Synthesis and Metal Complex Selectivity of Macrocyclic DTPA and EDTA Bis(amide) Ligands

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Several macrocyclic bis(amide) derivatives of DTPA and EDTA were prepared. Protonation constants as well as Gd^{3+} and Zn^{2+} complex stability constants of these ligands were determined by potentiometric titrations. The stability constants of the Gd^{3+} complexes increase with increasing ring size of the DTPA bis(amide) macrocycle series, reflecting the enhanced participation of the amide carbonyl oxygens in metal ion coordination. On the other hand, the stability constants of the Zn^{2+} complexes do not exhibit any particular trend throughout the series. The logarithmic selectivity constant of the 15-membered EDTA-DAM ligand for Gd^{3+} over Zn^{2+} (the difference between logarithmic stability constants of Gd(EDTA-DAM) and Zn(EDTA-DAM)) is 6.12. This indicates that the ligand EDTA-DAM exerts by far the highest selectivity for Gd^{3+} over Zn^{2+} among known polyamino polycarboxylates, presumably through a more favorable structural discrimination by this ligand. In contrast, the structural isomer DTPA-EAM shows a less favorable selectivity for Gd^{3+} over Zn^{2+}.

Introduction

At least four gadolinium(III) complexes of polyamino polycarboxylates have been used as magnetic resonance imaging (MRI) contrast agents:[1] $(NMG)_2$[Gd(DTPA)] and (NMG)-[Gd(DOTA)] are both ionic; the other two are nonionic, i.e., Gd(DTPA-BMA) and Gd(HP-DO3A),[2] where NMG is the methylglucamine cation, DTPA is diethylenetriaminepentaacetate, DOTA is 1,4,7,10-tetraazacyclododecane-1,4,7,10-tetraacetate, DTPA-BMA is the bis(methylamide) derivative of DTPA, and HP-DO3A is 10-(2-hydroxypropyl)-1,4,7,10-tetraazacyclododecane-1,4,7-triacetate. To qualify as an effective MRI contrast agent, the complex has to be efficacious (i.e., provide a significant enhancement in the proton relaxation rates of water) and safe at a clinically effective administered dosage. It is known from chelation therapy that metal complexes tend to be less toxic than their constituent free metal ions and free ligands,[3] and it was originally believed that the toxicity of a metal complex was simply related to its thermodynamic stability. The higher the thermodynamic stability, the less the degree of complex dissociation which can potentially lead to toxic free metal ions and ligands. However, a metal complex in vivo may also undergo other types of reactions including ligand displacement and transmetallation. Endogenously available metal ions[4] such as Cu^{2+} and Zn^{2+} can potentially compete for the ligand of a contrast agent and promote the release of Gd^{3+} ions. Indeed, a recent study[5] has demonstrated that the relative thermodynamic stabilities of Gd^{3+} and Zn^{2+} complexes (i.e., ligand selectivities for Gd^{3+} over Zn^{2+}) correlate well with observed acute toxicities for several acyclic Gd^{3+} complexes. The correlation is poor between the complex thermodynamic stability alone and acute toxicity measurements. The study successfully explains why Gd(DTPA-BMA), with a relatively low thermodynamic stability (as compared to other clinically useful Gd^{3+}-based MRI contrast agents) has a surprisingly high LD_{50} (mice) value.

Gd(DTPA-BMA) is the first nonionic MRI contrast agent developed for clinical applications.[6] Phase I–III clinical trials confirmed that this agent is quite effective in enhancing a wide variety of lesions in the brain and spine.[7] This contrast agent also showed satisfactory clinical tolerance in assessment of safety during these studies.[7] The logarithmic selectivity constant of the ligand DTPA-BMA for Gd^{3+} over Zn^{2+} is 4.81, which is the difference between the logarithmic stability constants of Gd-(DTPA-BMA) and Zn(DTPA-BMA). In order to understand ligand selectivity in more detail and to design newer generations of MRI contrast agents, several macrocyclic bis(amide) derivatives of DTPA and EDTA (Figure 1) have been prepared. Macrocyclic ligands have advantages over acyclic ones, potentially being "preorganized" for metal complexation,[8] being able to exert size selectivity, and having the ability to structurally discriminate in binding various metal ions.[9] The synthesis, protonation constants, and Gd^{3+} and Zn^{2+} complex stability constants of a series of amide macrocyclic ligands are reported in this paper.

Experimental Section

Materials and Solutions. $ZnSO_4$, KOH, and Na_2EDTA were purchased from J. T. Baker in DILUT-IT Analytical Concentrate form. $ZnSO_4$ and Na_2EDTA concentrates were diluted to 20 mM with deionized water. Gd^{3+} stock solution (Standard Reference Material, 63.59 mM Gd^{3+} in 10% HCl) was obtained from NBS. HCl standard solution (0.1000 M) was purchased from Fisher Scientific. EM Science was the source for ACS reagent grade KCl crystals.

All reagents used for the synthesis of ligands were obtained from commercial sources unless otherwise noted. 1,5-Diazabicyclo[4.3.0]non-5-ene (DBN) was purified by distillation from CaH_2 and stored over 4-Å molecular sieves. Elemental analyses were performed by Galbraith Laboratories. NMR spectra were obtained on a Bruker AM 250 spectrometer. Positive ion FAB mass spectra were obtained on an HP 5985A spectrometer using a glycerol matrix and xenon gas.

Synthesis. The DTPA bis(amide) macrocycles were synthesized by the condensation of the appropriate diamine with DTPA bis(anhydride) according to the following general procedure. In a typical preparation the diamine and 1,5-diazobicyclo[4.3.0]nonene (DBN) were dissolved in

(1) (a) Watson, A. D.; Rocklage, S. M.; Carvlin, M. J. Contrast Media. In *Magnetic Resonance Imaging*, 2nd ed., D. D., Stark, W. G., Bradley, Jr., C. V. Mosby, C. V., Eds.; Mosby Year Book, Inc.: St. Louis, MO, 1991. Chapter 14. (b) Chang, C. A. *Eur. J. Solid State Inorg. Chem.* **1991**, *28*, 237–244.
(2) Chang, C. A.; Sieving, P. F.; Watson, A. D.; Dewey, T. M.; Karpishin, T. B.; Raymond, K. N. *J. Magn. Reson. Imaging* **1992**, *2*, 95–98.
(3) Lauffer, R. B. *Chem. Rev.* **1987**, *87*, 901–927.
(4) Rubin, M.; Martell, A. E.; Gohil, R.; Penhos, J. C.; Weiss, P.; Motekaitis, R. S. *Inorganic Chemistry in Biology and Medicine*; ACS Symposium Series 391; American Chemical Society, Washington, DC, 1980.
(5) Cacheris, W. P.; Quay, S. C.; Rocklage, S. M. *Magn. Reson. Imaging* **1990**, *8*, 467–481.
(6) (a) Quay, S. C. PCT 86/02841, 1986. (b) Quay, S. C. U.S. Patent 4,687,659, 1987.
(7) (a) Van Wagoner, M.; O'Toole, M.; Quay, S. C. *Invest. Radiol.* **1990**, *25*, S39–S41. (b) Kaplan, G. D.; Aisen, A. M.; Aravapalli, S. R. *J. Magn. Reson. Imaging* **1991**, *1*, 57–62. (c) Greco, A.; McNamara, M. T.; Lawthiez, P.; Quay, S. C.; Michelozzi, G. *Radiology* **1990**, *176*, 416–456.
(8) Cram, D. J. *Science* **1988**, *240*, 760–767.
(9) (a) Adam, K. R.; Dancey, K. P.; Leong, A. J.; Lindoy, L. F.; McCool, B. J.; McPartlin, M.; Tasker, P. A. *J. Am. Chem. Soc.* **1988**, *110*, 8471–8477. (b) Chang, C. A.; Ochaya, V. *Inorg. Chem.* **1986**, *25*, 355–358.

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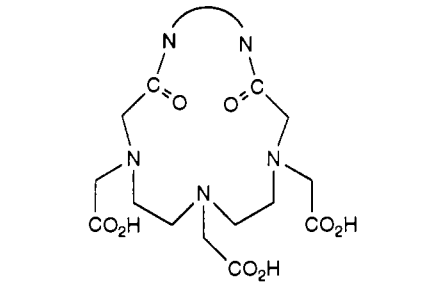
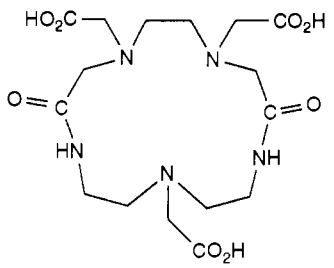


MACROCYCLE	N N
DTPA-EAM	HN NH
DTPA-PAM	HN NH
DTPA-BAM	HN NH
DTPA-cis$^{C=C}$BAM	HN NH
DTPA-PenAM	HN NH
DTPA-XAM	HN NH
DTPA-OAM	HN O O NH



EDTA-DAM

Figure 1. Structural formulas of DTPA and EDTA bis(amide) macrocycles.

DMSO (250-mL flask) and DTPA bis(anhydride) was added neat at once at ambient temperature. The reaction mixture was stirred for 24 h, concentrated under reduced pressure to an oil, diluted with water (40 mL), and adjusted to pH 10.0 using either 1 N HCl or 1 N NaOH. The solution was then applied to AG1-X8 (100–200 mesh, acetate form) resin in a 2.5 × 20 cm column, the column washed well with deionized water, and the product eluted with acetic acid to yield the title compound as a white solid after removal of acetic acid followed by lyophilization and recrystallization.

1,4,7-Tris(carboxymethyl)-9,14-dioxo-1,4,7,10,13-pentaazacyclopentadecane (DTPA-EAM). 1,2-Diaminoethane (0.336 g, 5.60 mmol) in DMSO/DBN (122 mL/3 mL) was condensed with DTPA bis(anhydride) (2.00 g, 5.60 mmol) with continuous stirring for 24 h. The crude product was applied to AG1-X8 resin (70-mL column bed) and after washing with water (100 mL) was eluted with 250 mL of acetic acid using a 0.5–1.0 N gradient. The product (0.71 g, 30%) was obtained as a white solid after recrystallization from ethanol/acetone (50 mL/25 mL). ^{1}H NMR (D_2O): δ 3.0 (s, 8 H), 3.14 (s, 4 H), 3.40 (s, 4 H), 3.45 (s, 4 H), 3.50 (s, 2 H). MS: m/e 418 $(M + H)^+$. Anal. Calcd (found) for $C_{16}H_{27}N_5O_8 \cdot 0.75H_2O$: C, 44.60 (44.62); H, 6.67 (6.36); N, 16.25 (16.11).

1,4,7-Tris(carboxymethyl)-9,15-dioxo-1,4,7,10,14-pentaazacyclohexadecane (DTPA-PAM). 1,3-Diaminopropane (0.415 g, 5.60 mmol) in DMSO/DBN (175 mL/3 mL) was condensed with DTPA bis(anhydride) (2.00 g, 5.60 mmol) with continuous stirring for 20 h. The crude product was applied to AG1-X8 resin (70-mL column bed) and, after washing with water (100 mL), eluted from the column with 300 mL of 0.5 N acetic acid. The product (0.428 g, 18%) was obtained as a white solid. ^{1}H NMR (D_2O): δ 1.58 (br t, J = 6 Hz, 2 H), 3.15 (m, 12 H), 3.43 (s, 2 H), 3.48 (s, 4 H), 3.67 (s, 4 H). MS: m/e 432 $(M + H)^+$. Anal. Calcd (found) for $C_{17}H_{29}O_8N_5 \cdot 1.00H_2O$: C, 45.43 (45.62); H, 6.95 (6.73); N, 15.58 (15.30).

1,4,7-Tris(carboxymethyl)-9,16-dioxo-1,4,7,10,15-pentaazacycloheptadecane (DTPA-BAM). 1,4-Diaminobutane (0.740 g, 8.40 mmol) in DMSO/DBN (96 mL/4 mL) was condensed with DTPA bis(anhydride) (3.00 g, 8.40 mmol). The crude product was applied to AG1-X8 (75-mL column bed) and after washing with water (200 mL) was eluted with 500 mL of 0.5 N acetic acid. The title compound (0.52 g, 14%) was obtained as a white solid after recrystallization from water/acetone (1 mL/5 mL). ^{1}H NMR (D_2O): δ 1.35 (s 4 H), 3.05 (s, 12 H), 3.50 (s, 4 H), 3.51 (s, 2 H), 3.55 (s, 4 H). MS: m/e 446 $(M + H)^+$.

1,4,7-Tris(carboxymethyl)-9,16-dioxo-1,4,7,10,15-pentaaza-*cis*-12-cycloheptadecene (DTPA-*cis*$^{C=C}$-BAM): (i) Preparation of 1,4-Bis(phthalimido)-*cis*-2-butene.[10] Potassium phthalimide (63 g, 0.34 mmol) was suspended in 200 mL of DMF and brought to 50 °C. 1,4-Dichloro-*cis*-2-butene (12.5 g, 0.10 mmol, 10.52 mL) was added dropwise over 3 h while the temperature was maintained between 50 and 55 °C. The mixture was heated at 60 °C for 5 h and stirred for a further 15 h at ambient temperature. The mixture was then poured into 400 mL of ice–water, stirred for 10 min, and filtered. The white filter cake was washed with 100 mL of 1% $NaHCO_3$ and 50 mL of H_2O and dried. Recrystallization from CH_3CH_2OH/CH_3CN yielded 27 g (78%) of the bis(phthalimido) compound as a white solid. ^{1}H NMR (D_2O): δ 4.53 (d, J = 4.5 Hz, 4 H), 5.66 (t, J = 5.8 Hz, 2 H), 7.67 (t, J = 2.4 Hz, 4 H), 7.80 (t, J = 2.5 Hz, 4 H).

(ii) Preparation of 1,4-Diamino-*cis*-2-butene Dihydrochloride.[10] 1,4-Bis(phthalimido)-*cis*-2-butene (20.0 g, 0.058 mol) was refluxed in ethanol (125 mL). A solution of hydrazine monohydrate (6.37 mL, 0.130 mol) in water (7 mL) was added over 0.5 h. The solution was then refluxed for an additional 3 h and stirred overnight at ambient temperature. The mixture was brought to pH 1 by the slow dropwise of 10 N HCl, stirred for 0.5 h, and allowed to sit overnight. Water (100 mL) was added, a chalky white solid filtered out, and the filtrate stripped to yield an orange-yellow solid. Recrystallization from methanol yielded 7.15 g (78%) of the diamine dihydrochloride salt. ^{1}H NMR (D_2O): δ 3.54 (d, J = 5.26 Hz, 4 H), 5.67 (t, J = 5.26 Hz, 2 H).

(iii) Preparation of DTPA-*cis*$^{C=C}$-BAM. 1,4-Diamino-*cis*-2-butene dihydrochloride (0.89 g, 5.59 mmol) in DMSO/DBN (96 mL/4.5 mL) was condensed with DTPA bis(anhydride) (2.0 g, 5.59 mmol). The crude product was applied to AG1-X8 (70-mL column bed) and after washing with water (750 mL) was eluted with 400 mL of 0.5 N acetic acid. The title compound (750 mg, 32%) was obtained as a white solid after recrystallization from water/acetone (10 mL/5 mL). ^{1}H NMR (D_2O): δ 2.97 (br t, J = 5.0 Hz, 4 H), 3.15 (t, J = 5.0 Hz, 4 H), 3.30 (s, 4 H), 3.35 (s, 2 H), 3.59 (s, 2 H), 3.70 (d, J = 6.0 Hz, 4 H), 5.47 (t, J = 6.0 Hz, 2 H). MS: 444 $(M + H)^+$. Anal. Calcd (found) for $C_{18}H_{29}N_5O_8 \cdot 1.00H_2O$: C, 46.85 (46.99); H, 6.77 (6.66), 15.18 (14.95).

1,4,7-Tris(carboxymethyl)-9,17-dioxo-1,4,7,10,16-pentaazacyclooctadecane (DTPA-PenAM). 1,5-Diaminopentane (1.143 g, 11.19 mmol) in DMSO/DBN (350 mL/6.9 mL) was condensed with DTPA bis(anhydride) (4.00 g, 11.19 mmol). The crude product was applied to AG1-X8 (75-mL column bed) and, after washing with water (250 mL), eluted with 1 L of 1 N acetic acid. The product (2.01 g, 39%) was obtained as a white solid after recrystallization from 2-propanol. ^{1}H NMR (D_2O): δ 1.13 (m, 2 H), 1.28 (m, 4 H), 3.05 (br t, J = 5 Hz, 8 H), 3.10 (br d, J = 5 Hz, 4 H), 3.40 (s, 4 H), 3.49 (s, 4 H), 3.55 (s, 2 H). MS: m/e 460 $(M + H)^+$. Anal. Calcd (found) for $C_{19}H_{33}N_5O_8 \cdot 1.10H_2O$: C, 47.61 (47.61); H, 7.40 (7.47); N, 14.61 (14.48).

6,9,12-Tris(carboxymethyl)-4,14-dioxo-3,6,9,12,15-pentaazabicyclo[15.2.2]heneicosa-1(19),18,20-triene (DTPA-XAM). *p*-Xylyenediamine (0.761 g, 5.59 mmol) in DMSO/DBN (97 mL/3.1 mL) was condensed with DTPA bis(anhydride) (2.00 g, 5.59 mmol). The crude product was applied to AG1-X8 (80-mL column bed) and after washing with water (1 L) eluted with 1.5 L of 0.5 N acetic acid. The product (0.3916 g, 14%) was obtained as a white solid after recrystallization from water. ^{1}H NMR (D_2O): δ 2.60 (t, J = 5 Hz, 4 H), 2.77 (t, J = 5 Hz, 4 H), 3.35 (m, 10 H), 4.14 (s, 4 H), 7.18 (s, 4 H). MS: m/e 494 $(M + H)^+$. Anal. Calcd (found) for $C_{22}H_{31}N_5O_8 \cdot 1.50H_2O$: C, 50.76 (50.94); H, 6.58 (6.26); N, 13.45 (13.05).

(10) Feigenbaum, A.; Lehn, J.-M. *Bull. Soc. Chim. Fr.* **1973**, 198.

1,4,7-Tris(carboxymethyl)-9,20-dioxo-13,16-dioxa-1,4,7,10,19-pentaazacyclobeneicosane (DTPA-OAM). 2,2′-(Ethylenedioxy)diethylamine (0.828 g, 5.59 mmol) in DMSO/DBN (250 mL/4.45 mL) was condensed with DTPA bis(anhydride) (2.00 g, 5.59 mmol). The crude product was applied to AG1-X8 (80 mL column bed) and after washing with water (1 L) eluted with 250 mL of 1 N acetic acid. The product (1.27 g, 45%) was obtained as an off-white solid. ^{1}H NMR (D_2O): δ 3.50 (m, 8 H), 3.25 (t, J = 6 Hz, 4 H), 3.41 (m, 14 H), 3.47 (s, 2 H), 3.51 (s, 4 H), 3.66 (s, 4 H). MS: m/e 506 $(M + H)^+$. Anal. Calcd (found) for $C_{20}H_{35}N_5O_{10}\cdot 2H_2O$: C, 44.36 (44.44); H, 7.26 (7.10); N, 12.93 (12.92).

4,10,13-Tris(carboxymethyl)-8,15-dioxo-1,4,7,10,13-pentaazacyclopentadecane (EDTA-DAM). (i) Preparation of 10,13-Bis(carboxymethyl)-8,15-dioxo-1,4,7,10,13-pentaazacyclopentadecane (EDTA-DET). To a solution of diethylenetriamine (1.61 g, 15.61 mmol) in DMSO/DBN (250 mL/5.8 mL) was added neat EDTA bis(anhydride) (4.0 g, 15.61 mmol) all at once. The reaction mixture was stirred for 24 h, concentrated under reduced pressure to an oil, diluted with water (30 mL), and the pH adjusted to 9.5 using 1 N HCl. The solution was applied to AG1-X8 (100–200 mesh), acetate form (70 mL column bed) resin, the column washed with deionized water (1 L), and the product eluted with 0.5 N CH_3COOH to give 1.25 g (23%) as a white solid after recrystallization from water/acetone (10 mL/4 mL). ^{1}H NMR (D_2O): δ 3.45 (s, 4 H), 3.16 (m, 4 H), 3.35 (s, 4 H), 3.41 (m, 4 H), 3.62 (s, 4 H). MS: m/e 360 $(M + H)^+$. Anal. Calcd (found) for $C_{14}H_{25}N_5O_6\cdot 3.35H_2O$: C, 40.06 (40.38); H, 7.61 (7.59); N, 16.69 (16.47).

(ii) Preparation of EDTA-DAM. To a solution of EDTA-DET (300 mg, 0.84 mmol) and diisopropylethylamine (1.08 g, 8.35 mmol) in CH_3OH (125 ML) was added *tert*-butyl bromoacetate (0.25 mL, 1.67 mmol) neat at once. The reaction mixture was stirred for 24 h at ambient temperature, stripped to dryness, and trifluoroacetic acid/methylene chloride (15 mL/2 mL) added. The solution was stirred for 4 h at ambient temperature, stripped to dryness, and the acid treatment repeated four times (until ^{1}H NMR showed complete removal of the *tert*-butyl group). The residue was taken up in 15 mL of water, the pH adjusted with 1N NaOH, and the solution applied to AG1-X8 acetate resin (75 mL column bed) and the product eluted with 250 mL of 0.05 N CH_3COOH to yield 215 mg (61%) of the title compound as a white solid after recrystallization from water/acetone. ^{1}H NMR (D_2O/NaOD): δ 2.52 (s, 4 H), 2.60 (br s, 4 H), 3.01 (s, 4 H), 3.07 (s, 2 H), 3.08 (s, 8 H). MS: m/e 418 $(M + H)^+$. Anal. Calcd (found) for $C_{16}H_{27}N_5O_8\cdot 0.25H_2O$: C, 45.55 (45.52); H, 6.57 (6.65); N, 16.60 (16.56).

Potentiometric Measurements. The potentiometric titrations were performed with an automated titration system that has been previously described in detail.[11] The autotitrating system consists of a Fisher digital pH meter, a Ross combination pH electrode, and a Metrohm digital autoburette. The pH electrode was calibrated using the data obtained from titration of 8×10^{-3} M KOH solution with 0.1000 M HCl, from which pK_w was also estimated. All calibrations and titrations were carried out under a N_2 atmosphere at 25.0 °C ± 0.1, and at an ionic strength of 0.10 M KCl.

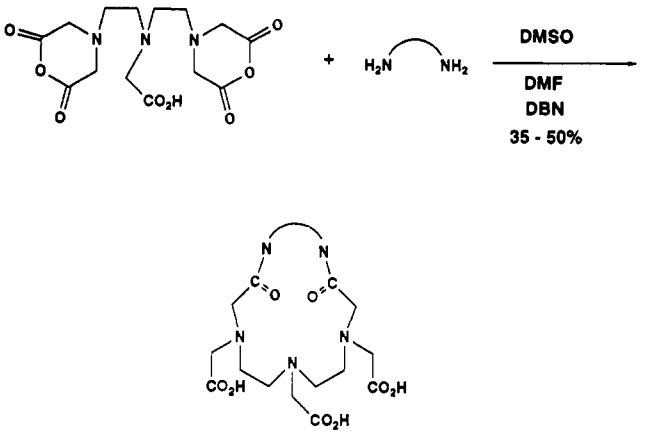
For the preparation of sample solutions, deionized water was degassed by boiling and subsequently saturated with N_2. The titration mixtures were prepared using 20–50 mM stock solutions of free ligand and standardized Zn^{2+} or Gd^{3+} stock solutions, and solid KCl. The pH of the titration mixture was then adjusted by the addition of a known volume of standard base. The concentration of metal ions and free ligands in the sample solution was maintained between 1.5×10^{-3} and 4×10^{-3} M during titrations and 0.1000 M HCl was used as a titrant to minimize ionic strength changes during the course of titrations. The pH of the initial solution was adjusted to 11 by adding NaOH solution, prior to the titrations.

The stability constants of all Gd^{3+} complexes, with the exception of GdDTPA-EAM, were determined from the titration data of a 1:1:1 molar ratio of Gd^{3+}, the respective ligand, and EDTA. The ratio of the two Gd^{3+} complexes at various pH allowed the estimation of the unknown stability constant from the known value of GdEDTA. Proton association constants and stability constants were calculated using a computer program described previously.[5]

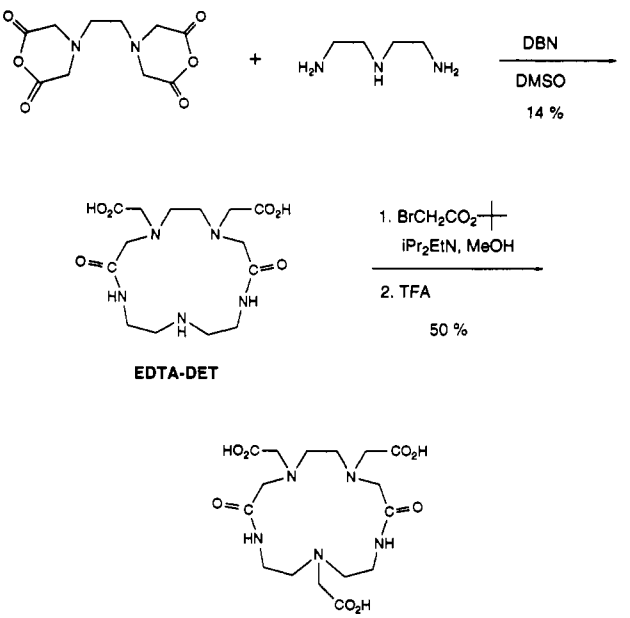
Results and Discussion

Ligand Synthesis. New DTPA-based bis(amide) macrocycles have been prepared through the cyclization of DTPA bis-(anhydride) and a diamine producing compounds containing ionizable functionalities (Scheme I). Neither high dilution nor simultaneous addition methods were required to achieve the moderate yields obtained, demonstrating the ease of synthesis. The preparation of the EDTA-based bis(amide) macrocycle was accomplished through the cyclization of EDTA bis(anhydride) and diethylenetriamine followed by alkylation (Scheme II). The yield from this cyclization reaction is lower than that for the DTPA series due to the reactivity associated with the unprotected secondary amine of diethylenetriamine.

Scheme I. Synthesis of DTPA-Based Bis(amide) Macrocycles



Scheme II. Synthesis of the EDTA-Based Bis(amide) Macrocycle



DTPA Bis(amide) Macrocycles. Table I lists the protonation constants of DTPA bis(amide) macrocycles and their stability constants with Gd^{3+} and Zn^{2+}. Three protonation constants for all DTPA bis(amide) macrocycles were observed in the pH range 2–10 and are associated with the amine groups. Stability constants of ZnDTPA bis(amide) macrocycles, and that of Gd(DTPA-EAM) were determined through competition with protons. For Gd^{3+} complexes which are dissociated less than 50% at pH 2, competitive titration techniques with EDTA were used to determine their stability constants. During the competitive titrations, the Gd^{3+} ion initially bound to EDTA at high pH migrated to the DTPA bis(amide) macrocycle at lower pH. The rate of metal transfer from EDTA to the macrocycle was sufficiently fast that equilibrium was established within 60 s after each addition of acid. A half life for the proton-assisted

(11) Rocklage, S. M.; Sheffer, S. D.; Cacheris, W. P.; Quay, S. C.; Hahn, E. F.; Raymond, K. N. *Inorg. Chem.* **1988**, *27*, 3530–3534.

Table I. Protonation Constants and Metal Chelate Stability Constants (25 °C, μ = 0.10 M (KCl))

equilibrium	DTPA-BMA[a]	DTPA-EAM	DTPA-PAM	DTPA-BAM	DTPA-*cis*$^{C-C}$-BAM	DTPA-PenAM	DTPA-XAM	DTPA-OAM	EDTA-DAM
[HL]/[L][H]	9.37	9.45 (0.03)	9.61 (0.04)	8.98 (0.01)	9.68 (0.04)	9.09 (0.03)	8.30 (0.02)	8.98 (0.07)	7.42 (0.02)
[H_2L]/[HL][H]	4.38	4.21 (0.02)	4.21 (0.01)	4.46 (0.01)	4.67 (0.01)	4.48 (0.02)	4.29 (0.02)	4.61 (0.13)	7.22 (0.01)
[H_3L]/[H_2L][H]	3.31	3.39 (0.03)	3.52 (0.08)	3.21 (0.06)	3.32 (0.04)	3.13 (0.03)	3.31 (0.10)	3.49 (0.04)	3.72 (0.02)
[H_4L]/[H_3L][H]	1.43	1.96 (0.21)	1.75 (0.21)	1.75 (0.19)	1.98 (0.11)	1.84 (0.23)	1.52 (0.26)	1.63 (0.21)	1.41
$\sum pK_a$	18.49	19.01	19.09	18.39	19.65	18.54	17.42	18.71	19.77
[GdL]/[Gd][L]	16.85	11.15 (0.03)	14.49 (0.32)	15.39 (0.14)	15.56 (0.04)	15.94 (0.19)	12.34 (0.16)	17.44 (0.57)	15.14 (0.57)
[GdHL]/[GdL][H]		2.87 (0.02)			2.28 (0.29)	2.06 (0.39)	3.58 (0.41)		
[ZnL]/[Zn][L]	12.04	12.08 (0.31)	11.74 (0.08)	11.56 (0.05)	12.19 (0.19)	11.46 (0.03)	10.65 (0.20)	12.19 (0.03)	9.03 (0.12)
[ZnHL]/[ZnL][H]	4.04	4.66 (0.04)	4.78 (0.23)	4.29 (0.04)	4.18 (0.02)	4.26 (0.02)	4.05 (0.05)	3.78 (0.04)	7.39 (0.03)
[ZnH_2L]/[ZnHL][H]		2.31 (0.07)	1.59 (0.04)		2.18 (0.13)		1.73 (0.19)		
log K(Gd/Zn)	4.81	–0.94	2.75	3.83	3.38	4.48	1.69	5.25	6.12

[a] Reference 5.

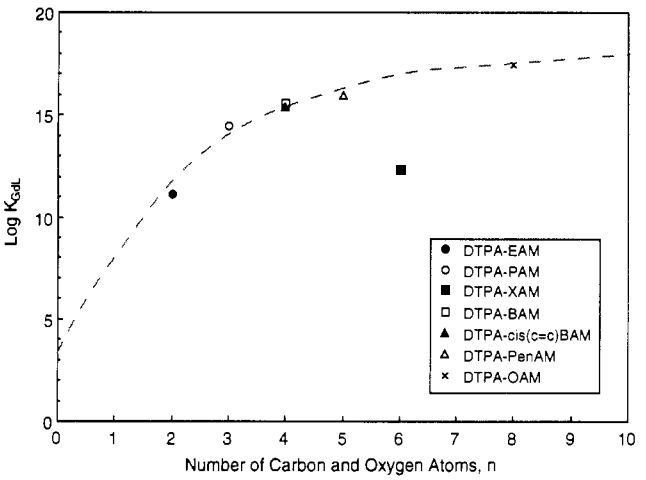


Figure 2. Plot of log K values of some Gd complexes of DTPA bis(amide) macrocycles as a function of number of carbon and oxygen atoms between two amide nitrogens.

dissociation of Gd(EDTA) to Gd^{3+} and free ligand was reported to be less than 5 s at 20 °C.[12]

Figure 2 plots the stability constants of GdDTPA bis(amide) complexes according to the number of carbon and oxygen atoms between two amide nitrogens. An increase in the stability constant was observed as the number of carbon and oxygen atoms increased. The X-ray crystal structures of Gd(DTPA-BEA)[13] and DyDTPA-BMA[14] illustrate that the amide carbonyl oxygens of the flexible, acyclic ligands are bound directly to the lanthanide metal ion. The observed increased stability of the gadolinium complexes of the bis(amide) macrocycles with larger pocket size probably reflects the participation of amide carbonyl oxygens in metal coordination. For favorable coordination of Gd^{3+}, the amide carbonyl oxygens should point toward the inside of the macrocycle pocket. To best achieve this conformation, at least eight atoms between the two amide nitrogens of the macrocycle are needed. Only Gd(DTPA-OAM) in this series shows a slightly higher stability constant than Gd(DTPA-BMA). As the macrocycle cavity size gradually increases, the rigidity of the macrocycle gradually decreases, causing the size selectivity and the degree of preorganization to gradually diminish. The stability constant of Gd(DTPA-XAM) was low, as compared to most other complexes in this study. Presumably, the aromatic ring may reside in such a manner as to decrease the cavity size of the macrocycle, consequently causing a weak interaction with Gd^{3+}. In addition to geometric considerations, the lower basicity of DTPA-XAM (pK_a = 17.42) relative to other DTPA bis(amide) macrocycles (pK_a = 18.54–19.65) may also contribute to the lower stability of Gd(DTPA-XAM).

On the other hand, the stability constants of the ZnDTPA bis(amide) macrocyclic complexes, with the exception of Zn(DTPA-XAM), do not change significantly throughout the series and are very similar to that of Zn(DTPA-BMA). Since the size of Zn^{2+} is much smaller than that of Gd^{3+}, the pocket sizes of DTPA bis(amide) macrocycles do not affect their stability constants. The low stability constant of Zn(DTPA-XAM) is primarily determined by the low basicity of the ligand (vide supra).

The logarithmic selectivity constants of DTPA-bis(amide) macrocycles, log K(Gd/Zn), are also listed in Table I. As predicted from the thermodynamic stability constants, only DTPA-OAM in the macrocycle series shows a slightly more favorable selectivity toward Gd^{3+} over Zn^{2+} than DTPA-BMA does. Since the basicity of DTPA-OAM ($\sum pK_a$ = 18.71) is very similar to that of DTPA-BMA ($\sum pK_a$ = 18.49), the contribution of an enthalpy term to the thermodynamic stability should be similar in both Gd^{3+} complexes. It seems that the formation of the DTPA-OAM macrocycle in this case may not necessarily have a preorganized conformation for the coordination of Gd^{3+} and therefore does not provide any thermodynamic complex formation advantage over that of the linear DTPA-BMA.

DTPA-EAM vs EDTA-DAM. A 15-membered EDTA macrocycle, EDTA-DAM, was prepared to compare the properties of this ligand with its isomeric 15-membered congener, DTPA-EAM. The basicity of EDTA-DAM (pK_a = 19.77) is slightly higher than that of DTPA-EAM (pK_a = 19.01). However, the stability constant of Gd(DTPA-EAM) was 4 log units lower than that of Gd(EDTA-DAM), indicating a geometrically more favorable conformation of the latter ligand for the coordination of Gd^{3+} than that of the former. An examination of CPK models suggests that the three carboxylic groups of (DTPA-EAM) are more crowded through coordination to Gd^{3+} than those in Gd(EDTA-DAM), presumably causing electrostatic repulsion among the negative carboxylic groups, resulting in a lower stability for Gd(DTPA-EAM). On the other hand, the stability constant of Zn(EDTA-DAM) was lower by 3 log units than that of Zn(DTPA-EAM). Thus, DTPA-EAM has a significantly higher selectivity for Zn^{2+} while EDTA-DAM has a significantly higher selectivity for Gd^{3+}. An observed protonation constant of Zn(EDTA-DAM) at 7.39 was very similar to the highest pK_a value of the free ligand, which suggests that the isolated amine nitrogen located between the two amide nitrogens does not bind to Zn^{2+}. Consequently, the enhanced stability of Gd(EDTA-DAM) and the reduced stability of Zn(EDTA-DAM) significantly improved the selectivity constant of this ligand, log K(Gd/Zn) = 6.12, over that of DTPA-EAM, log K(Gd/Zn) = –0.94. The selectivity of EDTA-DAM is by far the highest for Gd^{3+} over Zn^{2+} among all known polyamino polycarboxylates and is a good example of ligand structural discrimination for metal complex formation.

(12) Brücker, E.; Servas, P. *Inorg. Chim. Acta* **1970**, *4*, 632–636.
(13) Konings, M. S.; Dow, W. C.; Love, D. B.; Raymond, K. N.; Quay, S. C.; Rocklage, S. M. *Inorg. Chem.* **1990**, *29*, 1488–1491.
(14) Ehnebom, L.; Pederson, B. F. Manuscript submitted for publication.