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Contrast Agents for Magnetic Resonance Imaging Synthesized with Ring-Opening Metathesis Polymerization

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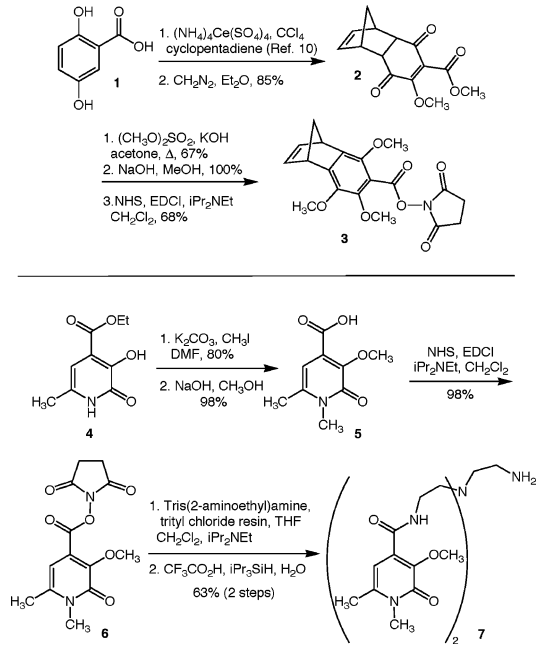
Magnetic resonance imaging (MRI) is indispensable for diagnostic clinical medicine and biomedical research.[1] By enhancing specific features of an image, contrast agents improve the sensitivity and therefore utility of MR images. Poly(amino carboxylate)-based Gd^{III} chelates are currently the only agents used clinically. Recently, Raymond and co-workers developed hydroxypyridonate (HOPO)-based Gd^{III} chelates that are more effective at enhancing the contrast of MR images.[2,3] This attribute of HOPO complexes encourages the development of even more effective chelates. Here, we describe oligomeric HOPO-based Gd^{III} chelates generated by using ring-opening metathesis polymerization (ROMP). This approach provides contrast agents with extraordinary sensitivity and versatility.

The strength of Gd^{III}-based contrast agents can be improved by increasing the number of coordinated water molecules, optimizing the water exchange rate between bound and bulk water molecules, increasing the rotational correlation time, or increasing the number of Gd^{III} ions per molecule.[4] The HOPO-based agents have an increased number of water molecules in the innersphere environment and a near optimal water exchange rate; together, these features lead to a higher relaxivity.[3] Increasing the rotational correlation time can further improve these agents as with poly(aminocarboxylate)-based systems involving proteins, linear polymers, dendrimers, and micellar aggregates.[1,5] We reasoned that highly sensitive and tunable contrast agents could be made through incorporation of multiple HOPO-based Gd^{III} chelates into an easily functionalizable macromolecule.

Our strategy for generating contrast agents with the desired attributes relies on ROMP. We envisioned that this synthetic strategy could afford the first multivalent HOPO-based agents. ROMP is an ideal polymerization method for this purpose because it can give rise to polymers with multiple sites that can be functionalized.[7] Such flexibility is beneficial because the utility of contrast agents can be increased by equipping them with targeting moieties or fluorescent probes. Moreover, the use of ROMP is advantageous because it can yield polymers of well-defined length.[6] Thus, our strategy allows for multiple Gd^{III} ions to be incorporated into polymers of varying lengths, which should yield a series of agents with controlled relaxivities. We embedded a HOPO-based chelating moiety within a benzonorbornadiene unit that constitutes the backbone of the polymer upon ROMP. We integrated the chelator into the backbone to increase the rotational correlation time of the resulting macromolecules.

Polymeric contrast agents **10a** and **10b** were generated via a modular synthesis from benzonorbornadiene monomer **3** and HOPO-chelator **7** (Scheme 1). This strategy utilizes the novel attributes of the new monomer **3**: a lanthanide binding catechol and a reactive site from which a complete chelate can be generated. This convergent strategy provides a means to use the succinimidyl esters to conjugate other groups, for example, for targeting or fluorescence imaging.

Scheme 1. Synthetic Route to Building Blocks Used in the Assembly of Multivalent Imaging Agents



To synthesize the target polymers **8a** and **8b** we employed the ruthenium initiator $(H_2IMes)(3\text{-Br-py})_2(Cl)_2Ru{=}CHPh$. Its rate of initiation relative to propagation affords polymers of well-defined average lengths.[6,8] Another critical feature of this catalyst is functional group tolerance; our monomer **3** contains phenolic ether and activated ester groups. Initiation occurred at 0 °C with complete consumption of **3** occurring in <5 h. After the starting material was consumed, the ketone-containing enol ether was added to terminate the polymerization. The ketone terminator provides a site for orthogonal functionalization via its ability to form adducts with nucleophiles such as hydrazides.[9] Monomer-to-initiator ratios of 10:1 and 30:1 were used to explore the effect of various polymer lengths on contrast agent strength; the resulting polymers were characterized by using gel permeation chromatography and NMR spectroscopy.

From **8a** and **8b**, oligomeric contrast agents were generated (Scheme 2A). Compound **8a** was reacted with 0.25 equiv per monomer of HOPO-chelator **7** and 0.75 equiv per monomer of guanidinium-substituted spacer amine to yield oligomer **9a**. The guanidinium-containing amine was used to cap any unreacted sites, prevent problems that could arise from having too many chelates per polymer, and increase the aqueous solubility of the polymer. Poor water solubility is a major drawback of HOPO-type chelates, and methods to improve solubility are currently being investigated by other research groups.[11] Subsequent methyl ether removal and metalation with $GdCl_3$ yielded agent **10a**. Polymer **8b** was treated in a similar fashion to yield **10b**. Additionally, the monoionic

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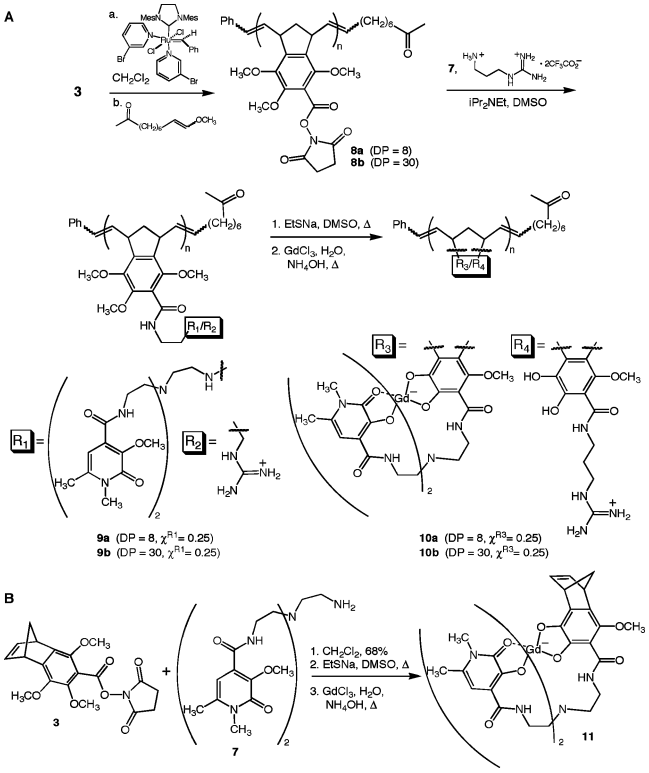
Scheme 2. (A) Synthetic Route to Multivalent Imaging Agents[a] and (B) Synthesis of Monomeric Chelator **11** for Comparison to Multivalent **10a** and **10b**



[a] Compounds **8**−**10** are shown as a single regioisomer; however, head-to-head and head-to-tail isomers are generated in the polymerization reaction. DP denotes the degree of polymerization.

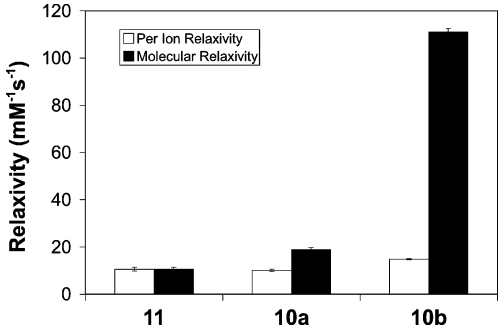


Figure 1. Gd^{III} ionic and molecular relaxivities at 60 MHz. Error bars represent 1 standard deviation.

complex **11** was generated for control measurements (Scheme 2B). As predicted, **10b** has an aqueous solubility (≥10 mM) that is superior to that of monoionic HOPO-based agents (<0.5 mM).[5a]

To evaluate the ability of agents **10a**, **10b**, and **11** to enhance contrast in MR images, the relaxivity values of these complexes were ascertained (Figure 1). Relaxivities were determined as the slope of the line generated by plotting the inverse of T_1 relaxation time versus concentration. The monomeric agent **11** has a relaxivity of 10.5 ± 0.8 mM^{-1} s^{-1}, which is similar to that of other HOPO-based agents and nearly 3-fold that of clinically used agents.[2] A significant increase in per Gd^{III} ion relaxivity was not observed upon transition from the monomeric agent to the shorter oligomeric agent; however, an increase in relaxivity of 1.4-fold per Gd^{III} ion was observed for the longer polymer. Not only is the Gd^{III} relaxivity (light bars) increased by 1.4-fold but also the molecular relaxivity (dark bars) increases 10.6-fold upon incorporation of multiple Gd^{III} chelates. Polymer **10b** has a molecular relaxivity of 111 ± 1.5 mM^{-1} s^{-1} (Figure 1); when compared with agents used in the clinic, this value is nearly 30-fold greater.

Our approach of incorporating multiple HOPO-type Gd^{III} chelates using ROMP yields polymers with extremely large molecular relaxivities. These large relaxivity values are proportional to rotational correlation times estimated from the molecular weights of linear polymers of Gd^{III} diethylenetriaminepentaacetic acid.[1a,12] The features of ROMP-derived polymers allow for functionalization of the monomer units and termini through orthogonal chemistry to yield multivalent, target-specific contrast agents. We envision that this strategy can be optimized to create hypersensitive, targeted imaging agents.

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Supporting Information Available: Synthetic methods and experimental details (PDF). This material is available free of charge via the Internet at http://pubs.acs.org.

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