In vivo MR spectroscopic changes in the brain and spinal cord after experimental spinal cord injury in rats

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1. Introduction

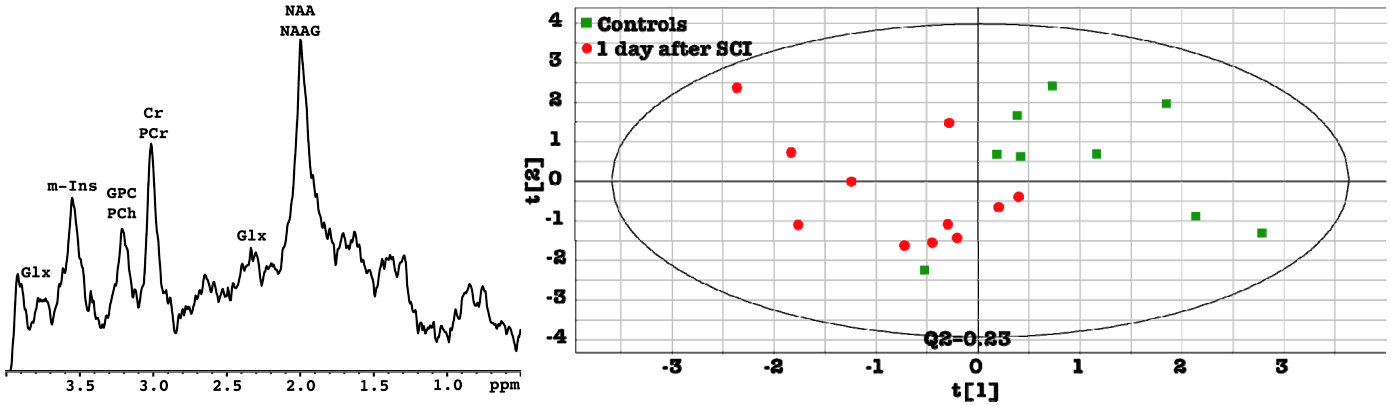
A variety of tests of sensorimotor function are used to characterize outcome after experimental spinal cord injury (SCI). However, these tests do not provide information about chemical and metabolic processes in the injured central nervous system (CNS). Spinal cord injury results in acute local primary injury and a more progressive secondary injury. The primary injury results from the destructive local impact to the spinal cord, whereas the secondary injury is characterized by a number of degenerative processes in local tissue and long projection pathways. In this work, proton magnetic resonance spectroscopy (MRS) was used to monitor chemical changes in the CNS (brain and spinal cord) *in vivo* following SCI in rats. Both long-term and short-term changes were investigated.

2. Method

A spinal cord transection injury model in adult Sprague-Dawley rats was used. In order to assure complete transaction a 3 mm long segment of the spinal cord was removed, at the vertebral level T9. Metabolic changes were monitored in rats (N=32) using *in vivo* proton MRS in cerebral cortex, thalamus/striatum and the spinal cord (lumbar enlargement, below injury). Individual rats were scanned at several time points in order to investigate both long-term and short-term changes. The time points studied were: prior to SCI, 1, 3, 14 days, 3 and 4 months after SCI.
All experiments were performed on a horizontal 4.7 T/40 cm Bruker magnet. PRESS was used for localization with repetition time = 3500 ms and echo time = 20 ms. 256-512 averages were acquired. Anaesthesia was induced with isoflurane and with a 2 % and maintainance dose during scans with spontaneous breathing. The program LCModel [2, 3] was used for quantification of metabolites in the spectra. Metabolite changes between groups over time were analysed with the SPSS software package (SPSS, Chicago, IL). The quantified metabolites were further analyzed with multivariate data analysis (with the SIMCA software (Umetrics AB, Umea, Sweden)) using the method PLS-DA as described in [1].

3. Results

The MR spectra were analyzed using LCModel, and the concentrations of 10-12 metabolites were reliably quantified according to Cramér-Rao lower bounds (CRLB) in cortex, striatum/thalamus and the spinal cord (left figure) of control rats as well as rats with SCI. The resolution, in therms of full width at half maximum (FWHM), and the signal-to-noise ratio (SNR) of the spectra were reported by LCModel and the means were calculated for each voxel. FWHM/SNR was 0.04/14 for brain spectra and 0.07/5 for spinal cord spectra.
In cerebral cortex significant changes of glutamate and N-acetylaspartate were found in rats prior to SCI compared to rats with SCI. Glutamate levels were decreased 1 day after injury with a tendency to recover towards the values of intact animals, but not reaching normal levels at the last time point investigated (4 months after SCI). No significant changes between control rats and rats with SCI were detected in the thalamus/striatum voxel, but in the spinal cord, significant changes were found for glutamine, glutamate, *myo*-inositol and N-acetylaspartate. Both long-term and short-term changes were detected. Models that included all quantified metabolites were built voxelwise with partial least squares discriminant analysis (PLS-DA) (example in right figure). Using these models, predictions for classification of control rats and injured rat were made with leave-one-out cross-validation. Sensitivity and specificity of the method were calculated from the predictions.



Example of spectrum from spinal cord (left) and scatter plot of PLS-DA of control rats and rats one day after SCI based on spectra from the spinal cord (right).

References

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