Longitudinal changes in the neurochemical profile of Hungtington R6/2 mice

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INTRODUCTION

The R6/2 transgenic mouse model is very useful for studying the pathogenesis of Huntington's disease (HD). Previous spectroscopy studies using HD models were limited in scope by quantifying a limited range of metabolites, or by measuring data within a narrow range of the disease progression, or by collecting data only from a single brain region, or by using in vitro methods [1-4]. The aim of this study was to expand the use of in vivo ^{1}H NMR spectroscopy to investigate neurochemical changes in striatum and cerebral cortex during the lifespan of R6/2 mice starting from a presymptomatic age with respect to behavioral and anatomical changes associated with the disease progression.

METHODS

All NMR measurements were performed using a Varian INOVA spectrometer interfaced to a 9.4 T magnet, equipped with powerful gradient/shim coils insert (Resonance Research Inc). First and second order shims were adjusted by FASTMAP [5]. Ultra-short echo-time STEAM (TE = 2 ms) combined with outer volume suppression and VAPOR water suppression was used for ^{1}H NMR spectroscopy [6,7]. Metabolite concentrations were quantified using LCModel with macromolecule spectra included in the database and the unsuppressed water signal was used as an internal reference [8]. High resolution multislice RARE images (fig.1) (in-plane resolution = 100 µm x 100 µm, slice thickness = 0.3 mm) were evaluated by user guided segmentation (Amira) to determine regional brain volume changes. The behavioral changes were evaluated by climbing test assay [9]. R6/2 mice (120-142 CAG repeats, n = 8) and their wild type (WT, n = 8) littermates, were obtained from Jackson Laboratories. ^{1}H NMR spectra were acquired from cortex and striatum at 4, 8, 12 and 15 weeks of age.

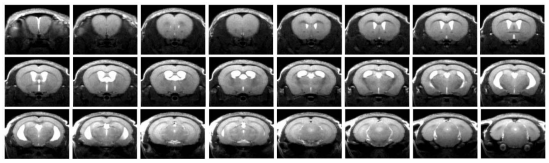

Fig.1 High resolution RARE images (300 µm slice thickness, 100 µm x 100 µm) for volumetric evaluation.

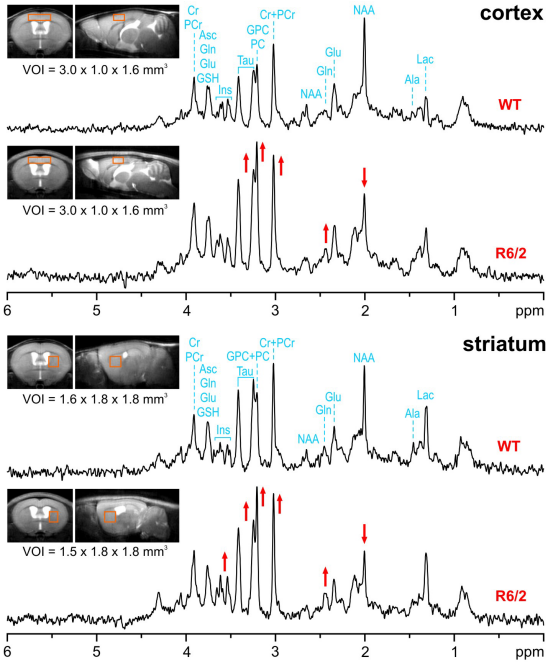

Fig. 2 In vivo ^{1}H NMR spectra acquired from the cerebral cortex and striatum of HD transgenic R6/2 and WT mice at 15 weeks of age. STEAM, TE = 2 ms, TR = 5 s, NT = 240.

RESULTS AND DISCUSSION

Shrinkage was seen in cortex, striatum, and anterior brain starting at 8 weeks but lateral ventricles were only significantly larger at 4wks and remained larger. The rate of volume change in the cortex was slower than that of the striatum. Routinely achieved spectral quality (Fig. 2) enabled reliable quantification of sixteen brain metabolites from each spectrum. Progressive changes in multiple brain metabolites were observed both in striatum and cerebral cortex. The most significant differences between R6/2 and WT mice were detected for Gln, NAA, Cr+PCr, GPC+PC and Tau, while the concentration of Glu was not affected (Fig. 3). Neurochemical concentrations became significantly different at 8 weeks of age and later, but trends for decreased NAA and increased Gln and total Cr were already obvious at 4 weeks. Observed changes in the neurochemical profiles and tissue volumes indicate neurodegenerative processes resulting in the neuronal loss, compromised neurotransmission, energy production and osmolytic regulation. Behavioral changes were observed at all time points with the latency to climb and instances of climbing distinguishing genotypes at early time points and time spent rearing distinguishing genotypes at older ages. In conclusion, these results clearly demonstrate that neurodegenerative processes are not restricted to striatum in the R6/2 mouse model of HD. Second, these results document the utility of in vivo ^{1}H NMR spectroscopy at high magnetic fields for longitudinal studies of transgenic mouse models of neurodegenerative diseases.

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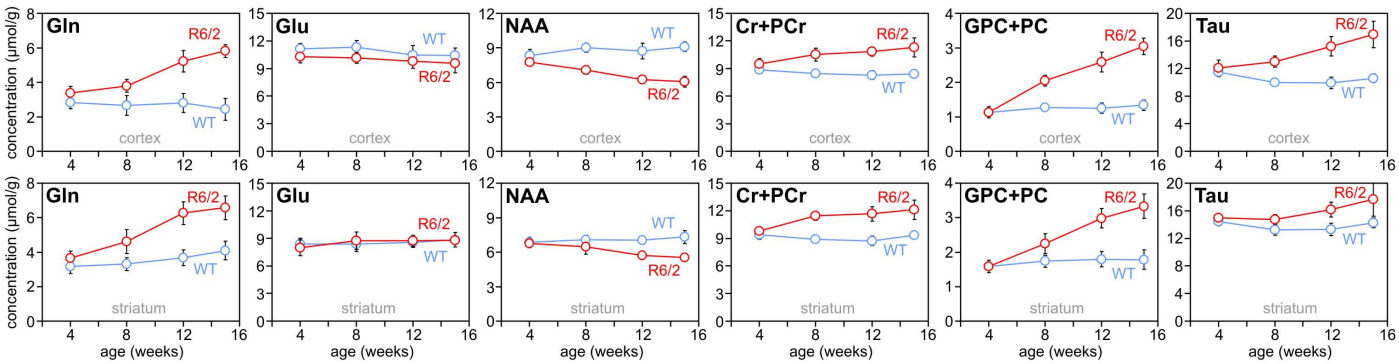

Fig. 3 Longitudinal changes in concentration of selected brain metabolites measured in the cerebral cortex and striatum of R6/2 (n = 8) and WT (n = 8) mice. Error bars indicate the SD.