Time-Dependent Influence of Cell Membrane Permeability on MR Diffusion

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Target audience: Researchers interested in diffusion weighted imaging and measurements of ADC.

Purpose: Cell membrane permeability has an important influence on MR diffusion measurements. For example, increased cell membrane permeability following apoptotic cell death[1] can significantly increase the apparent diffusion coefficient (ADC) because water is no longer as restricted. Although several models have previously been developed to account for permeability effects, recent studies that have used diffusion data to extract axon or cell sizes have usually ignored water exchange between the intra- and extracellular spaces for simplicity. This approximation assumes that the cell membrane permeability is small, so the intracellular lifetime is much longer than the diffusion time (Δ_{eff}). Intuitively, the influence of cell membrane permeability should be less pronounced at shorter diffusion times. However, the precise nature of this time-dependent influence has not been comprehensively quantified. In the current study, we developed an experimental protocol to selectively alter cell membrane permeability, and then investigated its influence on diffusion measurements over a broad range of effective diffusion times (0.4 ms < Δ_{eff} < 3000 ms).

Methods: Saponin is a natural detergent that selectively removes membrane cholesterol and thereby provides a selective way to increase membrane permeability without significantly altering other cell properties. Human myelogenous leukemia K562 cells were cultured and divided into three groups, each treated with vehicle (control), low (0.025% w/v) and high (0.05%) concentrations of saponin. There were six samples in each group. NMR diffusion measurements were performed on a Varian 7T scanner with a 12 mm micro-gradient coil with maximum gradient strength up to 1.42 T/m. ADC values were acquired with Δ_{eff} ranging from 0.4 to 3000 ms. Cosine-modulated oscillating gradient spin echo (OGSE) measurements were performed to achieve short Δ_{eff} ranging from 0.42 – 5 ms (i.e. ≈50 – 600 Hz with a gradient duration of 20 ms), while stimulated echo acquisitions (STEAM) were used to achieve longer Δ_{eff} from 11 to 2999 ms with a pulsed gradient duration of 3 ms. The cell membrane permeability of each sample was estimated using constant gradient (cg-) experiments[3].

Results and discussions: Fig. 1 shows the measured ADCs of samples acquired using STEAM (Fig. 1.a) and OGSE (Fig. 1.b). The estimated mean cell membrane permeabilities acquired using the cg-experiments were 0.011, 0.019, and 0.044 μm/ms for control, low (0.025%) and high (0.05%) concentrations of saponin, respectively, corresponding to intracellular lifetimes of 317, 183, and 82 ms. It is evident that the influence of membrane permeability on diffusion measurements was very different at different diffusion times. When Δ_{eff} was long (> 1000 ms), ADC was relatively independent of Δ_{eff}, and ADC differences between groups were maximized (0.45 μm^2/ms). This is consistent with previous results that ADC is determined by membrane density and permeability at long diffusion times. When Δ_{eff} was small (< 5 ms), ADC differences between groups became small (< 0.1 μm^2/ms) and completely disappeared when Δ_{eff} < 1 ms. This shows that the influence of cell membrane permeability is negligible with very short diffusion times. Interestingly, when Δ_{eff} is intermediate (i.e. 10 ms < Δ_{eff} < 60 ms), which is typical for PGSE-based measurements, the measured ADC can significantly increase (~ 0.1 μm^2/ms) with a slight increase in permeability (0.011 to 0.019 μm/ms).

Conclusion: The influence of membrane permeability on diffusion MR measurements is highly dependent on the choice of diffusion time. It can be negligible, but only when the diffusion time is very short, such as the case for OGSE measurements at moderately high frequency. By contrast, permeability has a major influence on diffusion measurements for intermediate/long diffusion times obtained in typical PGSE measurements.

References: (1) Bailey C, Giles A, Czarnota GJ, Stanisz GJ. Magn. Reson. Med. 2009; 62(1): 46-55. (2) Xu J, Does MD, Gore JC. Magn. Reson. Imaging. 2011: 29(3): 380-390. (3) Meier C, Dreher W, Leibfritz D. Magn. Reson. Med. 2003; 50(3): 510-514.

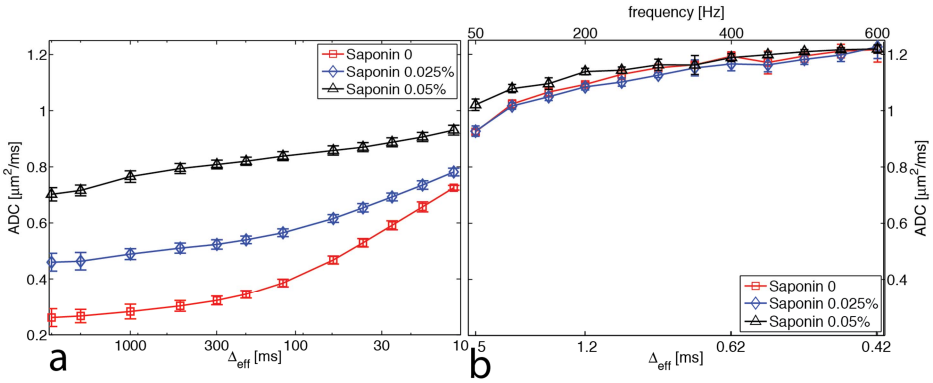

Fig. 1. The diffusion time (Δ_{eff}) dependent apparent diffusion coefficients (ADCs) by STEAM (a) or OGSE (b) of cells treated with different concentrations of saponin. The error-bars represent the inter-sample standard deviation in each group.