Encapsulated Gadolinium and Dysprosium Ions Within Ultra-Short Carbon Nanotubes for MR Microscopy at 11.75 and 21.1 T

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Introduction

Single-walled carbon nanotubes (SWCNT) have gained interest in recent years for their biocompatibility and multifunctional applications, such as drug delivery [1,2]. Ultra-short SWCNT have demonstrated the capability to encapsulate Gd^{3+} (GNT) and have been used to label cells for MRI [3]. In contrast to Gd, which displays decreased relaxation rates at high magnetic fields, Dy^{3+} shows the opposite trends above 3 T. The hollow interior and carbon surface may present additional benefits for lanthanide-doped US-tubes at higher magnetic fields due to nanoscale confinement and favorable water exchange. This study evaluates GNT and Dy-doped SWCNT (DNT) at 11.75 and 21.1 T both in solution and with a murine microglial cell line (Bv2).

Experimental

GNT and DNT were synthesized with methods outlined by others [3,4]. MRI was performed at 11.75 T and 21.1 T at the NHMFL. For solution experiments, GNT and DNT were serial diluted from stock solution at four concentrations to measure r_1 and r_2 relaxivity. For *in vitro* cell experiments, a rat Bv2 microglia cell line was used following methods outlined previously [3, 5]. Bv2 cells were transfected for 12 h with DNT and GNT at a final lanthanide concentration of 17 μM. Following harvest, 150k cells were immobilized with 2% agarose in a 10-mm NMR tube. DNT and GNT labeled cells were imaged together with cells exposed only to empty SWCNT (no lanthanide) and with unlabeled cells as controls. For T_1 and T_2 relaxation measurements, single slice 2D spin-echo (SE) sequences were used with TR and TE times varied. In addition, a 3D gradient recalled echo (GRE) sequence was acquired at 50-μm isotropic resolution with TE/TR = 7.5/150 ms.

Table 1: Relaxivity of DNT and GNT at 11.75 and 21.1 T

	r_1 ($mM^{-1}s^{-1}$)		r_2 ($mM^{-1}s^{-1}$)	
	11.75 T	21.1 T	11.75 T	21.1 T
DNT	**1.05**	**1.91**	**322.7**	**472.2**
Adj R^2	1.0	0.99	1.0	1.0
GNT	**17.25**	**8.62**	**352.4**	**400.9**
Adj R^2	0.94	0.94	1.0	1.0

Results

Comparing the effect of increased field strength in solution (**Table 1**), GNT show an overall larger r_1 that decreases at 21.1 T. DNT, on the other hand, shows an increase in r_1 value consistent with theoretical expectations [6,7]. For r_2, both DNT and GNT show an increase with field strength with DNTs having a larger increase. At 21.1 T, DNTs outperform GNTs as expected with the square dependence on magnetic field. When GNTs and DNTs are incorporated into Bv2 cells, T_1 contrast is quenched while T_2 and T_2* are the dominating contrast mechanisms (**Figure 1**). As in solution, the DNTs are the more effective intracellular contrast agent with a 5.2 ms shorter T_2 and a much larger susceptibility effect with a 8.2 ms shorter T_2*.

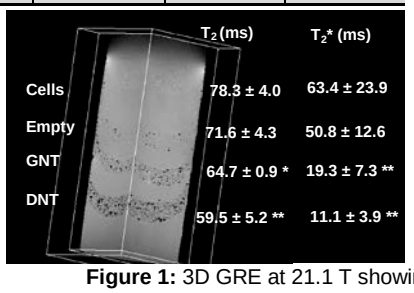

Figure 1: 3D GRE at 21.1 T showing increased contrast for DNT.

Conclusions

This study demonstrates that the DNTs provide a strong T_2/T_2*-agent at 21.1 T, potentially with higher contrast and detectability than the GNTs. Furthermore, both of these nanoparticle agents outperform many MRI contrast agents at high magnetic field, including some iron oxides. Due to the high relaxivities demonstrated both in solutions and with intracellular labeling, Gd^{3+} and Dy^{3+} encapsulated within US-tubes should provide excellent cell detectability for *in vivo* MRI-based cellular tracking.

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