Quantifying myelin kinetics in healthy subjects using deuterium labeling.

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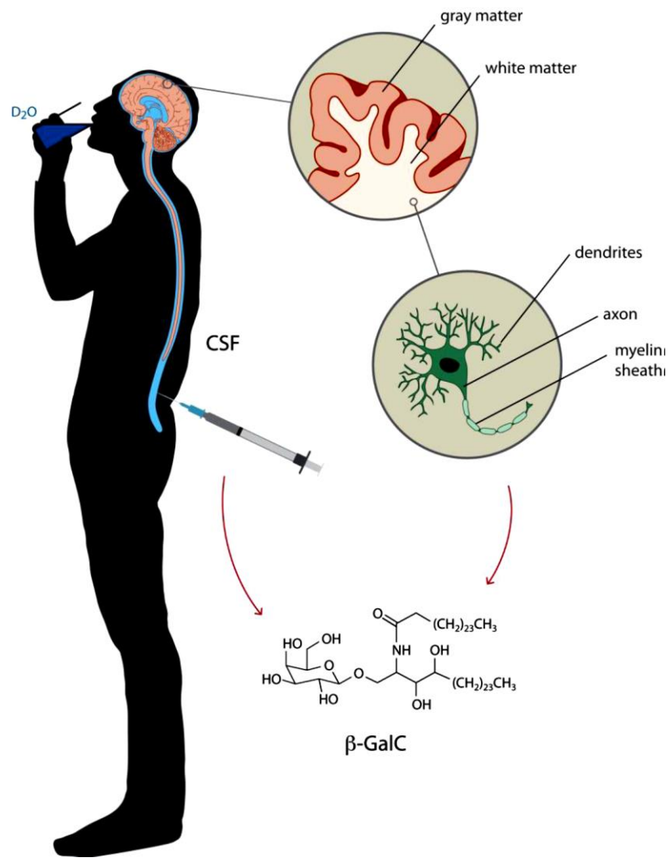
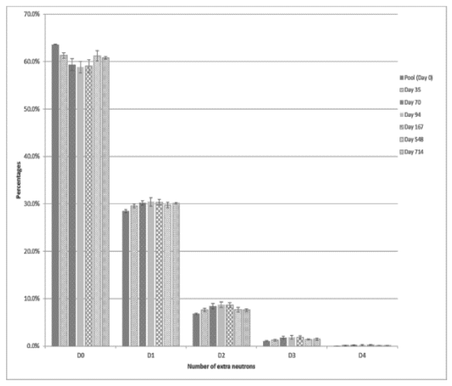
INTRODUCTION

Demyelinating diseases, such as multiple sclerosis (MS), are characterized by an increased breakdown of myelin with a subsequent failure of the remyelination process. Enhancement of remyelination may improve recovery after an exacerbation[1]. However, direct enhancement of remyelination can only be shown when myelin turnover rate can be quantified. The turnover rate of biomolecules can be determined by quantification of deuterium labeling after chronic administration of deuterated water (D_2O). Although the labeling of myelin cannot be determined directly in vivo, typical breakdown products or myelin precursors such as beta-galactosyceramide (BGalC) can be measured in cerebrospinal fluid (CSF)[2].

METHODS

- 6 healthy volunteers
- Daily 120mL 70% D_2O, 70 days
- 5 lumbar punctures (days 35, 70, 93, 167 and 548 or 714)
- LC/MS/MS System for BGalC analysis in CSF[3]
- BGalC turnover rate estimated using non-linear mixed effects modeling





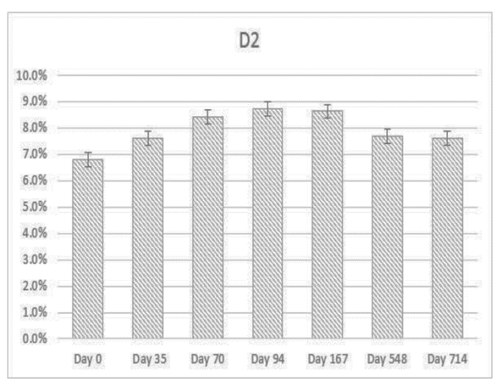


Figure 1: Figure left shows the iso-topomers of β-GalC and how the relative quantity of each iso-topomer changes over time during and after 10 weeks of D2O labeling. D0 implies that the β-GalC molecule has no extra neutron, while D1, D2, D3, D4 means that the β-GalC molecule has 1, 2, 3 and 4 extra neutrons as determined by mass spectrometry. In the baseline sample (pool, day 0) the D1, D2, D3, and D4 iso-topomers are also present, which is due to natural occurrence of stable radio-isotopes in food substances (primarily C13). In figure on the rightthe D2 iso-topomers are shown and how the relative abundance of the D2 iso-topomer changes over time.

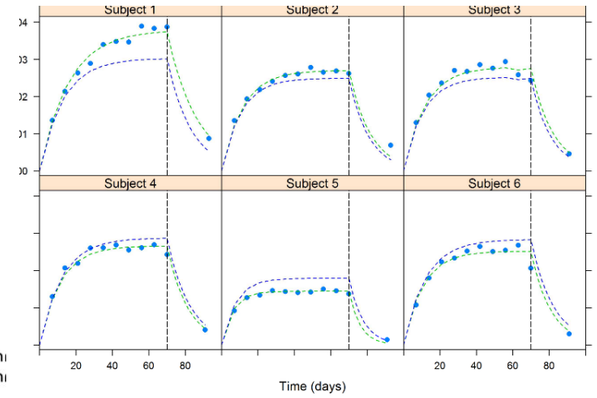


Figure 2: Body water enrichment. Shown are observations (closed blue circles), population predictions (dashed green line) and individual predictions (dashed blue line). The vertical dashed line is the end of the 70 day dosing period.

RESULTS

Although a high deuterium fraction of body water was observed (up to 3.9%, see figure 2), the deuterium fraction measured in BGalC was markedly lower (up to 0.14%, see figure 3). The deuterium fraction in GalC remained relatively stable during the study period, in subjects 1,2,4 and 5 even 100 days after the last dose of D_2O. A compartment turnover model best characterized the deuteration of body water and BGalC. The differences in BGalC deuteration were explained by interindividual variability in body water turnover. The estimated turnover rate of BGalC is 413 days (352 - 499, 95% confidence interval).

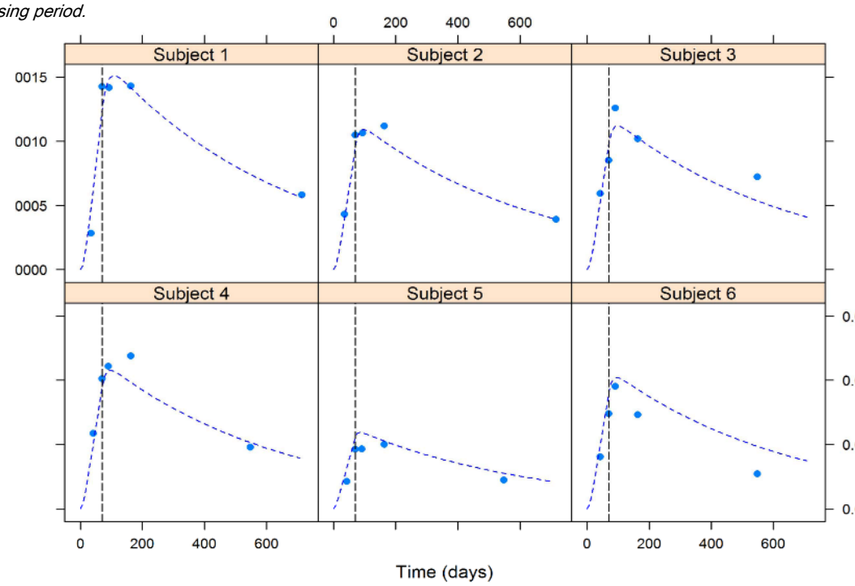


Figure 3: BGalC enrichment. Shown are observations (closed blue circles) and model predictions (dashed blue line). The vertical dashed line represents the end of the 70 day dosing period.

CONCLUSIONS

A slow rate-limiting biochemical step is suggested by the data and estimation of BGalC turnover rate. This is likely due to the production and/or degradation of myelin.

The estimated BGalC turnover rate is a potential, quantitative biomarker for myelin kinetics.

Further research:

- Repeat study in MS patients, eventually with remyelinating compound
- This approach may be applicable for other CSF biomarkers.

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