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A greener approach toward gadolinium-based contrast agents†

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Gadolinium-based contrast agents are widely used to enhance the contrast of images in magnetic resonance imaging procedures. In particular, Gd-DTPA (Magnevist™) and Gd-DOTA (Dotarem™) were the first contrast agents used in clinical practice. Herein, a new environmentally friendly synthetic approach for Gd-DTPA, Gd-DOTA and their derivatives has been described using ultrasound energy. Our protocol provides a simple procedure and short reaction times as well as elevated yields and purities. Additionally, in this novel method, no organic solvent is involved at any stage of the reaction, isolation or purification of products.

1 Introduction

Since the pioneering work of Lauterbur in 1973,[1] magnetic resonance imaging (MRI) has emerged as a powerful medical imaging tool. MRI technology has improved clinical diagnostics by enabling the noninvasive evaluation of various processes in complex living systems with superior spatial resolution and contrast.[2,3] Compared with other radio-diagnostic techniques, the most notable advantage of MRI is the use of lower-energy radiation understating the tissue damage produced by ionizing radiation exposure. It is important to note that epidemiological evidence has linked increasing exposure to such radiation with the development of cancers.[4]

The physical principle of MRI is based on the monitoring of spatial variation of hydrogen longitudinal (T_1) and transversal (T_2) magnetic relaxation times of water molecules. Indeed, T_1 and T_2 are higher sensitive to the biochemical conditions in healthy and pathological tissues displaying anatomical structural information with elevated contrast.[5] However, in some examinations, *e.g.*, the gastrointestinal tract or cerebral area, the relaxation times are sometimes superimposed onto one another, which makes dynamic scanning and thus their measurement difficult.[6] In these cases, the use of paramagnetic contrast agents has been detrimental to contrast enhancement and image resolution.[6]

Gadolinium(III)-based complexes have been the most widely utilized contrasting agents.[7] In these compounds, the metal ion is involved *via* a multidentate organic ligand forming resistant structures to transmetallation reactions from Zn(II) and Ca(II) ions, the two main endogenous competitors.[7,8] Since the late 1980s, several gadolinium(III)-based contrast agents have been approved for clinical use, such as gadopentetic acid (Gd-DTPA; Magnevist™) and gadoteric acid (Gd-DOTA; Dotarem™). These complexes were the first to be clinically employed for MRI and have been described as references for the development of novel contrast agents.[6,7]

Regarding preparation methods, the contrast agents have been synthesized under conventional thermal heating with long reaction times and with successive purification steps to obtain products of satisfactory purity.[9–12] Within an industrial context, longer reaction times and several purification steps are related to increased energy expenditure, increased time for the change of processes, higher operational risks, and higher waste generation compared with accelerated reactions with reduced byproducts. Moreover, the increasing number of unitary operations and personal involvement results in high production costs. Hence, the development of new synthetic strategies toward the preparation of these important compounds using a simple, economic and rapid method is of great significance and should be explored.

As an alternative to classical methods, sonochemistry offers a versatile and facile pathway for a large variety of chemical reactions and processes in accordance with sustainable chemistry concepts.[13] The use of this virtually innocuous energy has been reported to show notable results in synthetic chemistry with a remarkable rate of enhancement and an extraordinary reduction in reaction times.[14] Notwithstanding its similarities with mechanochemical activation,[15] the mechanism of such method relies on the acoustic cavitation phenomenon, which leads to the formation, growth, and adiabatic collapse of bubbles in liquid medium. This effect induces a very high local pressure and temperature inside the bubbles due to gas compression as well as the improvement of mass transfer and increase of turbulent flow

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in the liquid. The singular conditions attained during ultrasound-assisted processes have been applied to a range of organic and organometallic synthetic systems, resulting in products generated under milder and faster conditions.[16,17]

Therefore, because of the importance of gadolinium-based contrast agents in MRI procedures and in an attempt to improve their synthetic routes, a new, greener ultrasound-assisted method is described. Herein, the synthetic protocol employed diethylenetriaminepentaacetic acid (DTPA), 1,4,7,10-tetraazacyclododecane tetraacetic acid (DOTA) and ethylenediaminetetraacetic acid (EDTA) as chelating agents. The preparation of gadopentetic acid (Magnevist™) and gadoteric acid (Dotarem™) are also described.

2 Results and discussion

First, our efforts were focused on the synthesis of the two most important gadolinium-based contrast agents, Gd-DTPA (**2**) and Gd-DOTA (**3**). The current methodologies described for the preparation of Gd-DTPA perform this synthesis using variable reaction times from 3 to 48 hours, and the products are obtained with 92–99% yields. These processes include sequential steps, temperatures of 90–100 °C, water as a solvent and recrystallization for the purification of the product.[9–11] The synthetic approach for obtaining the meglumine salt of Gd-DOTA (**3**) has been reported *via* the direct mixture of sodium salt of the complex with one molar equiv. of meglumine (**1**) in water. Likewise, the conventional thermal heating methods for the synthesis of sodium salt of Gd-DOTA require reaction times of 2 to 6.5 hours, resulting in 83–95% yields. Such reactions are performed in water under temperatures of 50–95 °C followed by recrystallization to obtain the pure product.[10,12] Lipophilic Gd-DOTA derivatives have been synthesized in quantitative yields from complexation reactions of DOTA derivatives and gadolinium(III) oxide in water as a solvent at 80 °C for 24 h.[18]

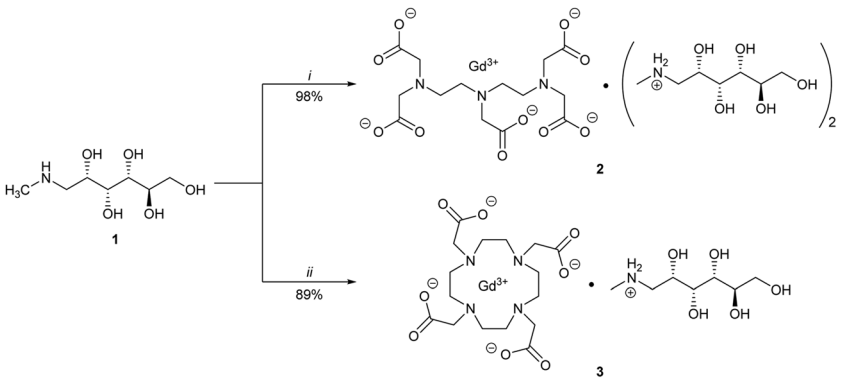
In our described methodology, compounds **2** and **3** were obtained after sonication with 98% and 89% yields, respectively (Scheme 1). The reactions were optimized in an open one-pot system using water as a solvent, thus allowing for the addition of reactants to the reaction mixture during sonication. The synthesis of Gd-DTPA and Gd-DOTA was preceded by salt formation among meglumine (**1**) and DTPA or DOTA, respectively, in a molar ratio of 1 : 1. This process increased the solubility of the ligands and the chemical rate of complexation with the subsequent formation of a host-guest system *via* the addition of gadolinium(III) oxide. For compound **2**, another molar equiv. of meglumine (**1**) was also added to the resulting coordination compound. It should be noted that in all experiments performed using ultrasound energy, the change in the addition order of the reactants did not produce the desired products.

Regarding the reaction time, complexes **2** and **3** were synthesized in shorter periods, furnishing the product after 30 and 20 min, respectively (Scheme 1). Moreover, an important aspect of the described protocol was the relative purity of products achieved *via* simple membrane filtration followed by lyophilization. As these products have been used by intravenous administration, their isolation and purification *via* filtration is an advantage in inducing sterile conditions. Additionally, from a sustainability view point, the isolation and purification steps have been reported as the main responsible by waste generated in synthetic chemistry.[19] Hence, to reduce the use of chemicals, solvents, unitary processes and operational time spent maintaining efficiency is highly desirable for any method aiming to increase environmental sustainability in this process.

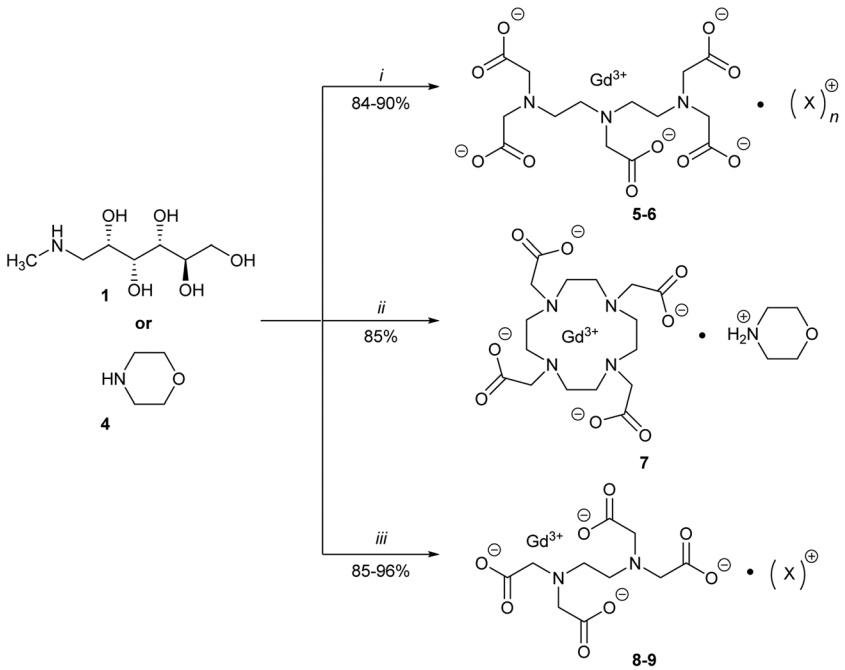
In evaluating the magnitude of waste produced by applying the E-factor[20] as a quantitative measure of greenness, one can conclude that E-factor values in this report are approximately zero. Because the water used in the processes is not considered to be waste, the E-factor values for Gd-DTPA and Gd-DOTA were 0.04 and 0.16, respectively. These reduced values are an important advantage, as the pharmaceutical industry has generated the largest amount of wastes per weight of product with E-factor values in the range of 25–100.[19]

To further explore the potential of this new method for the synthesis of **2** in gram quantities, the reaction was performed in a scale of up to 8 times (approximately 7.5 g). The protocol showed robustness leading to **2** with a 91% yield and elevated purity according to the elemental analysis.

To improve the scope of the sonochemical preparation of gadolinium-based coordination compounds, we attempted to obtain additional Gd-DTPA, Gd-DOTA and Gd-EDTA derivatives. Using the template protocol developed for **2** and **3**, compounds



Scheme 1 Reactants and conditions: (*i*) = (1) DTPA, H_2O, 2 min; (2) Gd_2O_3, H_2O, 10 min; (3) another equiv. of **1**, 18 min. (*ii*) = (1) DOTA, H_2O, 3 min; (2) Gd_2O_3, H_2O, 17 min.



Entry	(x)	*n*	Yield (%)
5	**4**	2	84
6	**1**	1	90
8	**1**	-	85
9	**4**	-	96

Scheme 2 Reactants and conditions: (*i*) = (1) DTPA, H_2O, 2 min; (2) Gd_2O_3, H_2O, 10 min; (3) another equiv. of **4**, 18 min (for compound **5**); (1) DTPA, H_2O, 2 min; (2) Gd_2O_3, H_2O, 28 min (for compound **6**). (*ii*) = (1) DOTA, H_2O, 3 min; (2) Gd_2O_3, H_2O, 17 min. (*iii*) = (1) EDTA, H_2O, 15 min; (2) Gd_2O_3, H_2O, 20 min.

5–9 were obtained in salt form using meglumine (**1**) or morpholine (**4**) in the salification step (Scheme 2). It is noteworthy that **4** had already been described in acid–base reactions procedures for the synthesis of gadolinium(III)-based compounds.[10] Complexes **5–6** were synthesized from DTPA with 84–90% yields after sonication for a total time of 30 min. The Gd-DOTA derivative **7** was prepared similarly to **3** and was achieved with an 85% yield in a reaction time of 20 min. Finally, compounds **8** and **9** were synthesized using EDTA as a chelating agent in 35 min. This hexadentate ligand granted the gadolinium complexes an 84% yield for meglumine's salt **8** and a 96% yield for compound **9** when a morpholine base was used as a counter-ion. When comparing the bases of **1** and **4** in sonochemical reactions, one can note that the meglumine derivative compounds were obtained with better efficiency when the ligands were DTPA and DOTA. However, for EDTA derivative complexes, morpholine produced a better yield (96%) than meglumine (84%).

3 Conclusions

The preparation of gadolinium-based coordination compounds has been successfully accomplished *via* sonochemical conditions. The simplicity of execution, significantly shorter reaction times (20–35 min), satisfactory yields (84–98%), use of water as a solvent and purity of the products make this environmentally friendly methodology highly attractive. Furthermore, under sonication, two of most import gadolinium-based contrast agents (Gd-DTPA and Gd-DOTA) were efficiently obtained in milder and faster conditions, with an E-factor approaching zero. Additionally, the open-vessel condition of this protocol enabled the creation of the products with the desired characteristics by the modification or addition of reactants during the reaction. Finally, the use of the ultrasound-assisted method described may be applicable in the generation of a wide variety of metal-based coordination compounds for important pharmaceutical applications to reduce environmental and production costs.

4 Experimental

4.1 Apparatus and analysis

All common reactants were used as obtained from commercial suppliers without further purification. The reactions were carried out with a standard probe (25 mm) connected to a 1500

Watt Sonics Vibra-Cell ultrasonic processor (Newtown, Connecticut, USA) equipped with integrated temperature control. The device operates at 20 kHz, and the amplitude was set to 20% of the maximum power output. In all experiments, the temperature was raised to 75–80 °C after sonication for 4–6 min and was maintained at this level until the end of the reaction times. High-resolution mass spectra were obtained for all compounds on a LTQ Orbitrap Discovery mass spectrometer (Thermo Fisher Scientific, Bremen, Germany). This hybrid system combines the LTQ XL linear ion trap mass spectrometer and an Orbitrap mass analyzer. The experiments were performed *via* the direct infusion of the sample in CH_3CN:H_2O (50%) (formic acid (0.1%) (flow: 10 μL min^{-1}) in the positive-ion mode) using electrospray ionization (ESI). Elemental composition calculations were executed using the specific tool included in the Qual Browser module of the Xcalibur (Thermo Fisher Scientific, release 2.0.7) software. Fourier transform infrared (FTIR) spectra were recorded using a universal attenuated total reflectance (UATR) attachment on a PerkinElmer Spectrum 100 spectrometer in the wavenumber range of 650–4000 cm^{-1} at a resolution of 4 cm^{-1}. The CHN elemental analyses were performed on a PerkinElmer 2400 CHN elemental analyzer (São Paulo University, USP/Brazil).

4.2 Synthetic procedures

Gadopentate dimeglumine (2). Diethylenetriaminepentaacetic acid (DTPA) (0.393 g, 1.0 mmol) and *N*-methylglucamine (**1**) (0.195 g, 1.0 mmol) were mixed in water (15 mL) in a 50 mL beaker. The reaction mixture was sonicated for 2 min using an ultrasonic probe. Afterward, gadolinium(III) oxide (0.181 g, 0.5 mmol) in water (5 mL) was added to the reaction mixture, and the resulting suspension was sonicated for 10 min. Another molar equivalent of the *N*-methylglucamine (0.195 g, 1.0 mmol) in water (10 mL) was then added to the mixture. After sonication for 18 min, the reaction was allowed to cool to room temperature and was subsequently filtered (Millipore 0.22 μm) and lyophilized. The product did not require subsequent purification, affording 0.922 g (98%) of **2** as a white powder. pH range of 7.7–7.9 (aqueous product solution); FT-IR (UATR, cm^{-1}): 3,242, 1,581, 1,401, 1,088, 710; HRMS (ESI) calcd for $C_{28}H_{54}GdN_5O_{20}$ + H: 939.2676, found. 939.2690 $(M + H)^+$; anal. calcd for $C_{28}H_{54}GdN_5O_{20}$ + H_2O: C, 35.18; H, 5.90; N, 7.33. Found C, 34.68; H, 6.18; N, 7.50%.

Gadoterate meglumine (3). In a 50 mL beaker, 1,4,7,10-tetraazacyclododecane tetraacetic acid (DOTA) (0.101 g, 0.25 mmol) and *N*-methylglucamine (**1**) (0.049 g, 0.25 mmol) were suspended in water (15 mL). The suspension was then sonicated *via* an ultrasonic probe for 3 min. Afterward, gadolinium(III) oxide (0.045 g, 0.125 mmol) in water (5 mL) was added to the mixture and sonicated for an additional 17 min. The reaction mixture was cooled to room temperature, filtered (Millipore 0.22 μm) and lyophilized. The product did not require subsequent purification affording 0.168 g (89%) of **3** as a gray powder. pH 7.9 (aqueous product solution); FT-IR (UATR, cm^{-1}): 3,285, 1,583, 1,391, 1,083, 715; HRMS (ESI) calcd for $C_{23}H_{42}GdN_5O_{13}$ + H: 755.2093, found. 755.2057 $(M + H)^+$; anal. calcd For $C_{23}H_{42}GdN_5O_{13}$ + $(H_2O)_4$: C, 33.45; H, 6.10; N, 8.48. Found C, 33.08; H, 5.84; N, 8.08%.

Gadopentate dimorpholine (5). Compound **5** was prepared using a procedure similar to that used to prepare compound **2**. Diethylenetriaminepentaacetic acid (DTPA) (0.393 g, 1.0 mmol) and morpholine (**4**) (0.087 g, 1.0 mmol) were mixed in water (15 mL) in a 50 mL beaker. The reaction mixture was sonicated using an ultrasonic probe for 2 min. Afterward, gadolinium(III) oxide (0.181 g, 0.5 mmol) in water (5 mL) was added to the reaction mixture, and the resulting suspension was sonicated for 10 min. Another molar equivalent of morpholine (0.087 g, 1 mmol) in water (5 mL) was then added to the mixture. After sonication for 18 min, the reaction was allowed to cool to room temperature, was subsequently filtered (Millipore 0.22 μm) and lyophilized. The product did not require subsequent purification, affording 0.301 g (84%) of **5** as a yellow powder. pH 5.5 (aqueous product solution); FT-IR (UATR, cm^{-1}): 3,371, 1,571, 1,398, 1,096, 708; anal. calcd for $C_{22}H_{39}GdN_5O_{12}$ + H_2O: C, 35.67; H, 5.58; N, 9.45. Found C, 35.44; H, 5.62; N, 9.45%.

Gadopentate monomeglumine (6). Compound **6** was prepared using a procedure similar to that used to prepare compound **2**. A suspension of DTPA (0.393 g, 1.0 mmol) and *N*-methylglucamine (**1**) (0.195 g, 1.0 mmol) in water (15 mL) were sonicated using an ultrasonic probe. After 2 min, gadolinium(III) oxide (0.181 g, 0.5 mmol) in water (5 mL) was added to the mixture. The reaction was then sonicated for 28 min; water (2 × 10 mL) was added in portions to prevent evaporation. The compound was obtained at a 90% yield (0.333 g) as a white powder. pH 2.1 (aqueous product solution); FT-IR (UATR, cm^{-1}): 3,244, 1,579, 1,400, 1,089, 710; HRMS (ESI) calcd for $C_{21}H_{37}GdN_4O_{15}$ + H: 744.1569 Found. 744.1579 $(M + H)^+$; anal. calcd for $C_{21}H_{37}GdN_4O_{15}$ + H_2O: C, 33.15; H, 5.17; N, 7.36. Found C, 33.33; H, 5.24; N, 7.39%.

Gadoterate morpholine (7). Compound 7 was prepared using a procedure similar to that used to prepare compound **3**. In a 50 mL beaker, 1,4,7,10-tetraazacyclododecane tetraacetic acid (DOTA) (0.101 g, 0.25 mmol) and morpholine (**4**) (0.022 g, 0.25 mmol) were suspended in water (15 mL). The rest of the procedure was identical to that performed to obtain the product **3**, affording 0.137 g (85%) of **7** as a colorless powder. pH 5.7 (aqueous product solution); FT-IR (UATR, cm^{-1}): 3,381, 1,589, 1,398, 1,083, 715; HRMS (ESI) calcd for $C_{20}H_{34}GdN_5O_9$ + H: 647.1670 Found. 647.1673 $(M + H)^+$; anal. calcd for $C_{20}H_{34}GdN_5O_9$ + $(H_2O)_4$: C, 33.46; H, 5.90; N, 9.76. Found C, 33.41; H, 5.39; N, 9.71%.

Gadolinium ethylenediamine tetraacetic acid meglumine salt (8). In a 50 mL beaker, ethylenediaminetetraacetic acid (EDTA) (0.146 g, 0.5 mmol) and *N*-methylglucamine (0.097 g, 0.5 mmol) were suspended in water (15 mL). The suspension was then sonicated using an ultrasonic probe for 15 min. Afterward, Gadolinium(III) oxide (0.09 g, 0.25 mmol) in water (5 mL) was added. The reaction mixture was sonicated for an additional 20 min. After cooling to room temperature, the reaction mixture was filtered (Millipore 0.22 μm) and lyophilized. The product did not require subsequent purification, affording 0.271 g (84%) of **8** as a white powder. pH 11.7 (aqueous product solution); FT-IR (UATR, cm^{-1}): 3,287, 1,586, 1,401, 1,095, 722; anal. calcd for

$C_{17}H_{30}GdN_3O_{13}$ + $(H_2O)_3$: C, 29.35; H, 5.22; N, 6.04. Found C, 29.39; H, 5.51; N, 5.92%.

Gadolinium ethylenediaminetetraacetic acid morpholine salt (9). Compound **9** was prepared using a procedure similar to that used to prepare compound **8**. In a 50 mL beaker, ethylenediaminetetraacetic acid (EDTA) (0.146 g, 0.5 mmol) and morpholine (**4**) (0.044 g, 0.5 mmol) were suspended in water (15 mL). The rest of the procedure was identical to that performed to obtain the product **8**, affording 0.256 g (96%) of **9** as a white powder. pH 11.1 (aqueous product solution); FT-IR (UATR, cm^{-1}): 3,240, 1,587, 1,407, 1,096, 715; anal. calcd for $C_{22}H_{39}GdN_5O_{12}$ + $(H_2O)_3$: C, 33.92; H, 8.15; N, 8.99. Found C, 34.12; H, 8.30; N, 9.02%.

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