Anisotropy, Compartmentalization, and Anomalous Diffusion of Intracellular Metabolites in the Axons and Glia of the Human Brain at 7T

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Target Audience Those interested in new, compartment-specific tools to probe neural tissue with diffusion weighted spectroscopy (DWS).

Purpose Anomalous diffusion has been reported in several studies of neural tissue[1-3]. Recently, there was the first report of anomalous diffusion in DWS measurements of intracellular metabolites in the rat brain at 7T utilizing ultra-short diffusion times (Δ < 10 ms) enabled by oscillating gradients[4]. Here, we show the first *in vivo* measurements of anomalous diffusion for N-acetylaspartate (tNAA), (Phospho)choline (tCho), and (Phospho+glycerophospho)creatine (tCr) in the human brain at 7T with conventional diffusion times (Δ ~ 60 ms). As tNAA is almost exclusively present in the intra-axonal space, and whereas tCho and tCr exist both within the axon and the glia structures, diffusion of these species allows for clear investigations of the effects of both anisotropy and compartmentalization when considering the relatively organized fiber tracts in the body of the corpus callosum (BCC)[5]. To characterize the diffusion, we make use of the continuous time random walk (CTRW) theory, in which the Fick's 2nd Law in Eq. (1) is generalized to fractional order for the time and space derivatives to $0 < \alpha \leq 1$ and $1 < \beta \leq 2$, respectively. In CTRW theory, the solution to Eq. (1) in the q domain is given by Eq. (2) as the Mittag-Leffler function (MLF), E_α, which through α and β are able to capture diffusion decay signals that are not mono-exponential. Conveniently, estimations of α and β allow for the mean squared displacement (MSD) in Eq. (3) to be characterized in power-law form as sub-($2\alpha/\beta < 1$), Gaussian ($2\alpha/\beta = 1$), or super-diffusion ($2\alpha/\beta > 1$)[3].

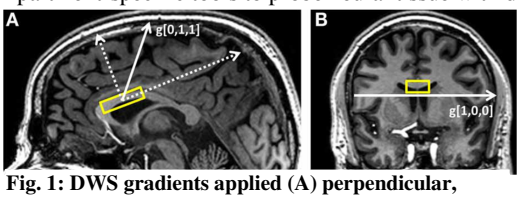

Fig. 1: DWS gradients applied (A) perpendicular, g[0,1,1], and (B) parallel, g[1,0,0], to the principal fiber direction of the BCC.

$$\frac{\partial^\alpha P(x,t)}{\partial t^\alpha} = D\frac{\partial^\beta P(x,t)}{\partial x^\beta} \quad (1) \qquad p(q,\Delta) = E_\alpha\left(-Dq^\beta\Delta^\alpha\right) \quad (2) \qquad \langle x^2(t)\rangle \sim t^{2\alpha/\beta} \quad (3)$$

Methods 10 healthy volunteers (26 ± 4 years) were scanned on a 7T Philips Achieva MRI system. Fig. 1 shows the 2 cm^3 volume of interest (VOI) in the BCC. The DWS data were acquired with a point-resolved spectroscopy (PRESS) sequence TR = 3 s, TE = 121 ms with a bipolar diffusion-weighting scheme and cardiac synchronization. Two directions with respect to the VOI coordinates were chosen for the diffusion weighting: one mostly perpendicular to the BCC fibers denoted as g[0,1,1] in Fig. 1A, and one mostly parallel to the direction of the BCC fibers denoted as g[1,0,0] in Fig. 1B. Diffusion-weighting parameters were: Δ = 60.5 ms, δ = 34 ms, τ = 17 ms, and 8 arrayed gradient amplitudes to produce a set of *b*-values of 0 – 6594s/mm^2 for g[0,1,1] and 0 – 3297 s/mm^2 for g[1,0,0]. The spectra were analyzed using LCModel[6] and the DWS data were fit to Eq. (2) using the Levenberg-Marquardt algorithm and custom code in Matlab.

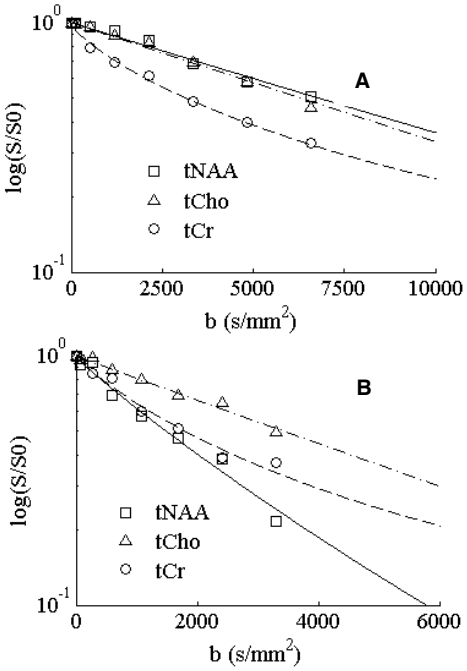

Fig. 2: Signal plots and MLF fits with diffusion weighting perpendicular (A) and parallel (B) to the principal fiber direction of the BCC.

Table 1: CTRW parameters, parallel and perpendicular measurements combined. p<0.01 ** and p<0.001 * indicate differences in mean values of tCr compared to tNAA and tCho.**

	tNAA	tCho	tCr
α	0.95 ± 0.06	0.94 ± 0.08	0.63 ± 0.14***
β	1.81 ± 0.16	1.82 ± 0.15	1.65 ± 0.18**
$2\alpha/\beta$	0.99 ± 0.13	1.03 ± 0.06	0.76 ± 0.15***

Results Measurements of the diffusion coefficient, *D*, are consistent with measurements made in previous DWS studies[4-5], and thus are not reported here. Fig. 2 shows example log-linear plots of the diffusion decay signals of tNAA, tCho, and tCr in the perpendicular (2A) and parallel (2B) orientations to the BCC. The fits to the MLF in Eq. (2) show that for both Figs. 2A and 2B, tNAA and tCho decays follow straight lines, representing Gaussian diffusion. However, the fits for the tCr data in Figs. 2A and 2B show clear deviation from a straight line as is apparent in the curved decay, possibly representing anomalous diffusion. In general, for each of the metabolites, there was no significant difference in estimations of α and β with respect to perpendicular and parallel gradient orientations. As such, the perpendicular and parallel CTRW measurements were combined in Table 1. For tNAA and tCho, the power law exponent in Eq. (3) confirms Gaussian dynamics ($2\alpha/\beta$ ~ 1) of the MSD, whereas for tCr, $2\alpha/\beta$ ~ 0.76 indicating sub-diffusive behavior of the MSD. Furthermore, these CTRW parameter estimations for tNAA, tCho, and tCr were consistent when performing additional *b*-value acquisitions up to ~10600 s/mm^2 in the perpendicular direction and up to ~5300 s/mm^2 in the parallel direction.

Discussion This study suggests that for the metabolites studied: 1) white matter anisotropy does not contribute to Gaussian (tNAA, tCho) or anomalous (tCr) diffusion patterns, 2) compartmental distribution does not necessarily contribute to anomalous diffusion as both tCho and tCr exist within the axons and glia, and 3) the unique sub-diffusive behavior of tCr may be influenced by other factors such as molecular weight differences between Cr and PCr[3], chemical exchange, binding, or active transport[4]. Although the tCr spectra provides, to our knowledge, the first report of anomalous diffusion of metabolites *in vivo* in the human brain, caution should be considered in explicitly excluding tNAA and tCho as the PRESS sequence produces data the long diffusion time regime, whereas others[4] have found that tNAA, tCho, and tCr in the rat brain exhibit anomalous diffusion at ultra-short diffusion time measurements which can only be performed with oscillating gradient acquisitions on animal systems.

Conclusion The CTRW approach to measure the DWS of intracellular metabolites provides new insight into neural tissue microstructure.

References [1] De Santis S, et al. Magn Reson Med 2011. [2] Ozarslan E, et al. Neuroimage 2012. [3] Ingo C, et al. Magn Reson Med 2013. [4] Marchadour C, et al. J Cereb Blood Flow Metab 2012. [5] Ronen I, et al. Front Integr Neurosci 2013. [6] Kan H E, et al. Magn Reson Med 2012.