Synthesis and evaluation of a biodegradable macromolecular contrast agent containing macrocyclic Gd(III) chelates for cancer MRI

Zhen Ye[1,2], Xueming Wu[2], and Zheng-Rong Lu[2]

[1]*Pharmaceutics and Pharmaceutical Chemistry, University of Utah, Salt Lake City, UT, United States,* [2]*Biomedical Engineering, Case Western Reserve University, Cleveland, Ohio, United States*

Introduction:

Macromolecular Gd(III) chelates are more effective MRI contrast agents than low molecular weight Gd(III) chelates and provide better characterization of tumor vascularity as shown in preclinical studies. However, the slow-excretion and prolonged tissue retention of macromolecular contrast agents have raised significant safety concerns. Polydisulfide Gd(III) complexes have been developed as biodegradable macromolecular MR contrast agents to facilitate excretion of Gd(III) chelates and to alleviate the safety concerns. The agents can be readily degraded and excreted via renal filtration with minimal tissue retention [1]. However, the reported polydisulfide Gd(III) complexes are based on linear Gd(III) chelates of a DTPA derivative, which has poor kinetic stability. Macrocyclic Gd(III) chelates have shown much higher kinetic chelation stability than linear chelates [2]. It is desirable to develop a new generation of biodegradable contrast agents based on a macrocyclic ligand, such as DOTA. In this study, we have synthesized and evaluated a neutral polydisulfide containing Gd-DOTA monoamide as a new biodegradable macromolecular MRI contrast agent with high kinetic stability. We have investigated the biodegradability, kinetic stability and in vivo contrast enhancement of the agent.

Materials and Methods:

The contrast agent, N^1-lysylethylenediamine Gd-DOTA monoamide and dithiobis-propionic acid copolymers (GOLS), was synthesized by copolymerization of N^1-Lysylethylenediamine DOTA monoamide and N-hydroxysuccinimide dithiobispropionate, followed by Gd(III) complexation. The physicochemical properties of GOLS, including molecular weight, Gd(III) content and relaxivities, were determined using size exclusion chromatography (SEC), ICP-OES and the Bruker® Minispec relaxometer (1.5T, 60 Hz). The degradability of GOLS was evaluated by incubating with L-cysteine (15 μM) in PBS at physiological pH and 37ºC. The kinetic stability of the agent was studied by incubating with and without endogenous metal ions Cu^{2+} and Zn^{2+} and compared to that of DTPA based polydisulfide Gd(III) complex, GDCC [3]. The efficacy for tumor contrast-enhancement of GOLS was investigated in an orthotopic 4T1 mouse breast cancer model on a Bruker 9.4T MR Biospec small animal scanner using ProHance® as a control. Statistical analysis was performed using a paired two-tailed Student's t-test.

Results:

The structure of GOLS is shown in Figure 1. The number average and weight average molecular weights of GOLS were 26.5 and 38.6 kD, respectively. The T_1 and T_2 relaxivities of GOLS were 7.20 $mM^{-1}s^{-1}$ and 9.72 $mM^{-1}s^{-1}$ per Gd at 1.5T. A gradual decrease of molecular weight of GOLS was observed at 30, 60, 120 and 240 minutes post incubation of cysteine at plasma concentration (Figure 2). Figure 3 shows the kinetics stability against transmetallation of GOLS and GDCC in incubation with endogenous metal ions Cu^{2+} and Zn^{2+}. The kinetic stability of GOLS was much higher than that of GDCC. Little release of free Gd(III) ions was detected with the macrocyclic agent GOLS. Figure 4 shows the axial T_1-weighted 2D spin echo MR images of the tumor tissues of the mice bearing orthotopic 4T1 breast tumor before and after injection of GOLS and ProHance®. GOLS resulted in significantly higher tumor enhancement than the clinical agent ProHance®.

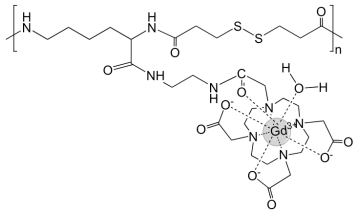

Figure 1. Chemical structure of N^1-Lysyl-ethylenediamine Gd-DOTA monoamide and dithiobispropionic acid copolymer (GOLS)

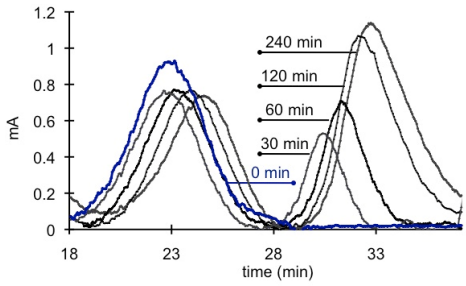

Figure 2. Molecular weight distribution of GOLS before (0 min) and at different time points post incubation with cysteine at plasma concentration (15 μM) under physiological pH and 37°C.

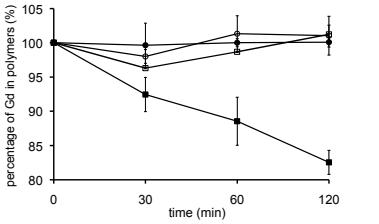

Figure 3. Kinetic stability of GDCC (square) and GOLS (circle) with (filled) or without (no fill) incubation of Cu^{2+} and Zn^{2+} at plasma concentra-tions in PBS (pH=7.4).

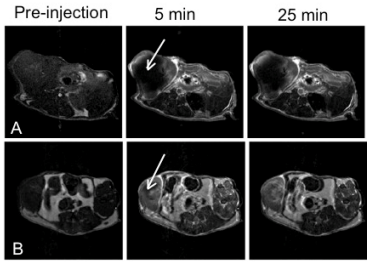

Figure 4. T_1-weighted 2D spin-echo images (axial) of mice bearing 4T1 mouse breast cancer before and at 5 and 25 minutes post injection of ProHance® (A) and GOLS (B) at a dose of 0.1 mmol-Gd/kg.

Conclusion:

A neutral biodegradable macromolecular contrast agent based on macrocyclic Gd chelates, GOLS, was synthesized and evaluated for MR cancer imaging. Our preliminary study showed that the macromolecules can be readily degraded, and the agent had higher kinetic stability against transmetallation than the agent based on linear Gd chelates. In vivo MR study showed that GOLS generated significantly higher tumor enhancement than a clinical agent. GOLS is promising as a safe and effective contrast agent in contrast enhanced MRI.

References:

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