where the homonuclear J coupling dominated the linewidth. Additionally, the feasibility of studying PD patients was demonstrated by CRLB similar to controls. Interestingly, GABA concentrations were substantially higher than the ~1mM reported in cortex (6, 7). The quantification of high GABA concentrations was consistent with the Glu H4 resonance being shifted upfield to ~2.32 ppm in spectra averaged from several subjects (Fig. 1), indicating comparable concentrations of GABA and Glu (Table 1). In addition, MEGA-PRESS editing provided an estimate of ~5mM GABA concentration in a healthy subject (bottom trace, Fig.1). Finally, the GABA and Glu concentrations (Table 1), at 3-4 fold and approximately half the cortical values, respectively, were in very good agreement with autopsy results and animal studies (8) consistent with lower number of glutamatergic and higher number of GABAergic neurons in SN.

In conclusion, MRS of the SN at 4T can be used to evaluate neurotransmitters GABA and Glu, as well as the antioxidant GSH. This suggests that further study is merited to evaluate the utility of high field MRS in assessing pathogenetic theories of PD.

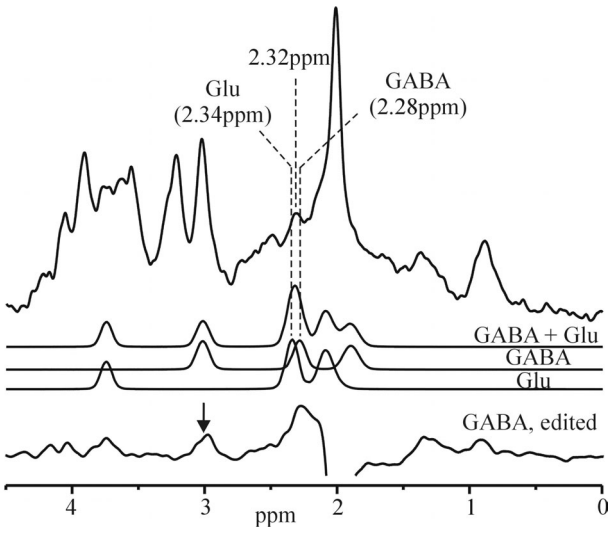


**Fig. 1.** Averaged $^1$H spectrum acquired from the substantia nigra of PD patients (top, n=9, VOI=2.2ml, 30min acquisition per patient). The contributions of GABA and Glu found by LCModel, as well as their sum, are shown below the *in vivo* spectrum. Also shown is the edited GABA spectrum (the 3ppm H4 resonance marked with an arrow) obtained from a healthy subject (bottom, VOI=5.3 ml, 80 min).

**Table 1.** Concentrations (mean ± SD) and average Cramer-Rao lower bounds (estimates of the SD of the fit) for metabolites quantified in the SN of healthy volunteers (VOI=2.2ml, n=4). Metabolites with high cross-correlation are reported as sums rather than individual values. (GABA: γ-aminobutyric acid, Glu: glutamate, GSH: glutathione, Ins: *myo*-inositol, Lac: lactate, NAA: N-acetylaspartate, NAAG: N-acetyl-aspartyl-glutamate, GPC: glycerophosphoryl-choline, PCho: phosphoryl-choline, Cr: creatine, PCr: phosphocreatine, Glc: glucose, Tau: taurine).

| | GABA | Glu | GSH | Ins | Lac | NAA+NAAG | GPC+PCho | Cr+PCr | Glc+Tau |
|---|---|---|---|---|---|---|---|---|---|
| Conc (mM) | 3.6 ± 0.3 | 5.3 ± 1.5 | 2.5 ± 0.7 | 9.0 ± 0.5 | 2.2 ± 0.3 | 14.5 ± 0.2 | 2.7 ± 0.4 | 8.5 ± 0.4 | 5.1 ± 2.3 |
| Avg CRLB | 30% | 24% | 28% | 8% | 32% | 6% | 15% | 9% | 29% |

## References

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