Inorganic Chemistry

Article

pubs.acs.org/IC

Complexation of Metal Ions with TRAP (1,4,7-Triazacyclononane Phosphinic Acid) Ligands and 1,4,7-Triazacyclononane-1,4,7-triacetic Acid: Phosphinate-Containing Ligands as Unique Chelators for Trivalent Gallium

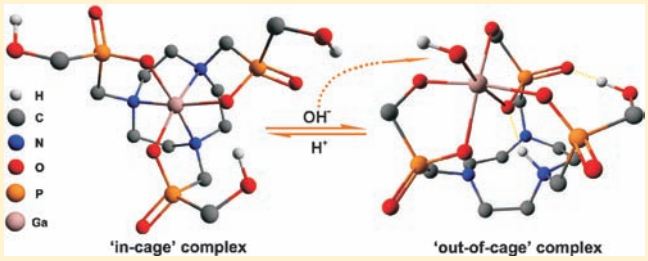
Jakub Šimeček,[†,‡] Martin Schulz,[§] Johannes Notni,[†,‡] Jan Plutnar,[†] Vojtěch Kubíček,[†] Jana Havlíčková,[†] and Petr Hermann*[,†]

[†]Department of Inorganic Chemistry, Univerzita Karlova (Charles University), Hlavova 2030, 12843 Prague 2, Czech Republic

[§]School of Chemical Sciences, Dublin City University, Glasnevin, Dublin 9, Ireland

Supporting Information

ABSTRACT: Three phosphinic acid 1,4,7-triazacyclononane (TACN) derivatives bearing methylphosphinic (TRAP-H), methyl(phenyl)phosphinic (TRAP-Ph), or methyl-(hydroxymethyl)phosphinic acid (TRAP-OH) pendant arms were investigated as members of a new family of efficient Ga^{3+} chelators, TRAP ligands (triazacyclononane phosphinic acids). Stepwise protonation constants of ligands and stability constants of their complexes with Ga^{3+}, selected divalent metal, and Ln^{3+} ions were determined by potentiometry. For comparison, equilibrium data for the metal ion−NOTA (1,4,7-triazacyclononane-1,4,7-triacetic acid) systems were redetermined. These ligands exhibit high thermodynamic selectivity for Ga^{3+} over the other metal ions (log K_{GaL} − log K_{ML} = 7−9) and a selective complexation of smaller Mg^{2+} over Ca^{2+}. Stabilities of the Ga^{3+} complexes are dependent on the basicity of the donor atoms: [Ga(NOTA)] (log K_{GaL} = 29.6) > [Ga(TRAP-OH)] (log K_{GaL} = 23.3) > [Ga(TRAP-H)] (log K_{GaL} = 21.9). The [Ga(TRAP-OH)] complex exhibits unusual reversible rearrangement of the "in-cage" N_3O_3 complex to the "out-of-cage" O_6 complex. The in-cage complex is present in acidic solutions, and at neutral pH, Ga^{3+} ion binds hydroxide anion, induces deprotonation and coordination of the *P*-hydroxymethyl group(s), and moves out of the macrocyclic cavity; the hypothesis is supported by a combination of results from potentiometry, multinuclear nuclear magnetic resonance spectrometry, and density functional theory calculations. Isomerism of the phosphinate Ga^{3+} complexes caused by a combination of the chelate ring conformation, the helicity of coordinated pendant arms, and the chirality of the coordinated phosphinate groups was observed. All Ga^{3+} complexes are kinetically inert in both acidic and alkaline solutions. Complex formation studies in acidic solutions indicate that Ga^{3+} complexes of the phosphinate ligands are formed quickly (minutes) and quantitatively even at pH <2. Compared to common Ga^{3+} chelators (e.g., 1,4,7,10-tetraazacyclododecane-1,4,7,10-tetraacetic acid (DOTA) derivatives), these novel ligands show fast complexation of Ga^{3+} over a broad pH range. The discussed TRAP ligands are suitable alternatives for the development of ^{68}Ga radiopharmaceuticals.



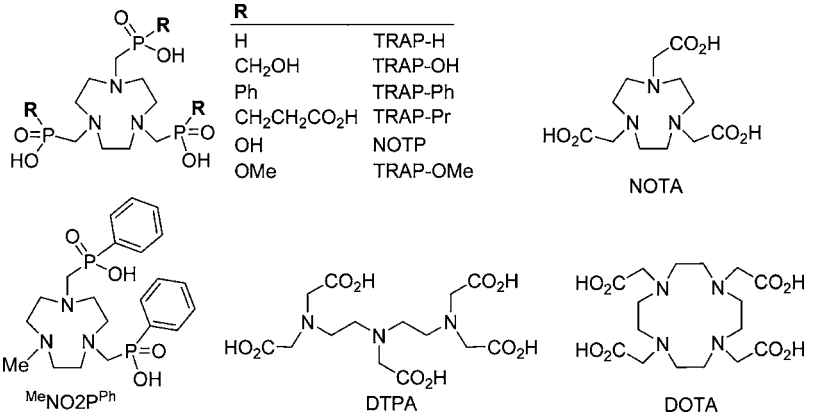
INTRODUCTION

Positron emission tomography (PET) is a noninvasive highly sensitive imaging modality providing a unique window for quantifying physiological functions and biochemical processes in living organisms. PET has become widely used over the past decade mainly in oncology and cardiology. The majority of PET examinations are conducted using [^{18}F]-fluorodeoxyglucose ([^{18}F]FDG), an agent for imaging of enhanced metabolic activity. Recent trends tend to combine PET with other imaging modalities as PET/CT[1] or PET/MRI.[2] Chemistry needed for progress in probe design should be developed to reach the full potential of the emerging scanners. In addition, new PET isotopes (mostly those of metallic elements) have been suggested for utilization in human medicine. Among the β^+-emitting radiometals, generator-produced[3] isotope ^{68}Ga [89% β^+; $\tau_{1/2}$ = 67.7 min; $E_{av}(\beta^+)$ = 740 keV] is the most promising, and its production, chemistry, and clinical use have been reviewed recently.[4−7] The metal radioisotopes cannot be used directly but must be bound by suitable ligands into thermodynamically stable and kinetically inert complexes. Also, other parameters, such as fast complexation kinetics, a pH suitable for quantitative complex formation, the solubility of the ligand and the complex, hydrophilicity and lipophilicity, an ability to attach the ligand and/or complex to a biomolecule, etc., have to be considered in designing radiometal-binding ligands.

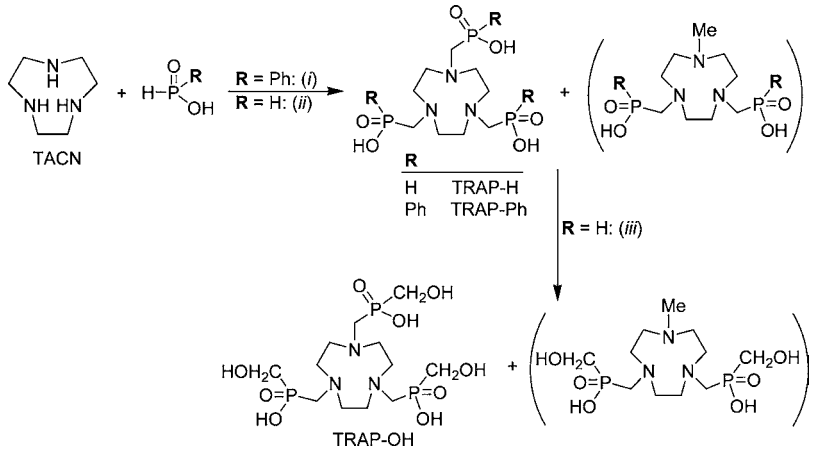
Received: September 27, 2011
Published: December 20, 2011

   dx.doi.org/10.1021/ic202103v | *Inorg. Chem.* 2012, 51, 577−590

Chart 1. Ligands Discussed Herein



Scheme 1. Synthesis of TRAP-R Ligands[a]



[a]Conditions: (i) $(CH_2O)_n$, 6 M aqueous HCl, 90 °C, 24 h; (ii) $(CH_2O)_n$, H_2O, room temperature, 24 h; (iii) $(CH_2O)_n$, 6 M aqueous HCl, reflux, 24 h.

Generally, hydrated Ga^{3+} ion, $[Ga(H_2O)_6]^{3+}$, is stable only in highly acidic solutions. Between pH 3.5 and 7, an insoluble precipitate of colloidal $Ga(OH)_3$ is formed. At pH ~7.5 and above, the hydroxide solubilizes due to formation of $[Ga(OH)_4]^-$ anion. The Ga^{3+} ion is classified as a hard Lewis acid and prefers octahedral coordination geometry. It forms the most stable complexes with ligands containing groups with hard Lewis donor atoms such as oxygen or nitrogen, e.g., carboxylates, phenols, phosphonates, phosphinates, hydroxamates, and amines.

Among applicable chelators, those derived from macrocyclic ligands 1,4,7,10-tetraazacyclododecane-1,4,7,10-tetraacetic acid derivatives and 1,4,7-triazacyclononane-1,4,7-triacetic acid (DOTA and NOTA, respectively) (Chart 1) are preferred because of the increased thermodynamic stability as well as kinetic inertness of their gallium(III) complexes[8−10] compared with their open-chain analogues as DTPA derivatives.[11] Several DOTA-based ligands,[12] mainly in combination with small peptides, have shown their clinical potential in nuclear medicine. Among the ^{68}Ga PET tracers, ^{68}Ga-labeled somatostatin analogues (e.g., the golden standard ^{68}Ga-[DOTA^0,D-Phe^1,Tyr^3]-octreotide, ^{68}Ga-DOTATOC) are extensively studied because of their superior results in neuroendocrine tumor imaging.[13−15] Despite an extensive use of DOTA-like ligands for complexation of trivalent lanthanides, they cannot be considered as ideally suited for coordination of Ga^{3+} ion; it should be mentioned that Ga^{3+} complexes of the DOTA conjugates are the most commonly used because some bifunctional DOTA-like ligands are commercially available and their chemistry is well-established. The DOTA cavity geometry on one hand and the preference of Ga^{3+} ion for the regular octahedral coordination on the other hand result in bad fitting of the metal ion into the ligand cavity.[5,16,17] In contrast, NOTA-like ligands present a cavity that is almost ideal for small octahedral metal ions such as Ga^{3+};[16,18−20] thus, a number of NOTA-like ligands have been synthesized for gallium(III) complexation.[20−25]

It has been shown that complexation properties of 1,4,7-triazacyclononane derivatives as well as other macrocyclic chelators with respect to metal ions can be altered by replacement of carboxylate group(s) with phosphinates.[26] Early examples of such ligands showed good fitting of Ga^{3+} ion into the ligand cage.[27] A phosphinic acid derivative of TACN tailored for very efficient ^{68}Ga complexation is TRAP-Pr (Chart 1).[28] Using TRAP-Pr, radiolabeling could be performed even at room temperature and at pH <1 with a radiochemical yield of >90% within just a few minutes.

However, the reasons for the almost ideal properties of TRAP-Pr in complexation of carrier-free ^{68}Ga are not fully understood. Thus, we decided to study a set of macrocyclic phosphinate-containing ligands and to reinvestigate the complexation properties of NOTA, as data for the latter ligand

dx.doi.org/10.1021/ic202103v | Inorg. Chem. 2012, 51, 577−590

Table 1. Stepwise Protonation Constants of the Title Ligands and Comparison with the Values for Other TACN-Based Ligands

constant	TRAP-H[a]	TRAP-Ph[a]	TRAP-OH[a]	TRAP-Pr[28]	TRAP-OMe[41]	NOTP[42]	NOTA
log K_1	**10.48**,[a] 10.16[b]	**12.08**	**11.47**	11.48	11.8	12.1	11.98,[c] 13.0,[d] 13.17[e]
log K_2	**3.28**,[a] 3.13[b]	**3.24**	**3.85**	5.44	3.65	9.4	5.65,[c] 5.6,[d] 5.74[e]
log K_3	1.11[b]	**1.44**	**1.30**	4.84	1.4	7.5	3.18,[c] 2.5,[d] 3.22[e]
log K_4				4.23		5.9	1.9,[d] 1.96[e]
log K_5				3.45		2.9	
log K_6				1.66			

[a]From this work (bold), at 25 °C, I = 0.1 M $(NMe_4)Cl$. [b]From ref 35 (0.1 M KNO_3). [c]From ref 8 (0.1 M KCl). [d]From ref 40. [e]From ref 43 [0.1 M $(NMe_4)Cl$]; this set of constants was used for the metal ion stability constant determinations.

Table 2. Complex Stability and Stepwise Protonation Constants for Gallium(III) Complexes of the Studied Ligands and Some Others[a]

	log K_{LGa} or log K_A					
equilibrium[b]	TRAP-H	TRAP-OH	TRAP-Pr[28]	NOTA		DOTA[10]
L + Ga ⇄ LGa	21.91	23.3	26.24	29.60	31.0[8]	26.05
LGa + H^+ ⇄ HLGa		1.6	0.7[d]	0.9		1.57[e]
$H_{-1}LGa + H^+ ⇄ LGa + H_2O$	7.97	6.96[c]	9.8	9.83	9.70[8]	
$H_{-2}LGa + H^+ ⇄ H_{-1}LGa + H_2O$		9.0[c]				

[a]At 25 °C and I = 0.1 $(NMe_4)Cl$; for ligand structures, see Chart 1. [b]Charges of ligand and complex species have been omitted for the sake of simplicity. [c]For specification of the protonation sites and for structures of the corresponding deprotonated species, see the text. [d]Value corresponding to the protonation of the coordinated phosphinate groups and, so, to the $H_4LGa ⇄ H_3LGa + H^+$ equilibrium.[28] [e]Value corresponding to the protonation of the coordinated acetates and, so, to the $H_3LGa ⇄ H_2LGa + H^+$ equilibrium.[10]

are not fully consistent in the literature.[8,29,30] These ligands bearing acetate or phosphinic acid moieties with different substituents on the phosphorus atoms are used to study the influence of the TACN pendant arms on coordination behavior. TRAP-Pr and this series represent differences in coordinating groups [CO_2H vs $P(R)O_2H$], hydrophilicity and lipophilicity, and the ability to form weak complexes employing donor atoms only from the pendant arms (CH_2OH and $CH_2CH_2CO_2H$ phosphorus atoms substituents).

RESULTS AND DISCUSSION

Throughout this Article, the abbreviations, e.g., TRAP-H, will be used regardless of ligand protonation state, except in cases in which the distinction is necessary for comprehension or used in formulae of distinct complex species; then, the abbreviations will be used according to IUPAC nomenclature, e.g., H_3trap-H.

Ligand Synthesis. The title TRAP ligands were synthesized via Mannich-type Moedritzer–Irani reaction (Scheme 1).[31] However, the syntheses were complicated by problems commonly connected with Mannich-type reaction of organophosphorus compounds in aqueous media. The main side reaction is formation of *N*-methylated species;[32] it led to problematic purification of the reaction mixtures and decreased yields of the isolated products. The presence of the *N*-methylated compounds was proven by isolation of the corresponding byproduct in the case of TRAP-Ph synthesis. The reductive methylation of a nitrogen atom(s) can be suppressed by conducting the reaction at the high overall concentration of the reactants and lowering the reaction temperature with the optimal acidity of the solvent.[32,33] The synthesis of TRAP-Ph was complicated by low reactivity of phenylphosphinic acid requiring a high acidity of the reaction medium (6 M HCl) and a high temperature (90 °C).[34] As the reaction conditions support N-methylation, the spectroscopic reaction yield of TRAP-Ph never exceeded 70% [^{31}P nuclear magnetic resonance (NMR) of the reaction mixture] and was further decreased during purification.

To prepare TRAP-H, the reaction could be conducted even at 20 °C with hypophosphorus acid as the only acidifying agent. Under these mild conditions, the reaction resulted in a high spectroscopic yield (90%; ^{31}P NMR of the reaction mixture). However, in this case, small amounts of other common byproducts, *P*-hydroxymethylated compounds, were present. Further lowering the temperature led only to a prolonged reaction time with no further suppression of the methylation. Unfortunately, because of a very inefficient separation of these byproducts, the isolated yield was significantly lowered (33%). Regardless, TRAP-H was prepared with a slightly higher yield compared to that with the procedure described previously.[35]

The preparation of TRAP-OH proceeded in two steps. The first one was identical with the synthesis of TRAP-H. In the second step, nonpurified TRAP-H was converted directly to TRAP-OH by the reaction with an excess of newly added paraformaldehyde. Upon complete *P*-hydroxymethylation, the purification from any *N*-methylated species was much more efficient and TRAP-OH was then obtained in 70% yield.

Equilibrium Studies. Stepwise protonation constants (Table 1; the full set of experimental data is presented in Table S1.1 of the Supporting Information) were determined by potentiometry in the presence of tetramethylammonium cation to prevent the undesirable formation of the alkali metal ion complexes that occurs for various polyaminopolycarboxylates and similar ligands. In the case of NOTA, its weak interaction with alkali metal ions has been confirmed in solution by NMR measurements.[36] The first protonation constant can be assigned to protonation of a ring nitrogen atom and is the most important for the overall basicity of the ligands. As expected, the phosphinate ligands exhibit lower log K_1 values than the carboxylate (NOTA) or phosphonate (NOTP) analogues. For aminoalkylphosphinic acids,[26] the value depends on the electronic properties of the phosphorus-bound substituents. The hydrogen atom can be considered as the most electron-withdrawing substituent in this series, leading to the least basic nitrogen atoms; surprisingly, the electron-withdrawing phenyl

dx.doi.org/10.1021/ic202103v | Inorg. Chem. 2012, 51, 577–590

Table 3. Stability Constants (log K_{ML}) of Selected Metal Ions with the Title Ligands and Chosen NOTA Analogues[a]

metal ion	TRAP-H	TRAP-OH	TRAP-Ph	TRAP-Pr[28]	NOTA	
Ga^{3+}	21.91	23.3	*c*	26.24	29.63	31.0[d]
Mg^{2+}	5.32[b]	6.59	5.38	7.84	10.97	9.69[e]
Ca^{2+}	4.29[b]	4.87	3.77	6.04	10.32	8.92[e]
Cu^{2+}	13.43[b]	15.53	15.18	16.85	21.99	21.63[e]
Zn^{2+}	13.04[b]	16.12	*c*	16.88	21.58	
La^{3+}	7.42	8.56	*c*	11.26		13.5[f]
Gd^{3+}	8.75	10.10	*c*	13.46		14.4[f]
Y^{3+}	8.69	9.96	*c*			

[a]A full set of the determined stability constants is given in the Supporting Information; for ligand structures, see Chart 1. [b]From ref 35. [c]Not determined because of the precipitation of the complex. [d]From ref 8. [e]From ref 29. [f]From ref 47.

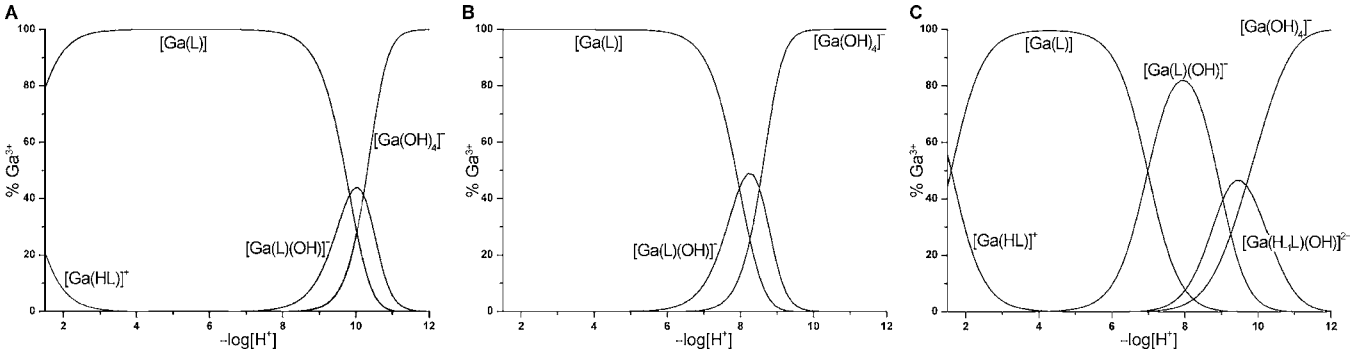


Figure 1. Distribution diagrams for Ga^{3+}–NOTA (A), Ga^{3+}–TRAP-H (B), and Ga^{3+}–TRAP-OH (C) systems ($c_{Ga} = c_L = 0.004$ M).

substituent exhibits the highest basicity. This effect can be caused by a shielding of the last *N*-bound proton by hydrophobic benzene rings from an interaction with surrounding water; a similar order of constant values has been observed for phosphinic acid derivatives of other polyaza macrocycles.[34,37,38] In general, the ligand basicity trends are similar to those observed for other amino acids: phosphonates > carboxylates > phosphinates ~ phosphonic monoesters.[26]

The log K_2 value could be assigned to the protonation of the second ring nitrogen atom. Next, protonation (corresponding to K_3) should take place on acetate or phosphinate/phosphonate pendant arms;[39,42] the next constant, K_4, was determined only for NOTA (acetate arm protonation).[40] The three remaining possible protonation constants for the phosphinate ligands could not be determined as the protonations occur only in strongly acidic media; this has been directly proven by NMR titration of phosphinic acid derivatives of 1-oxa-4,7-diazacyclononane.[34] This observation agrees with the general knowledge that phosphinic acids exhibit higher acidity than carboxylic ones; in the title ligands, even at pH ~1, some of the pendant arms are deprotonated and, thus, "pre-prepared" for the binding of metal cations.

The stability constants for complexes of NOTA and the title phosphinate ligands with gallium(III) as well as with some other cations were determined by potentiometric titrations (Tables 2 and 3; the full set of the experimental data is given in Table S1.2 of the Supporting Information). In the case of TRAP-Ph, determination of stability constants was possible only for Mg^{2+}, Ca^{2+}, and Cu^{2+} ions because of the insolubility of the other complexes.

In an acidic solution, some of the in-cage Ga^{3+} complexes are formed too slowly (see below) for a conventional potentiometric titration. Similarly in an alkaline solution, the rearrangement of the in-cage complexes and their full decomposition is also slow processes. Therefore, the titrations had to be performed using the out-of-cell technique with an equilibration time of 4 weeks at room temperature. As one can see from distribution diagrams (Figure 1), full complexation is reached even at pH 1.5, that is, in the beginning of the titrations. Therefore, the stability constants had to be determined from the equilibrium in the alkaline region where the macrocyclic ligands compete with hydroxide anions {$[Ga(OH)_4]^-$ formation}. Full hydroxide-induced dissociation occurs for the TRAP-H and NOTA complexes at slightly alkaline pH values, and the dissociation proceeds through formation of simple hydroxido species, $[Ga(L)(OH)]^-$; a similar chemical model was found for the Ga^{3+}–TRAP-Pr system.[28] A more complicated situation was observed in the case of the Ga^{3+}–TRAP-OH system where species with formal $[H_{-1}GaL]^-$ and $[H_{-2}GaL]^{2-}$ stoichiometries had to be included in the chemical model. The species start to form even in slightly acidic solutions (Figure 1) and could correspond either to the hydroxido complexes or to the complexes with deprotonated *P*-hydroxymethyl group(s). The site of deprotonation cannot be distinguished by potentiometry, because this method can observe only the amount of protons in the titrated solution. The presence of these species was surprising and led to a more thorough investigation as discussed below. The correctness of the potentiometric models was confirmed by NMR measurements; the determined concentrations of the species agreed with the abundances determined from the distribution diagram (Figure 1). The deprotonation of an alcohol group induced by its coordination to Ga^{3+} ion is not very common in aqueous solutions, and it was observed in only a few cases. The deprotonated alcoholate group is coordinated in the $[Ga(Hcitrate)_2]^{3-}$ anion,[44] and the only alcoholate groups are bound in the gallium(III) complex of an inositol derivative.[45] The data presented here extend this phenomenon to a new structural motif, the *P*-hydroxymethyl group.

dx.doi.org/10.1021/ic202103v | *Inorg. Chem.* 2012, 51, 577–590

In the monoprotonated $[Ga(HL)]^+$ species present in acidic solutions, Ga^{3+} cation is located inside the cavity of the phosphinate ligands and the proton is bound to an oxygen atom of a coordinated phosphinate group as suggested by NMR data (no free ligand was detected after the full equilibration at these pH values); the same phenomenon has been observed in the $[Ga(trap\text{-}Pr)]^{3-}$ complex[28] and in metal ion complexes of some polyaza macrocyclic polyphosphonate ligands[46] or for the $[Ga(dota)]^-$ complex.[10]

The stability constant of the [Ga(NOTA)] complex was redetermined in this work, but a comparison with the previous value is problematic as the original work[8] does not provide enough data on the equilibration time. The stability constants for gallium(III) complexes of the macrocyclic phosphinate ligands are lower than that for the [Ga(NOTA)] complex as a result of the lower basicity of the phosphinate ligands. It is clearly documented by the linear dependence of log K(GaL) on the sum of the protonation constants corresponding to ring nitrogen protonations, log K_1 + log K_2 (Figure S1.1 of the Supporting Information). Regardless, the thermodynamic stability of the complexes is high enough for possible in vivo utilizations. Comparison of log K_{GaL} values of the studied complexes with those of divalent metal ion complexes (Table 3) indicates an excellent thermodynamic selectivity of the studied ligands for Ga^{3+} ion. At least hexadentate macrocyclic ligands are required for the formation of thermodynamically stable complexes as stabilities of gallium(III) complexes with series of pentadentate 1-oxa-4,7-diazacyclononane derivatives with the same pendant arms are much lower [log K(GaL) = 8.9−14.9].[34]

To determine the selectivity of the ligands for gallium(III) complexation and to accumulate and/or redetermine data for more metal ions, we ran potentiometric titrations with the ligands and selected metal ions. The stability constants, K_{ML}, of complexes of the title ligands and similar TACN derivatives are listed in Table 3, and the full set of stability constants together with selected distribution diagrams and more comparisons of data for different systems are given in the Supporting Information (Tables S2 and S3 and Figures S1.2−S1.5). The stability constant values (Table 3) show high selectivity of all ligands for complexation of small metal ions, and this property is even more pronounced for the phosphinate ligands. The binding selectivities of the TRAP ligands for Ga^{3+} are 1−3 orders of magnitude greater than that of NOTA, and it might be a source of the efficient binding of carrier-free $^{68}Ga^{3+}$ (e.g., in the presence of the decay product, Zn^{2+}) observed previously.[28] This can be explained by a combination of the small internal ligand cavity, the difference in pendant arm donor atom hardness, and/or the coordination requirements of the metal cations. The data suggest that (i) log K_{ML} values are similar for copper(II) and zinc(II) complexes as in TACN-like ligands, one nitrogen atom has to be bound axially to Cu^{2+} ion leading to a lower thermodynamic stability of the complexes; (ii) complex stabilities are higher for the smaller and harder Mg^{2+} ion when compared with those of the Ca^{2+} ion and are more pronounced for hard phosphinate-containing ligands; (iii) there is a pronounced difference between La^{3+} and Gd^{3+} complex stability constants as the smaller Gd^{3+} ion fits the small ligand cavity better; and (iv) we noticed a high selectivity for the small and hard Ga^{3+} ion as mentioned above. In general, the values of stability constants depend on the overall basicity of the ligands and, therefore, are ordered as follows: phosphonate > carboxylate > phosphinate.

To obtain more data for trivalent metal ions relevant for biomedical applications and to test the correctness of the chemical model for the Ga^{3+}−TRAP-OH systems, we determined the stability constants of selected rare earth metal ions. The values of stability constants of phosphinate ligands are very low. However, potentiometric chemical models for these ions (requiring higher coordination numbers, mostly 8 or 9) and TRAP-OH (theoretically, a nonadentate ligand) were analogous to that for the Ga^{3+}−TRAP-OH system. Therefore, we can speculate that the hard Ln^{3+} or Y^{3+} ions can also induce a deprotonation of the *P*-hydroxymethyl group(s) with their simultaneous coordination to the central ions. Such a hypothesis is supported by facts that more deprotonated species ($H_{-1}LM$ and $H_{-2}ML$, or even $H_{-3}ML$ for Y^{3+}) had to be involved in the chemical model (Table S1.2 of the Supporting Information); their formation starts at neutral pH, and they have unusually high abundances and may contain coordinated hydroxide and/or alcoholate anion(s) (Figure S1.4 of the Supporting Information). As the analogous species were also suggested in the Ga^{3+}−TRAP-OH system (vide infra), the metal ion-induced deprotonation and simultaneous coordination of the hydroxymethyl group(s) seem to be a general case for this particular ligand. The phenomenon observed here is the first example of the formation of a chelate with an α-hydroxymethylphosphonic/phosphinic acid group involving alcoholate coordination.

DFT Calculations for Ga^{3+}−TRAP-OH Complexes. As one can see from Figure 1B, the [Ga(L)] species (H_3L = TRAP-OH) is dominant in acidic solutions; Ga^{3+} ion is bound in the in-cage complex having an octahedral N_3O_3 arrangement as found in other complexes of phosphinate[27,28] or phosphonate[48] TACN derivatives. With an increase in pH, the deprotonated species $[H_{-1}LGa]^-$ and $[H_{-2}LGa]^{2-}$ appear, followed by complex decomposition and formation of the free ligand and the $[Ga(OH)_4]^-$ anion. Because these species appear at higher pH values, the hydroxide anion seems to play a role in the processes. It can act as a Brönsted base deprotonating one or more *P*-hydroxymethyl moieties of the in-cage complex and/or coordinate to the Ga^{3+} ion that is still inside the ligand cage with replacement of a bound pendant arm(s). The other possibility is deprotonation of the *P*-hydroxymethyl moieties to furnish nucleophilic alcoholate anions that could coordinate Ga^{3+} ion; it induces the movement of the central ion out of the ligand cage with formation of an out-of-cage complex where Ga^{3+} ion is bound to the phosphinate side arms but not to the ring nitrogen atoms. In addition, this out-of-cage complex can bind water molecules or hydroxide anions if not all coordination sites are occupied by ligand oxygen atoms. These processes lead to an O_6 coordination arrangement. With these considerations in mind, a set of possible structures for the $[H_{-1}LGa]^-$ and $[H_{-2}LGa]^{2-}$ species was suggested. These structures cannot be distinguished by potentiometry, because this method can reveal only a number of protons in the titrated solution. However, from the comparison with the other complexes investigated here (see above), coordination of only hydroxide anion that would start at pH 5 seems to be rather improbable.

Once the equilibrium was reached, the unusual deprotonated species have high abundances. Unfortunately, any attempts to determine their structure by multinuclear NMR failed because of extremely broad and/or complicated spectra. NMR measurements of the purified [Ga(L)] complex prepared in an acidic solution showed the expected[27,28,48] spectra [Figure

dx.doi.org/10.1021/ic202103v | *Inorg. Chem.* 2012, 51, 577−590

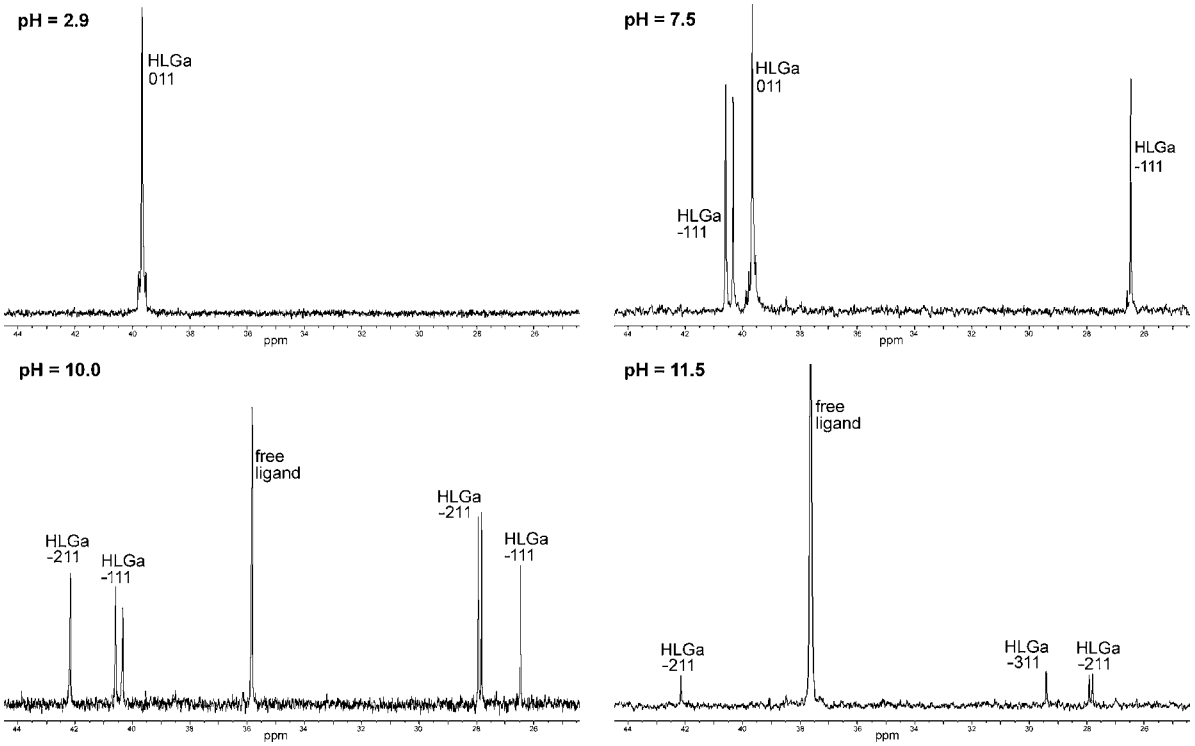
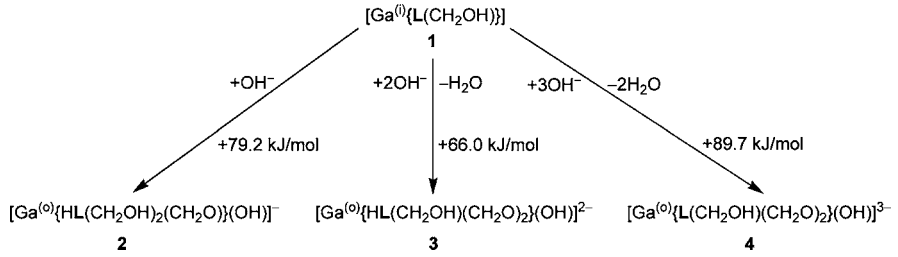


Figure 2. $^{31}P\{^{1}H\}$ NMR spectra of fully equilibrated solutions prepared by mixing Ga^{3+} ion and TRAP-OH ($c_{Ga} = c_L = 0.005$ M). The given pH values are those for the equilibrated solutions.

Scheme 2. The Most Suitable Species in the Ga^{3+}–TRAP-OH System As Suggested by the DFT Calculations[a]



[a]Relative free energies, ΔG_{calc}, are referenced to a $\Delta G_{calc}(\mathbf{1})$ of 0 kJ/mol.

S3.1 of the Supporting Information; δ_P 37.3, δ_{Ga} 135.2 ($\omega_{1/2}$ = 305 Hz)] and the corresponding ^{1}H NMR spectrum with the presence of a single diastereoisomer (see below) of the complex. It is consistent with the structure of other gallium(III) in-cage complexes with NOTA-like ligands.[16] However, among the NMR spectra recorded for different nuclei on the complex solutions at higher pH values, only the $^{31}P\{^{1}H\}$ NMR spectra were reasonably resolved to be interpretable (Figure 2). The ^{71}Ga NMR signals of any deprotonated species were not detected. This points to rather unsymmetrical structures of the $[H_{-1}LGa]^{-}$ and $[H_{-2}LGa]^{2-}$ species. These species were not exchanging with each other or with the free ligand on the NMR time scale.

Thus, DFT calculations were employed to suggest structures of the species that can be present in the Ga^{3+}–TRAP-OH system. Because of a larger number of degrees of freedom in these molecules, the energies presented below must be considered only as estimates, and some considerations were taken into account prior to the calculations. At pH >6, the main ligand species present in solution is the $(Htrap\text{-}OH)^{2-}$ anion (Figure S2.1 of the Supporting Information) in which the proton is bound to the ring nitrogen atoms; in the considered out-of-cage complex species, the ring nitrogen atoms may also bind a proton as the pK_a for such deprotonation in the free ligand is as high as 11.5 (Table 1). It is well-known that phosphorus acid oxygen atoms are able to form strong hydrogen bonds. The most probable coordination environment of Ga^{3+} ion in such complexes should be close to the octahedron. During the calculations, a modeling of the solvent influence was conducted with conditions as close as possible to the experimental conditions. Stoichiometries, binding modes, and relative energies of selected species considered during DFT calculations are shown in Scheme 2. More details about the calculations and a full set of the species considered in the calculations together with figures of their structures can be found in the Supporting Information (Figures S2.2–S2.5). In the following text, the protonated ligand sites will be distinguished according to the following examples (charges will be omitted for the sake of simplicity). For ligand monoprotonated on a ring nitrogen atom and having all *P*-hydroxymethyl groups protonated, the notation will be $\{HL(CH_2OH)_3\}$ (with a charge of −2). For a ligand with

dx.doi.org/10.1021/ic202103v | *Inorg. Chem.* 2012, 51, 577–590

deprotonated ring nitrogen atoms and with two deprotonated *P*-hydroxymethyl groups, the notation will be $\{L(CH_2OH)(CH_2O)_2\}$ (having a charge of −5). Gallium(III) in the in-cage complex will be labeled as $Ga^{(i)}$ and in the out-of-cage complex as $Ga^{(o)}$.

In-Cage Complexes. The neutral in-cage complex $[Ga^{(i)}\{L(CH_2OH)_3\}]$ (**1**) was taken as a reference with a relative Gibbs free energy (ΔG) of 0.0 kJ/mol (Figure 3). To test the correctness of our calculations, the calculated geometric parameters were compared with the experimental X-ray diffraction data {for the $[Ga(H_3trap\text{-}Pr)]$ complex},[28] and the agreement was found to be very good (Table S2.1 of the Supporting Information). Although hydrogen bonds should be generally considered with great care within the presented calculations, a strong hydrogen bond between the PCH_2OH group and a coordinated phosphinate oxygen atom of the adjacent pendant arm [O_C−H···O_P, 2.819 Å; ∠O_C−H···O_P, 162° (Figure 3)] can be noted. Analogous hydrogen bonds were found in structures of most species discussed in this section. The structure of **1** corresponds to the measured NMR spectra. Sequential deprotonation of the external *P*-hydroxymethyl group(s) leading to $[Ga^{(i)}\{L(CH_2OH)_2(CH_2O)\}]^-$ (**1a**) and $[Ga^{(i)}\{L(CH_2OH)(CH_2O)_2\}]^{2-}$ (**1b**) species (Figures S2.2 of the Supporting Information) was considered, as well. However, the pK_a for deprotonation of the *P*-hydroxymethyl group in aqueous solution calculated from the Gibbs free energy difference using equations from ref 49 equals 15.1. This value is too high to be accessible in an aqueous solution under common conditions; therefore, these structures can be excluded as those corresponding to the $[H_{-1}LGa]^-$ and $[H_{-2}LGa]^{2-}$ species.

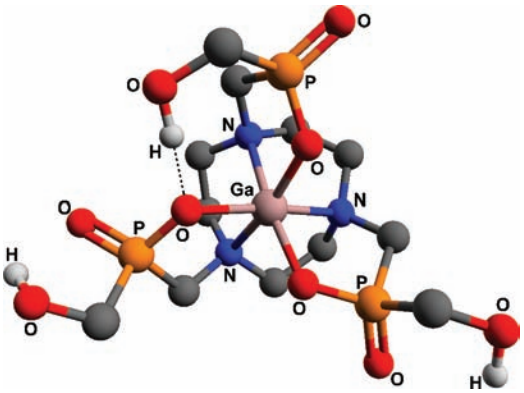


Figure 3. Calculated structure of the in-cage $[Ga^{(i)}\{L(CH_2OH)_3\}]$ (**1**) complex (H_3L = H_3trap-OH) taken as the lowest-energy complex. Hydrogen atoms except those in OH groups have been omitted for the sake of clarity, and the hydrogen bond is shown as a dashed line.

Out-of-Cage Complexes. As a first attempt, species having Ga^{3+} coordinated in an octahedral fashion by three phosphinate oxygen atoms and three oxygen atoms coming from the *P*-hydroxymethyl groups (1-, 2-, or 3-fold deprotonated) were tested. For the input structures, the Ga^{3+} ion was moved off the cage with a simultaneous transfer of a proton into the cage. In the output structure, the proton was found to be bound to a ring nitrogen atom with a rather strong hydrogen bond contact to a coordinated phosphinate oxygen atom (N−H···O, 2.6−2.9 Å; ∠N−H···O, 150−155°) and also exhibits a weak interaction with the other ring nitrogen atoms. However, the calculations for any simple isomerization or deprotonation did not lead to an octahedral Ga^{3+} coordination and always resulted in distorted pentacoordinate environments. All structures of the species [**A**−**C** (Figure S2.3 of the Supporting Information)] formed by sequential removal of the proton(s) from the reference in-cage $[Ga^{(i)}\{L(CH_2OH)_3\}]$ (**1**) complex have high relative energies (see the Supporting Information). The inaccessibility of the octahedral coordination sphere in these complexes can be explained by high sterical strain induced by the simultaneous presence of various Ga(−O−P−CH_2O−) chelate rings. In addition, the number and type of structurally clearly distinguishable phosphorus atoms in species **A**−**C** do not fit the peak pattern in experimentally observed $^{31}P\{^1H\}$ NMR spectra (Figure 2). Thus, the simple flip from the in-cage coordination sphere to an out-of-cage sphere with all three side arms coordinating in the same way can be ruled out.

This might be overcome by the introduction of a coligand, such as water or hydroxide anion; trivalent gallium has a high affinity for these ligands, and species with coordinated hydroxide anions had to be involved in the best chemical models for the gallium(III) systems with other NOTA analogues. As mentioned above, most of the structures were calculated with a protonated ring nitrogen atom (the proton is probably shared among all nitrogen atoms). The proton is bound inside the $(Htrap\text{-}OH)^{2-}$ anion in an analogous manner (Figure S2.1 of the Supporting Information), and a similar sharing of the last proton inside the ligand cavity was suggested for the 2-thioethyl TACN derivative.[50]

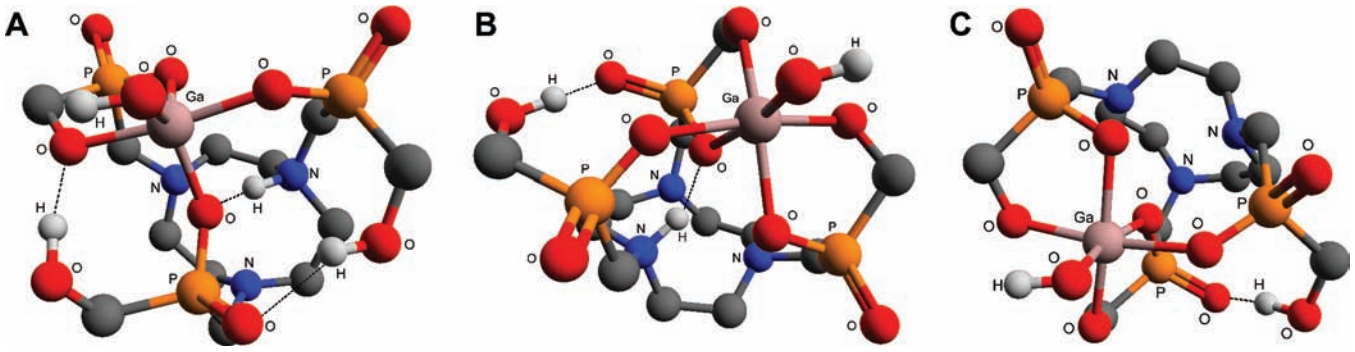


Figure 4. Structures of the out-of-cage species as suggested by calculations: (**A**) $[H_{-1}LGa]^-$ = $[Ga^{(o)}\{HL(CH_2OH)_2(CH_2O)\}(OH)]^-$ (**2**), (**B**) $[H_{-2}LGa]^{2-}$ = $[Ga^{(o)}\{HL(CH_2OH)(CH_2O)_2\}(OH)]^{2-}$ (**3**), and (**C**) $[H_{-3}LGa]^{3-}$ = $[Ga^{(o)}\{L(CH_2OH)(CH_2O)_2\}(OH)]^{3-}$ (**4**), where H_3L = TRAP-OH. Hydrogen atoms except those in OH or NH groups have been omitted for the sake of clarity, and the hydrogen bonds are shown as dashed lines.

dx.doi.org/10.1021/ic202103v | *Inorg. Chem.* 2012, 51, 577−590

Filling one coordination site with a water molecule furnished energetically disfavored (from 100 to 185 kJ/mol) and highly distorted pentacoordinated complexes [species **D** and **E** (Figures S2.4 of the Supporting Information)]. It was found that the water molecule is bound only loosely ($Ga-O_w$, 2.12−2.43 Å). As trivalent gallium has a high affinity for the hydroxide anion, the anion should be the most suitable coligand. In addition, deprotonated species found in other Ga^{3+}−ligand systems and experimentally studied in this Article contain a coordinated hydroxide anion. There are several possibilities for how to arrange various numbers of deprotonated *P*-hydroxymethyl moieties and the OH^- coligand. The structures are shown in Figure 4 and Figure S2.5 of the Supporting Information. Most of the calculated structures exhibit a distorted octahedral coordination arrangement expected for trivalent gallium and can be stabilized by intramolecular hydrogen bonds.

Among the species having a potentiometric $[H_{-1}LGa]^{1-}$ stoichiometry, the $[Ga^{(o)}\{HL(CH_2OH)_2(CH_2O)\}(OH)]^-$ (**2**) complex (Scheme 2 and Figure 4) is the most energetically favored species, although the coordination polyhedron of this species, a trigonal bipyramid, is not very common in gallium(III) complexes. Bond distances point to a strong coordination of the central ion ($Ga-O_P$, 1.96−1.99 Å; $Ga-O_C$, 2.01 Å; $Ga-O_H$, 1.88 Å). The in-cage proton is bound to a ring nitrogen atom and is kept inside the ligand cavity by a system of hydrogen bonds between nitrogen and oxygen atoms, as shown above and in Figure 4. Other species, **2a** and **2b**, were also considered (Figure S2.5 of the Supporting Information). Although they exhibit octahedral structures that are more common for the gallium(III) ion, their relative energies are very high in comparison (175 and 251 kJ/mol, respectively) with that of **2**.

The experimentally observed 2-fold deprotonated species, $[H_{-2}LGa]^{2-}$, should have one protonated ring nitrogen atom and only one *P*-hydroxymethyl group, as in the calculated $[Ga^{(o)}\{HL(CH_2OH)(CH_2O)_2\}(OH)]^{2-}$ (**3**) anion (Figure 4 and Scheme 2). This species has the lowest energy (66 kJ/mol) among all out-of-cage species and exhibits a distorted octahedral environment around the Ga^{3+} ion [$Ga-O_P$, 2.05−2.13 Å; $Ga-O_C$, 2.01 Å; $Ga-O_H$, 1.93 Å; ∠O−Ga−O, 80−98° (for adjacent oxygen atoms)]. The other considered species with such a stoichiometry was $[Ga^{(o)}\{HL(CH_2OH)_3\}(OH)_3]^{2-}$ (**3a**) (Figure S2.5 of the Supporting Information), but it exhibited a much higher relative energy (228 kJ/mol).

The fully deprotonated complex $[H_{-3}LGa]^{3-}$ might be the species present in alkaline solutions; however, $[Ga(OH)_4]^-$ anion formation should be more favorable under such conditions. Thus, in solution, this species is probably present (if ever) only with low abundance and could not be involved in the potentiometric chemical model. Regardless, the small singlet peak at 29.4 ppm was observed in the $^{31}P\{^1H\}$ NMR spectrum of the solution at high pH (Figure 2). Hence, this stoichiometry was also heeded during calculations. At high pH values, the nitrogen atom(s) should be deprotonated [ligand log K_1 = 11.47 (Table 1)] and the $[Ga^{(o)}\{L(CH_2OH)(CH_2O)_2\}(OH)]^{3-}$ (**4**) species (Figure 4 and Scheme 2), possessing a structure analogous to that of **3** but without the in-cage proton, is the most likely for the $[H_{-3}LGa]^{3-}$ stoichiometry. The coordination polyhedron around the Ga^{3+} ion [$Ga-O_P$, 1.98−2.14 Å; $Ga-O_C$, 2.03 Å; $Ga-O_H$, 1.94 Å; ∠O−Ga−O, 82−101° (for adjacent oxygen atoms)] is still distorted. Although the $[Ga^{(o)}\{HL(CH_2O)_3\}(OH)]^{3-}$ [**4a** (Figure S2.5 of the Supporting Information)] species has an only slightly higher energy (93 kJ/mol), there is a protonated ring nitrogen atom; thus, this structure was excluded. All three low-energy structures might be further stabilized by another intramolecular hydrogen bond between the phosphinate and the neighboring hydroxymethyl group ($O_C-H\cdots O_P$, 2.6−2.9 Å; $\angle O_C-H\cdots O_P$, 146−168°), as shown in Figure 4.

Structures **2**−**4** are in accordance with $^{31}P\{^1H\}$ NMR spectra (Figure 2). As stated above, these solution species are not fluxional and do not convert into each other on the NMR time scale, probably because of the rigidifying role of the proton inside the ligand cavity. Thus, the phosphorus atom environment in species **2** ($[H_{-1}LGa]^-$ stoichiometry) should exhibit two signals like reference in-cage complex **1** (the *P*-hydroxymethyl groups are protonated) and one signal with a much smaller δ_P (with a more electronegative P substituent, i.e., the deprotonated alcoholate). Analogously, species **3** ($[H_{-2}LGa]^{2-}$ stoichiometry) should exhibit one signal close to reference complex **1** and two peaks in the 22−28 ppm region. The small singlet at 29.4 ppm observed at high pH might be attributed to species **4** ($[H_{-3}LGa]^{3-}$ stoichiometry), where phosphorus atoms are averaged on the NMR time scale because of fast proton exchange between *P*-hydroxymethyl groups.

Information from the calculations can be summarized as follows. (i) Reference in-cage complex **1** has the most stable structure of all species investigated. (ii) As expected, a simple deprotonation of the *P*-hydroxymethyl moiety in in-cage complex **1** is difficult to achieve in an aqueous solution. (iii) A simple flip from the in-cage arrangement to the octahedral out-of-cage coordination sphere with three fully coordinated side arms only (chelate rings formed by the $^-OPCH_2O^-$ or $^-OPCH_2OH$ moieties only) will not occur because of the steric strain. (iv) The out-of-cage species possessing an O_6 coordination environment can be obtained only by addition of the hydroxide anion as a coligand. (v) Structures with reasonably low relative energies can comprise both penta- and hexacoordinated Ga^{3+} ion. (vi) The structures might be stabilized by intramolecular hydrogen bonds.

Isomerism of Gallium(III) Complexes in Solution. As mentioned above, the most stable in-cage gallium(III) complexes of the NOTA-like ligands exhibit distorted octahedral coordination arrangements. From another point of view, the Ga^{3+} ion is "sandwiched" between twisted trigonal O_3 and N_3 planes. Such an arrangement leads to chiral complexes in which the chirality is caused by a combination of the conformation of macrocycle-containing chelate rings ($\delta\delta\delta/\lambda\lambda\lambda$) and the helicity of the coordinated pendant arms (Δ/Λ). It leads to two diastereomeric pairs, $\Delta\delta\delta\delta/\Lambda\lambda\lambda\lambda$ and $\Lambda\delta\delta\delta/\Delta\lambda\lambda\lambda$. The phenomenon is fully analogous to the isomerism well-documented in lanthanide(III) complexes of DOTA-like ligands[51] and has been observed for NOTA complexes with different metal ions.[36,52] In the phosphinic acid NOTA analogues, the phosphorus atoms become chiral (*R*/*S*) after coordination to a central metal ion; again, the same chirality originating from the metal ion coordination of the phosphinate group is commonly observed in lanthanide(III) complexes of phosphinic acid analogues of DOTA.[51] Hence, combination of all the chiralities in complexes of TACN bearing three phosphinate pendant arms results in four possible diastereomeric pairs, $\Lambda\delta\delta\delta$-*RRR*/$\Delta\lambda\lambda\lambda$-*SSS*, $\Lambda\delta\delta\delta$-*RRS*/$\Delta\lambda\lambda\lambda$-*SSR*, $\Lambda\delta\delta\delta$-*RSS*/$\Delta\lambda\lambda\lambda$-*SRR*, and $\Lambda\delta\delta\delta$-*SSS*/$\Delta\lambda\lambda\lambda$-*RRR* (Figure S2.7 of the Supporting Information). In the solid state, only one of

dx.doi.org/10.1021/ic202103v | *Inorg. Chem.* 2012, 51, 577−590

the diastereomers, Λδδδ-*RRR*/Δλλλ-*SSS*, was observed for the [Ga(trap-Ph)] and [Ga(H_3trap-Pr)] complexes.[27,28] In solution, isomerism was investigated only for the $[Ga(trap\text{-}Pr)]^{3-}$ complex; the isomer present in solution was shown to be identical to that in the solid state.[28]

It is known that the [Ga(NOTA)] complex is present as a single diastereomer in the solid state but is fluxional in solution at room temperature as its ^{1}H NMR spectrum exhibits average signals.[18] On the other hand, phosphonate or phosphinate complexes are more rigid and the diastereomers of their complexes do not convert into each other.[27,28,42] As described above, the NMR spectrum of the in-cage [Ga(trap-OH)] complex and DFT calculations indicate that, in solution, the complex is present as a rigid Λδδδ-*RRR*/Δλλλ-*SSS* isomer, like the $[Ga(trap\text{-}Pr)]^{3-}$ complex.[28]

A different situation was observed for the [Ga(trap-H)] complex as unusual NMR spectra were obtained. Despite the common quadrupole broadening of ^{71}Ga NMR spectra, four very narrow NMR signals in a 1:3:4:2 intensity ratio (δ_{Ga} values of 132.00, 134.96, 137.80, and 140.45 ppm and $\omega_{1/2}$ values of 154, 200, 186, and 162 Hz, respectively) were observed (Figure 5). The signals probably correspond to the presence of all four diastereomers of the complex (Figure S2.7 of the Supporting Information). The doublet in the ^{31}P NMR spectrum centered at 22.0 ppm ($^1J_{PH}$ = 599 Hz) shows inconspicuous shouldering of the 20.1 ppm signal (Figure 5); the doublet is in agreement with the ^{1}H NMR spectrum that shows two overlapping H−P doublets centered at 7.20 and 7.25 ppm with $^1J_{PH}$ values of 607 and 561 Hz, respectively. Unfortunately, one-dimensional and two-dimensional ^{1}H and/or ^{13}C NMR spectra (or correlations with ^{31}P NMR spectra) of the isomeric mixture were too complicated or had signals that were overly broad to be useful for assignment of the structures of the individual isomers (Figure S3.2 of the Supporting Information).

The extraordinarily narrow ^{71}Ga NMR resonances (compared with those of complexes of other TACN-based ligands)[18,24,25,27,28,48] point to the high symmetry of the donor atom arrangements in the [Ga(trap-H)] diastereomers. The ratio of the four ^{71}Ga NMR signals is independent of the conditions used for preparation of the complex [different temperatures ranging from 25 to 80 °C, different rates of reactant mixing, the source of the Ga^{3+} cation, $Ga(NO_3)_3$ or $GaCl_3$, and dilution of the reacting solutions]. In addition, the ratio of ^{71}Ga signals remained constant under all conditions applied during the preparation of the complex, such as heating. Evidently, the isomers are not fluxional, and an energetic barrier for their mutual transformation has to be relatively high. This hypothesis was examined by DFT calculations.

The relative Gibbs energies were calculated for these four diastereoisomers and were found to be very close to each other (Table S2.4 of the Supporting Information). This supports the assumption, made above, that four signals in the ^{71}Ga NMR spectrum originate from four different diastereomers. The obtained relative energies of particular isomers are 0.00, −3.68, −7.25, and −6.25 kJ/mol for Λδδδ-*RRR*, Λδδδ-*RRS*, Λδδδ-*RSS*, and Λδδδ-*SSS*, respectively. However, the conversion from one diastereomer to another is accomplished by rotation of the N−CH_2−PO_2 moiety or inversion of the ethylene chain in the ethylenediamine chelate ring; activation barriers of the processes should be substantially high. We calculated the barrier height for the conversion from Λδδδ-*SSS* to Λδδδ-*RSS* ($\Delta G^{\#}$) to be 130 kJ/mol relative to the Λδδδ-*SSS* isomer.

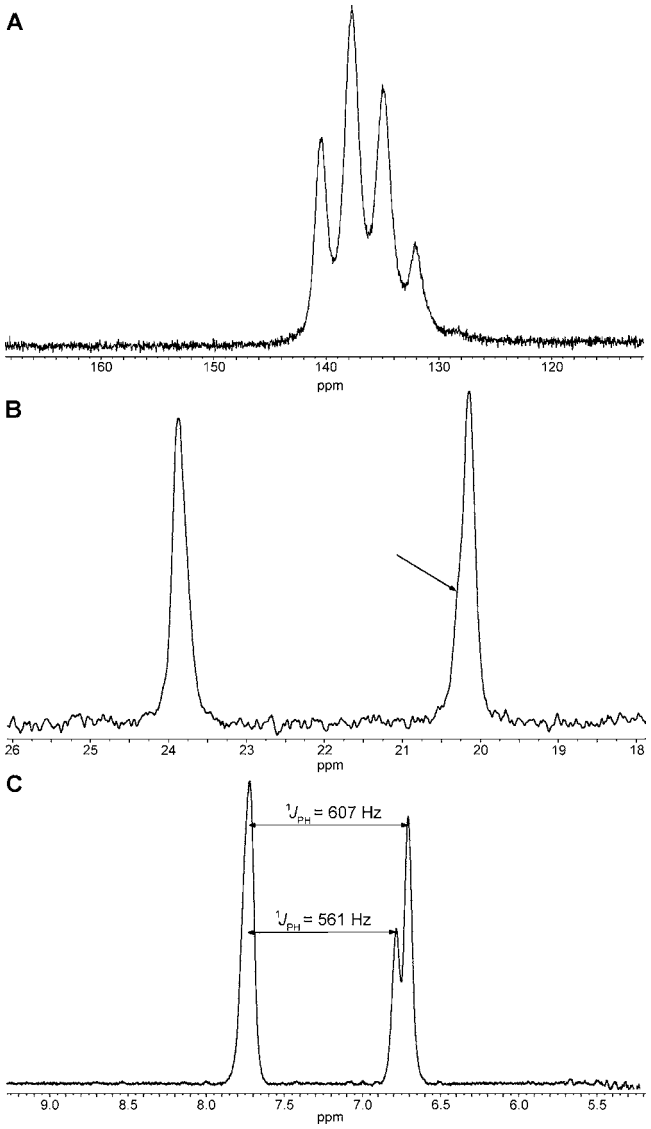


Figure 5. ^{71}Ga (A), ^{31}P (B), and a low-field part of ^{1}H (C) NMR spectra of the [Ga(trap-H)] complex (the shoulder in the ^{31}P NMR spectrum is marked with an arrow).

Interconversions among the other isomers should have similar energetic barriers.

The Ga^{3+}−TRAP-Ph system cannot be experimentally investigated in a similar manner because of the precipitation of a white solid (presumably the previously investigated complex, its Λδδδ-*RRR*/Δλλλ-*SSS* isomer[27]). However, the diluted supernatant solution taken before complete precipitation exhibits several ^{31}P{^{1}H} NMR signals in the region expected for the gallium(III) complex.

The differences in abundance of the complex diastereoisomers with varied substituents on phosphorus atoms show an interesting analogy with the lanthanide(III) complexes of the tetraphosphorus acid derivatives of DOTA.[38] In these complexes (with a square antiprismatic coordination geometry), only one major diastereoisomer was present in solution if the phosphorus atom substituents were alkyls such as Me, CH_2OH, Et, and Bn. For more electronegative phosphorus atom substituents such as H, Ph, and OR′, a full set of all possible diastereomers was observed in solution. The same phenomenon was observed here for gallium(III) complexes of

dx.doi.org/10.1021/ic202103v | *Inorg. Chem.* 2012, 51, 577−590

tris(phosphinic acid) derivatives, TRAP-R. The ligands with alkyl substituents ($CH_2CH_2CO_2H$ in TRAP-Pr or CH_2OH in TRAP-OH) give rise to only one diastereoisomer of the complexes, while those with more electron-withdrawing substituents (H in TRAP-H or probably also Ph in TRAP-Ph) form a mixture of diastereoisomers during complexation. However, such a hypothesis needs more data to be confirmed.

Formation and Decomposition of the Gallium(III) Complexes. To study the formation and decomplexation of the gallium(III) complexes, we used ^{31}P and ^{71}Ga NMR spectroscopy. Formation of the complexes at room temperature was monitored by ^{71}Ga NMR spectroscopy and quantified by comparison to the intensity of the $[Ga(OH)_4]^-$ signal as a standard; monitoring of the reaction progress by ^{71}Ga NMR spectroscopy has been recently used for the quantitative kinetic evaluations of complex formation and decomplexation in the Ga^{3+}–DOTA[10] and Ga^{3+}–NOTA–citrate[53] systems. The ^{31}P NMR spectra were used to show more detailed changes in the abundance of the free ligands, intermediates, and final gallium(III) complexes.

The rate of formation of the complex is strongly influenced by the substituent on the phosphorus atom, showing the important role of the substituents in the design of the TRAP ligands for trivalent gallium complexation (Table 4). At pH 2.8, all examined ligands except TRAP-H showed very fast complexation; gallium(III) was quantitatively bound in less than 5 min, which is the shortest possible time needed between preparation of a sample and measurement of the NMR spectra. At pH 1, TRAP-OH and TRAP-Pr showed much faster complexation than NOTA; however, the Ga^{3+}–TRAP-H complex needed approximately 15 days to be fully formed. At pH 0, all the phosphinate ligands were still able to form complexes with Ga^{3+} ion, albeit slowly (weeks); meanwhile, NOTA showed no signs of complex formation even after 40 days. Such measurements were not possible for TRAP-Ph; however, the fast precipitation even in acidic solutions points to rather fast complexation. The results emphasize the importance of careful pH control during the complexation with NOTA as described previously.[4] The higher acidity of the phosphinic acid groups and the lower basicity of the ring nitrogen atoms of the phosphinic acid TACN derivatives are responsible for complexation even at pH 0, where the more basic NOTA exhibits no gallium(III) binding. The results also show that ligands with electron rich side-chain substituents are able to interact with Ga^{3+} ion (TRAP-OH, TRAP-Pr, and probably TRAP-Ph) and thereby significantly accelerate the formation of the final in-cage complexes. The comparison of TRAP-OH and TRAP-H complexation can serve as an example; the Ga^{3+} ion is bound by the ligand with P–CH_2OH groups much faster than by the ligand with P–H groups, although both ligands do not have dramatically different basicities and the primary donor atoms are the same. Weakly interacting pendant arms move the metal ion closer to the macrocyclic cavity as seen Figure S3.3 of the Supporting Information. The deprotonated phosphinate group accelerates the transfer of the metal ion into the ligand cavity and helps to simultaneously remove nitrogen-bound protons from the cavity. The data for complexation at low pH are in accord with the recent observation that mixed acetate–phosphonate derivatives of TACN are more efficient chelators for carrier-free $^{68}Ga^{3+}$ than NOTA.[23]

Table 4. Half-Times ($t_{1/2}$) and Times of the Quantitative Formation ($t_{100\%}$) of the [Ga(L)] Complexes[a]

ligand	pH	$t_{1/2}$	$t_{100\%}$
TRAP-H	2.8	30 min	220 min
	1	21 h	15 days
	0	36 days	240 days
TRAP-OH	2.8	*c*	<5 min
	1	14 min	60 min
	0	3.8 days	31 days
TRAP-Pr[b]	2.8	*c*	<5 min
	1	3 min	12 min
	0.8	20 min	100 min
	0		12 days
NOTA	2.8	*c*	<5 min
	1	270 min	6 days
	0	*d*	*d*

[a]At 25 °C, 1:1 L:Ga molar ratio, c_{Ga} = 0.01 M. [b]From ref 28. [c]Not measurable because of a fast reaction. [d]No reaction observed.

We point out that Ga^{3+} complexation at pH values as low as 1 is of outstanding practical importance, as 0.1 M aqueous HCl is used for elution of some popular commercially available $^{68}Ge/^{68}Ga$ generators. Our study shows that, in contrast to NOTA, particularly TRAP-Pr is able to form a complex with Ga^{3+} rapidly under these conditions. In full accordance with this finding, our recently published results for the ^{68}Ga radiochemistry of the TRAP chelators[54] showed that these can indeed be readily labeled with ^{68}Ga at pH <1 and, therefore, also using the neat $^{68}Ge/^{68}Ga$ generator eluate. Despite not being applicable for all purposes, this method is compatible with many targeting moieties, such as most of the oligopeptides used for peptide receptor imaging. We therefore hold the view that TRAP ligands represent a big step forward in the development of kit production of ^{68}Ga radiopharmaceuticals in a "shake-and-shoot" synthetic approach, which is known from ^{99m}Tc radiotracers. Moreover, the highly selective and efficient Ga^{3+} complexation by TRAP ligands, as shown in this study, readily corresponds to the finding that much lower chelator concentrations are required for ^{68}Ga labeling.[54] TRAP-based ^{68}Ga radiopharmaceuticals can therefore be produced with hitherto unknown specific activitites, thereby allowing the smallest molar amounts of tracers to be administered for PET imaging. As an "ideal tracer" should be applied in the smallest possible amounts, to minimize interference with the biochemical equilibria governing the biosystem or organism under investigation, TRAP ligands can be considered of fundamental importance for future ^{68}Ga radiopharmaceutical research.

The kinetic inertness of the [Ga(trap-H)] and [Ga(trap-OH)] complexes was tested in 6 M $HClO_4$ at room temperature. No sign of decomposition was observed over 6 weeks, confirming the high kinetic inertness of these complexes against proton-assisted decomposition. The same complete inertness to proton-assisted decomplexation has been observed for the [Ga(NOTA)] and $[Ga(trap\text{-}Pr)]^{3-}$ complexes.[18,28]

Decomplexation in alkaline solutions proceeds for weeks for NOTA and TRAP-Pr complexes.[18,28] However, the situation was different for the [Ga(trap-OH)] complex. This in-cage complex is transformed to the out-of-cage complexes as discussed above. The reaction was followed only by $^{31}P\{^1H\}$ NMR spectroscopy as the out-of-cage complexes exhibit no ^{71}Ga NMR signal. The in-cage complex is fully stable up to pH ~5. At higher pH values, the transformation proceeds progressively faster. Equilibrium was reached after ~3 weeks

dx.doi.org/10.1021/ic202103v | Inorg. Chem. 2012, 51, 577–590

at pH 7.2, after ~6.5 days at pH 8.1, and after ~1.5 days at pH 9.5, and no signals or species other than those shown in the fully equilibrated samples (Figure 2) were observed during the course of the reaction. The rearrangement of the in-cage to the out-of-cage complex at physiological pH over hours to days does not interfere with the suitability of the TRAP-OH-based ligands for practical application as the decomposition is still slow in comparison with the half-life of the ^{68}Ga isotope and/or the labeled tracer pharmacokinetics (minutes to hours). Similar considerations concerning a relation of ^{68}Ga half-life and the rate of decomplexation of the Ga^{3+} complexes of DOTA and its monoamide in the slightly alkaline region have been pointed out recently.[10]

CONCLUSION

The properties of TRAP ligands, phosphinic acid derivatives of 1,4,7-triazacyclononane bearing different phosphorus substituents (H, Ph, CH_2OH, or $CH_2CH_2CO_2H$), were compared with those of the carboxylic acid analogue, NOTA. Thermodynamic studies showed that the phosphinic acid ligands are more acidic than NOTA and their acidity or basicity depends on the substituent on the phosphorus atom. The stability constants were determined for their complexes with Ga^{3+} and other metal ions; in addition, this parameter was redetermined for the [Ga(NOTA)] complex by applying the improved method. The thermodynamic stability of the gallium(III) complexes strongly depends on the ligand basicity, in the following order: [Ga(NOTA)] > [Ga(trap-Pr)] > [Ga(trap-OH)] > [Ga(trap-H)]. Although selectivity for Ga^{3+} complexation is very high for all TACN-based ligands, phosphinate ligands exhibit higher selectivity than NOTA for binding small metal ions. All investigated phosphinate ligands are able to bind trivalent gallium efficiently even at pH 0 and are, generally, more efficient than NOTA. The binding is very fast for TRAP-Pr and TRAP-OH, possessing coordinating substituents on the phosphorus atoms that increase the local metal ion concentration close to the macrocyclic cavity. Thus, the presence of phosphinic acid pendant arms improves the coordination ability of the ligands in acidic media, which represents one of the most important properties for the practical application in nuclear medicine. All complexes are fully inert against proton-assisted decomplexation. Our data suggest an importance of weakly coordinating side chains for the acceleration of the transfer of the metal ion into the macrocyclic cavity. Altogether, we found that the methylenephosphinic acid group is a potent alternative to the commonly used acetic acid pendant arm in macrocyclic ligands, and the phosphinic acid analogues of NOTA are excellent gallium(III) chelators, with ideal properties for ^{68}Ga-based PET imaging agent elaboration.

The in-cage [Ga(trap-OH)] complex exhibited unexpected behavior in solutions at pH >5. It slowly reacts with hydroxide anions to form out-of-cage complexes where an oxygen-only coordination environment is present. The possible structures of these species were suggested by DFT calculations, the study being only the second[55] in-depth computational treatment of gallium(III) complexes of macrocyclic ligands. The calculations showed some unexpected results. To form the out-of-cage complexes, hydroxide anion must be coordinated together with at least one deprotonated *P*-hydroxymethyl group. This deprotonation is facilitated by a polarization effect of a small and charged Ga^{3+} ion once it moves out; subsequently, a deprotonated hydroxymethyl group will coordinate. Simultaneously, a proton has to be moved into the ligand cavity where it is bound to a nitrogen atom and held in-cage by a network of intramolecular hydrogen bonds. Such reactivity is caused by the affinity of the hard Ga^{3+} ion for the hard oxygen atoms. This is the first direct observation of thermodynamically stable out-of-cage gallium(III) complexes in aqueous solution that are commonly assumed as kinetic intermediates during formation and decomplexation of the complexes of macrocyclic ligands.

EXPERIMENTAL SECTION

Materials and Methods. All reactants and solvents were commercially available analytical grade chemicals. The 1,4,7-triazacyclononane as a free base was purchased from CheMatech and as a trihydrochloride was prepared via modified Richman–Atkins cyclization.[56] Characteristic NMR spectra were recorded using Varian UNITY Inova (400 MHz) or VNMRS (300 MHz) spectrometers. ^{1}H and ^{13}C NMR shifts are referenced to the *t*-BuOH signal, and ^{31}P NMR shifts are referenced to 85% aqueous H_3PO_4. Elemental analyses were performed using the HERAEUS Varian EL III system. Mass spectra were recorded on a Bruker Esquire 3000 spectrometer with ESI as an ion source and ion trap as a detector in positive or negative mode.

1,4,7-Triazacyclononane-1,4,7-tris[methylene(phenyl)-phosphinic acid] (TRAP-Ph) and 1,4,7-Triazacyclononane-7-methyl-1,4-bis[methylene(phenyl)phosphinic acid] (^{Me}NO2P^{Ph}). 1,4,7-Triazacyclononane trihydrochloride (1.50 g, 6.29 mmol), paraformaldehyde (0.74 g, 24.67 mmol), and phenylphosphinic acid (13.40 g, 94.37 mmol) were mixed with 6 M aqueous HCl (15 mL). The solution was stirred at 90 °C for 24 h and, then, evaporated in vacuo. The crude product was purified on Dowex 50 resin (H^+ form, elution with water followed by 5% aqueous ammonia). The ammonia fractions were combined and evaporated in vacuo. The resulting mixture of TRAP-Ph and ^{Me}NO2P^{Ph} was purified on silica gel [elution with a 1:1 aqueous ammonia/EtOH mixture; R_f 0.85 (TRAP-Ph), 0.34 (^{Me}NO2P^{Ph})]. The fractions containing pure ^{Me}NO2P^{Ph} were evaporated in vacuo, yielding ^{Me}NO2P^{Ph}·2NH_3·8.5H_2O as a dark-brown oil (1.2 g, 32%). The fractions containing pure TRAP-Ph were combined and evaporated in vacuo. The target product was further purified on Dowex 1 resin (OH^- form, elution with water followed by 6 M aqueous HCl) to remove the traces of ammonia. After evaporation of HCl fractions containing the ligand in vacuo, the remaining oil was dissolved in a minimal amount of water and freeze-dried to give TRAP-Ph·2.4HCl·3.5H_2O (1.60 g, 35%).

TRAP-Ph. ^{1}H NMR (300 MHz, D_2O/NaOD, 25 °C): δ 2.72 (d, $^2J_{PH}$ = 6.9 Hz, N-C$\underline{H}_2$-P, 6H), 2.78 (s, ring C$\underline{H}_2$, 12H), 7.42–7.45 (m, $\underline{H}_{ar}$, 9H), 7.59–7.63 (m, $\underline{H}_{ar}$, 6H). ^{13}C{^{1}H} NMR (75.4 MHz, D_2O/NaOD): δ 50.2 (s, ring $\underline{C}H_2$, 6C), 54.7 (d, $^1J_{PC}$ = 100.1 Hz, $\underline{C}H_2$-P), 129.4 (s, $\underline{C}_{ar}$), 131.73 (s, $\underline{C}_{ar}$), 132.3 (s, $\underline{C}_{ar}$), 136.8 (d, $^1J_{PC}$ = 121.5 Hz, $\underline{C}_{ar}$-P). ^{31}P{^{1}H} NMR (121.4 MHz, D_2O/NaOD, 25 °C): δ 26.3 (s). MS (ESI, negative mode): m/z 612 [TRAP-Ph + Na^+ − 2H^+]. Anal. Calcd (%) for $C_{27}H_{36}N_3O_6P_3$·3.5H_2O·2.4HCl: C, 43.70; H, 6.17; N, 5.66; Cl, 11.47. Found: C, 43.76; H, 5.43; N, 5.63; Cl, 11.28.

^{Me}NO2P^{Ph}. ^{1}H NMR (300 MHz, D_2O/NaOD, 25 °C): δ 2.52 (s, N-C$\underline{H}_3$, 3H), 2.68 (s, ring C$\underline{H}_2$, 4H), 2.86–3.11 (m, ring C$\underline{H}_2$, 8H, and N-C$\underline{H}_2$-P, 4H), 7.50–7.57 (m, $\underline{H}_{ar}$, 5H), 7.68–7.74 (m, $\underline{H}_{ar}$, 5H). ^{13}C{^{1}H} NMR (75.4 MHz, NaOD, 25 °C): δ 41.8 (s, ring $\underline{C}H_2$), 47.1 (s, ring $\underline{C}H_2$), 51.2 (s, ring $\underline{C}H_2$), 53.5 (s, $\underline{C}H_3$), 56.1 (d, $^1J_{PC}$ = 105.1 Hz, $\underline{C}H_2$-P), 128.5 (d, $^2J_{PC}$ = 11.6 Hz, $\underline{C}_{ar}$), 130.8 (d, $^3J_{PC}$ = 9.4 Hz, $\underline{C}_{ar}$), 131.2 (s, $\underline{C}_{ar}$), 136.6 (d, $^1J_{PC}$ = 120.0 Hz, $\underline{C}_{ar}$-P). ^{31}P{^{1}H} NMR (121.4 MHz, D_2O/NaOD, 25 °C): δ 29.5 (s). MS (ESI, negative mode): m/z 450 [^{Me}NO2P^{Ph} − H^+]. Anal. Calcd (%) for $C_{21}H_{31}N_3O_4P_2$·8.5H_2O·2NH_3: C, 39.49; H, 8.52; N, 10.97. Found: C, 39.92; H, 8.43; N, 10.71; Cl, 11.28.

1,4,7-Triazacyclononane-1,4,7-tris(methylenephosphinic acid) (TRAP-H). Triazacyclononane (1.0 g, 7.75 mmol), paraformaldehyde (0.74 g, 24.67 mmol), and hypophosphorous acid (2.30 g, 34.8 mmol) were dissolved in water (20 mL) and stirred at room temperature for 48 h. The reaction mixture was evaporated in vacuo (bath temperature of <40 °C). The resulting oil was purified on a weak

dx.doi.org/10.1021/ic202103v | *Inorg. Chem.* 2012, 51, 577–590

cationic exchanger, Amberlite 50, with water elution. Fractions containing the pure ligand (by ^{31}P NMR) were combined, evaporated in vacuo as described above, and freeze-dried to give TRAP-H·H_2O (0.93 g, 33%). ^{1}H NMR (300 MHz, D_2O, 25 °C): δ 3.22 (d, $^2J_{PH}$ = 9.0 Hz, N-C$\underline{H}_2$-P, 6H), 3.45 (s, ring C$\underline{H}_2$, 12H), 7.14 (d, $^1J_{PH}$ = 543.9 Hz, P-$\underline{H}$, 3H). ^{13}C{^{1}H} NMR (150.9 MHz, D_2O, 25 °C): δ 52.20 (s, ring $\underline{C}H_2$), 56.21 (d, $^1J_{PC}$ = 90.5 Hz, N-$\underline{C}H_2$-P). ^{31}P{^{1}H} NMR (121.4 MHz, D_2O, 25 °C): δ 16.1 (s). ^{31}P NMR (121.4 MHz, D_2O, 25 °C): δ 16.2 (d, $^1J_{PH}$ = 543.9 Hz). MS (ESI, negative): m/z 362 [TRAP-H$^+$]. Anal. Calcd (%) for $C_9H_{24}N_3O_6P_3$·H_2O: C, 28.35; H, 6.87; N, 11.02. Found: C, 28.28; H, 6.70; N, 11.13.

1,4,7-Triazacyclononane-1,4,7-tris[methylene-(hydroxymethyl)phosphinic acid] (TRAP-OH). Triazacyclononane trihydrochloride (1.50 g, 6.23 mmol), paraformaldehyde (0.74 g, 24.67 mmol), and solid hypophosphorous acid (2.80 g, 42.42 mmol) were dissolved in water (20 mL) and stirred at room temperature for 48 h. The reaction mixture was evaporated in vacuo (bath temperature of <40 °C). The resulting oil was dissolved in 6 M HCl (50 mL). Paraformaldehyde (0.74 g, 24.67 mmol) was added, and the solution was heated at 105 °C in a bath for 24 h. The reaction mixture was evaporated in vacuo and purified on Dowex 50 resin (H^+ form, elution with water). The fractions containing product were combined, evaporated in vacuo, and freeze-dried to give TRAP-OH·0.3HCl·1.5-H_2O (2.16 g, 70%). ^{1}H NMR (300 MHz, D_2O, 25 °C): δ 3.47 (d, $^2J_{PH}$ = 6.8 Hz, N-C$\underline{H}_2$-P, 6H), 3.62 (s, ring C$\underline{H}_2$, 12H), 3.82 (d, $^2J_{PH}$ = 6.0 Hz, P-C$\underline{H}_2$-OH, 6H). ^{13}C{^{1}H} NMR (100.6 MHz, D_2O, 25 °C): δ 52.46 (s, ring $\underline{C}H_2$), 54.14 (d, $^1J_{PC}$ = 87.42 Hz, N-$\underline{C}H_2$-P), 60.36 (d, $^1J_{PC}$ = 113.8 Hz, O-$\underline{C}H_2$-P). ^{31}P{^{1}H} NMR (121.4 MHz, D_2O, 25 °C): δ 34.58 (s). MS (ESI, negative): m/z 474 [TRAP-OH + Na^+ − 2H^+]. Anal. Calcd (%) for $C_{12}H_{30}N_3O_9P_3$·1.5H_2O·0.3HCl: C, 29.34; H, 6.83; N, 8.55. Found: C, 29.30; H, 6.58; N, 8.28.

Potentiometric Measurements. Potentiometry was conducted according to the previously published procedures; for the preparation of stock solutions and chemicals, the equipment, electrode system calibration, titration procedures, and data treatment, see refs 10, 46b, and 57. The $Ga(NO_3)_3$ stock solution was acidified with aqueous HNO_3, and the excess of acids in the stock solution was determined independently by acid−base titration. Throughout the paper, pH means −log[H^+]. Protonation and stability constants were determined in 0.1 M (NMe_4)Cl at 25.0 °C with a pK_w of 13.81. Protonation constants and stability constants for the complexes with metal ions except Ga^{3+} were determined by in-cell titrations from data obtained in the pH range of 1.6−12 (or until precipitation of metal hydroxides) with ~40 points per full titration and four parallel titrations (c_L = 0.004 M; c_M = 0.004 or 0.002 M). The stability constants of the gallium(III) complexes were obtained by the out-of-cell method as described previously [starting pH of 1.5, 15−25 points per titration, points with precipitated $Ga(OH)_3$ excluded].[10,28] The full sets of determined constants (with their standard deviations given directly by the program) are given in the Supporting Information (Tables S1.1 and S1.2). The titration data were treated with OPIUM,[58] and the presented chemical models were chosen to have a chemical sense and to exhibit the best fitting statistics. Stability constants of metal hydroxy complexes were taken from ref 59.

Complexation−Decomplexation NMR Measurements. Formation of the Ga^{3+}−TRAP-R (R = H, Ph, or CH_2OH) complexes was followed by ^{31}P and ^{71}Ga NMR spectroscopy (25 °C, c_L = c_{Ga} = 10 mM). The experiments were conducted at pH 2.8 (1 M sodium chloroacetate buffer), pH 1.0 (0.1 M HCl), and pH 0 (1.0 M HCl); the solution pH was checked at the end of the complexation. The ^{71}Ga and ^{31}P{^{1}H} NMR signals were referenced to a 0.2 M aqueous $[Ga(OH)_4]^-$ solution and 85% aqueous H_3PO_4, respectively, in the insert tube. We preformed the complexes for other NMR measurements in solution by mixing of the ligand and Ga^{3+} salt stock solutions in a 1:1 Ga:L molar ratio and increasing the pH to 2.5 with a hydroxide solution (with heating if necessary for quantitative formation). Proton-assisted decomplexation of the complexes (c_{GaL} = 10 mM) in 6 M $HClO_4$ was followed by ^{31}P{^{1}H} NMR over a period of 6 weeks.

Computational Method. If not stated otherwise, all calculations were performed with the Gaussian 03 suite.[60] The geometries were optimized using a pure DFT functional with Ahlrichs' triple-ξ basis set (BP86/TZVP/auto).[61−63] The basis was automatically fitted to improve performance. Stationary points were confirmed as local minima by a frequency calculation (absence of imaginary frequencies). Transition states were also confirmed by a frequency calculation by the occurrence of one imaginary frequency. The conductor-like polarizable continuum model (C-PCM) was applied to model the influence of water (ε = 78.39) with the following nonstandard input.[64,65] Atomic radii were modeled with the United Force Field (UFF) model that uses individual spheres for all hydrogen atoms. The *nosymmcav* keyword was applied to break the symmetry of the cavity. The number of added spheres was decreased by setting O_{fac} equal to 0.8 and R_{min} equal to 0.5. In cases where the optimization did not converge, the GDIIS algorithm was applied to arrive at a stationary point.[66−68] Energies for the hydroxide anion and water molecule were taken from ref 49. Values reported therein were obtained at the BP/TZVP level of theory and with application of COMSO-RS, which combines the electrostatic advantages and computational efficiency of the dielectric continuum solvation model COSMO with a statistical thermodynamics method for the local interaction of surfaces. It also takes into account local deviations from dielectric behavior as well as hydrogen bonding (see ref 49 and references cited therein).

To obtain more accurate energies for the four TRAP-H diastereoisomers and the transition state of the isomerization, we optimized their geometries at the BP86/TZVP level of theory using the C-PCM solvent model with the alterations given above followed by a frequency calculation with the Gaussian 03 suite. With the obtained geometry, a single-point calculation was conducted with Gaussian 09 using Truhlar and Zhao's M06 functional and Ahlrichs' def2-TZVPP basis set.[69−72] The influence of the solvent water (water ε = 78.3553) was modeled with C-PCM and the alterations given above. Because the values for the thermal correction to the Gibbs free energy change only slightly with a different basis, they were taken from the TZVP calculation and added to the SCF energy obtained from the single-point calculation to yield more accurate Gibbs free energies.

■ ASSOCIATED CONTENT

Ⓢ Supporting Information

Additional experimental details of potentiometric titrations, experimentally determined protonation and stability constants, comparison of stability constants of more ligands, distribution diagrams, dependence of gallium(III) complexes on the basicity of the ligands, reactions considered in DFT calculations together with the complex species, summary of relative energies of all species, atomic coordinates of all species, diastereoisomers of the [Ga(trap-H)] complex, NMR spectra of [Ga(trap-R)] complexes, and time course of complexation of Ga^{3+} with TRAP-OH. This material is available free of charge via the Internet at http://pubs.acs.org.

■ AUTHOR INFORMATION

Corresponding Author

*Department of Inorganic Chemistry, Universita Karlova, Hlavova 2030, 12843 Prague 2, Czech Republic. Telephone: +420-22195-1263. Fax: +420-22195-1253. E-mail: petrh@natur.cuni.cz.

Present Address

‡Department of Pharmaceutical Radiochemistry, Technische Universität München, Walther-Meissner Strasse 3, 85748 Garching, Germany.

■ ACKNOWLEDGMENTS

Support from the Grant Agency of the Czech Republic (203/09/1056) and the Long-Term Research Plan of the Ministry of

Education of the Czech Republic (MSM0021620857) is acknowledged. The work was conducted under the framework of COST D38, CM0807, and BM607 Actions. J.Š. is thankful for support by the Grant Agency of Charles University (19310), and V.K. is thankful for support from RP MSMT 14/63. We are grateful to M. Pniok (Charles University, Prague, Czech Republic) for help with a large-scale synthesis of TRAP-Ph and MS measurements. J.N. and M.S. thank S. Schenk (BASF) for fruitful discussion. All the calculations were performed at the Universitätsrechenzentrum of the Friedrich-Schiller-Universität (Jena, Germany).

DEDICATION

Dedicated to Prof. Ernst Anders on the occasion of his 70th birthday.

REFERENCES

(1) (a) Gupta, T.; Beriwal, S. *Indian J. Cancer* **2010**, *47*, 126−133. (b) Bockisch, A.; Freudenberg, L. S.; Schmidt, D.; Kuwert, T. *Semin. Nucl. Med.* **2009**, *39*, 276−289.

(2) (a) Pichler, B. J.; Kolb, A.; Nägele, T.; Schlemmer, P. H. *J. Nucl. Med.* **2010**, *51*, 333−336. (b) Sauter, A. W.; Wehrl, H. F.; Kolb, A.; Judenhofer, M. S.; Pichler, B. J. *Trends Mol. Med.* **2010**, *16*, 508−515. (c) Boss, A.; Stegger, L.; Bisdas, L. S.; Kolb, A.; Schwenzer, N.; Pfister, M.; Claussen., C. D.; Pichler, B. J.; Pfannenberg, C. *Eur. Radiol.* **2011**, *21*, 1439−1446.

(3) (a) Zhernosekov, K. P.; Filosofov, D. V.; Baum, R. P.; Aschoff, P.; Bihl, H.; Razbash, A. A.; Jahn, M.; Jennewein, M.; Rösch, F. *J. Nucl. Med.* **2007**, *48*, 1741−1748. (b) Ocak, M.; Antretter, M.; Knopp, R.; Kunkel, F.; Petrik, M.; Bergisadi, N.; Decristoforo, C. *Appl. Radiat. Isot.* **2010**, *68*, 297−302.

(4) Fani, M.; André, J. P.; Mäcke, H. R. *Contrast Media Mol. Imaging* **2008**, *3*, 67−77.

(5) Wadas, T. J.; Wong, E. H.; Weisman, G. R.; Anderson, C. J. *Chem. Rev.* **2010**, *110*, 2858−2902.

(6) Rösch, F.; Riss, J. P. *Curr. Top. Med. Chem.* **2010**, *10*, 1633−1668.

(7) Rösch, F.; Baum, R. P. *Dalton Trans.* **2011**, *40*, 6104−6111.

(8) Clarke, E. T.; Martell, A. E. *Inorg. Chim. Acta* **1991**, *181*, 273−280.

(9) Clarke, E. T.; Martell, A. E. *Inorg. Chim. Acta* **1991**, *190*, 37−46.

(10) Kubíček, H.; Havlíčková, J.; Kotek, J.; Tircsó, G.; Hermann, P.; Tóth, É.; Lukeš, I. *Inorg. Chem.* **2010**, *49*, 10960−10969.

(11) (a) Wei, L.; Zhang, X.; Gallazzi, F.; Miao, Y.; Jin, X.; Brechbiel, M. W.; Xu, H.; Clifford, T.; Welch, M. J.; Lewis, J. S.; Quinn, T. P. *Nucl. Med. Biol.* **2009**, *36*, 345−354. (b) Li, J.; Vanbilloen, H.; Vermaelen, P.; Devos, E.; Mortelmans, L.; Bormans, G.; Ni, Y.; Verbruggen, A. *Bioorg. Med. Chem.* **2010**, *18*, 5274−5281.

(12) (a) Liu, S. *Adv. Drug Delivery Rev.* **2008**, *60*, 1347−1370. (b) Lattuada, L.; Barge, A.; Cravotto, G.; Giovenzana, G. B.; Tei, L. *Chem. Soc. Rev.* **2011**, *40*, 3019−3049.

(13) Heppeler, A.; Froidevaux, S.; Maecke, H. R.; Jermann, E.; Béhé, M.; Powell, P.; Hening, M. *Chem.—Eur. J.* **1999**, *5*, 1974−1981.

(14) Pool, S. E.; Krenning, E. P.; Koning, G. A.; van Eijck, C. H. J.; Teunissen, J. J. M.; Kam, B.; Valkema, R.; Kwekkeboom, D. J.; de Jong, M. *Semin. Nucl. Med.* **2010**, *40*, 209−218.

(15) Correia, J. G. G.; Paulo, A.; Raposinho, P. D.; Santos, I. *Dalton Trans.* **2011**, *40*, 6144−6167.

(16) Bandoli, G.; Dolmella, A.; Tisato, F.; Porchia, M.; Refosco, F. *Coord. Chem. Rev.* **2009**, *253*, 56−77.

(17) Cola, N. A.; Rarig, R. S. Jr.; Ouellette, W.; Doyle, R. P. *Polyhedron* **2006**, *25*, 3457−3462.

(18) (a) Craig, A. S.; Parker, D.; Adams, H.; Bailey, N. A. *J. Chem. Soc., Chem. Commun.* **1989**, 1793−1794. (b) Broan, C.; Cox, J. P.; Craig, A. S.; Kataky, R.; Parker, D.; Harrison, A.; Randall, A. M.; Ferguson, G. J. *J. Chem. Soc., Perkin Trans. 2* **1991**, 87−99.

(19) Jyo, A.; Kohno, T.; Terazono, Y.; Kawano, S. *Anal. Sci.* **1990**, *6*, 323−324.

(20) André, J. P.; Maecke, H. R.; Zehnder, M.; Macko, L.; Akyeld, K. G. *Chem. Commun.* **1998**, 1301−1302.

(21) Eisenwiener, K.-P.; Prata, M. I. M.; Buschmann, I.; Zhang, H.-W.; Santos, A. C.; Wenger, S.; Reubi, J. C.; Mäcke, H. R. *Bioconjugate Chem.* **2002**, *13*, 530−541.

(22) (a) Riss, P. J.; Kroll, C.; Nagel, V.; Rösch, F. *Bioorg. Med. Chem. Lett.* **2008**, *18*, 5364−5367. (b) McMurry, T. J.; Brechbiel, M. W.; Wu, C.; Gansow, O. A. *Bioconjugate Chem.* **1993**, *4*, 236−245. (c) Brechbiel, M. W.; McMurry, T. J.; Gansow, O. A. *Tetrahedron Lett.* **1993**, *34*, 3691−3694.

(23) Fellner, M.; Riss, P.; Loktionova, N.; Zhernosekov, K.; Thews, O.; Geraldes, C. F. G. C.; Kovács, Z.; Lukeš, I.; Rösch, F. *Radiochim. Acta* **2011**, *99*, 43−51.

(24) de Sá, A.; Prata, M. I. M.; Geraldes, C. F. G. C.; André, J. P. *J. Inorg. Biochem.* **2010**, *104*, 1051−1062.

(25) (a) Shetty, D.; Choi, S. Y.; Jeong, J. M.; Hoigebazar, L.; Lee, Y.-S.; Lee, D. S.; Chung, J.-K.; Lee, M. C.; Chung, Y. K. *Eur. J. Inorg. Chem.* **2010**, 5432−5438. (b) Shetty, D.; Jeong, J. M.; Ju, C. H.; Kim, Y. J.; Lee, J.-Y.; Lee, Y.-S.; Lee, D. S.; Chung, J.-K.; Lee, M. C. *Bioorg. Med. Chem.* **2010**, *18*, 7338−7347.

(26) Lukeš, I.; Kotek, J.; Vojtíšek, P.; Hermann, P. *Coord. Chem. Rev.* **2001**, *216*−*217*, 287−312.

(27) (a) Cole, E.; Copley, R. C. B.; Howard, J. A. K.; Parker, D.; Ferguson, G.; Gallagher, J. F.; Kaitner, B.; Harrison, A.; Royle, L. *J. Chem. Soc., Dalton Trans.* **1994**, 1619−1629. (b) Cole, E.; Parker, D.; Ferguson, G.; Gallagher, J. F.; Kaitner, B. *Chem. Commun.* **1991**, 1473−1475.

(28) Notni, J.; Hermann, P.; Havlíčková, J.; Kotek, J.; Kubíček, V.; Plutnar, J.; Loktionova, N.; Riss, P. J.; Rösch, F.; Lukeš, I. *Chem.—Eur. J.* **2010**, *16*, 7174−7185.

(29) Anderegg, G.; Arnaud-Neu, F.; Delgado, R.; Felcmann, J.; Popov, K. *Pure Appl. Chem.* **2005**, *77*, 1445−1495.

(30) Bevilacqua, A.; Galb, R. I.; Hebard, R. I.; Zompa, L. J. *Inorg. Chem.* **1987**, *26*, 2699−2706.

(31) Moedritzer, K.; Irani, R. R. *J. Org. Chem.* **1966**, *31*, 1603−1607.

(32) Remore, D. *J. Org. Chem.* **1978**, *43*, 992−996.

(33) Lázár, I.; Sherry, A. D. *Synthesis* **1995**, 453−457.

(34) Drahoš, B.; Pniok, M.; Havlíčková, J.; Kotek, J.; Císařová, I.; Hermann, P.; Lukeš, I.; Tóth, É. *Dalton Trans.* **2011**, *40*, 10131−10146.

(35) Bazakas, K.; Lukeš, I. *J. Chem. Soc., Dalton Trans.* **1995**, 1133−1137.

(36) Geraldes, C. F. G. C.; Marques, M. P. M.; Sherry, A. D. *Inorg. Chim. Acta* **1998**, *273*, 288−298.

(37) Lubal, P.; Kývala, M.; Hermann, P.; Holubová, J.; Rohovec, J.; Havel, J.; Lukeš, I. *Polyhedron* **2001**, *20*, 47−55.

(38) Kotková, Z.; Pereira, G. A.; Djanashvili, K.; Kotek, J.; Rudovský, J.; Hermann, P.; Elst, L. V.; Muller, R. N.; Geraldes, C. F. G. C.; Lukeš, I.; Peters, J. A. *Eur. J. Inorg. Chem.* **2009**, 119−136.

(39) Clegg, W.; Iveson, P. B.; Lockhart, J. C. *J. Chem. Soc., Dalton Trans.* **1992**, 3291−3298.

(40) Geraldes, C. F. G. C.; Sherry, A. D.; Marques, M. P. M; Alpoim, M. C.; Cortes, S. *J. Chem. Soc., Perkin Trans. 2* **1991**, 137−146.

(41) Lázár, I.; Ramasamy, R.; Brücher, E.; Geraldes, C. F. G. C.; Sherry, A. D. *Inorg. Chim. Acta* **1992**, *195*, 89−93.

(42) Geraldes, C. F. G. C.; Sherry, A. D.; Cacheris, W. P. *Inorg. Chem.* **1989**, *28*, 3336−3341.

(43) Drahoš, B.; Kubíček, V.; Bonnet, C. S.; Hermann, P.; Lukeš, I.; Tóth, É. *Dalton Trans.* **2011**, *40*, 1945−1951.

(44) O'Brien, P.; Salacinski, H.; Motevalli, M. *J. Am. Chem. Soc.* **1997**, *119*, 12695−12696.

(45) Hegetschweiler, K.; Ghisletta, M.; Fässler, T. F.; Nesper, R.; Schmalle, H. W.; Rihs, G. *Inorg. Chem.* **1993**, *32*, 2032−2041.

(46) (a) Kotek, J.; Lubal, P.; Hermann, P.; Císařová, I.; Lukeš, I.; Godula, T.; Svobodová, I.; Táborský, P.; Havel, J. *Chem.—Eur. J.* **2003**, *9*, 233−248. (b) Táborský, P.; Lubal, P.; Havel, J.; Kotek, J.; Hermann, P.; Lukeš, I. *Collect. Czech. Chem. Commun.* **2005**, *70*, 1909−1942.

(47) Cacheris, W. P.; Nickle, S. K.; Sherry, A. D. *Inorg. Chem.* **1987**, *26*, 958−861.

(48) Prata, M. I. M.; Santos, A. C.; Geraldes, C. F. G. C.; de Lima, J. J. P. *J. Inorg. Biochem.* **2000**, *79*, 359−363.
(49) Klamt, A.; Eckert, F.; Diedenhofen, M.; Beck, M. E. *J. Phys. Chem. A* **2003**, *107*, 9380−9386.
(50) Rong, M.; Welch, M. J.; Reibenspies, J.; Martell, A. E. *Inorg. Chim. Acta* **1995**, *236*, 75−82.
(51) Hermann, P.; Kotek, J.; Kubíček, V.; Lukeš, I. *Dalton Trans.* **2008**, 3027−3047.
(52) Geraldes, C. F. G. C.; Alpoim, M. C.; Marques, M. P. M.; Sherry, A. D.; Singh, M. *Inorg. Chem.* **1985**, *24*, 3876−3881.
(53) Morfin, J.-F.; Tóth, É. *Inorg. Chem.* **2011**, *50*, 10371−10378.
(54) Notni, J.; Šimeček, J.; Hermann, P.; Wester, H.-J. *Chem.—Eur. J.* **2011**, *17*, 14718−14722.
(55) Notni, J.; Pohle, K.; Peters, J. A.; Görls, H.; Platas-Iglesias, C. *Inorg. Chem.* **2009**, *48*, 3257−3267.
(56) Richman, J. E.; Atkins, T. J. *J. Am. Chem. Soc.* **1974**, *96*, 2268−2270.
(57) Försterová, M.; Svobodová, I.; Lubal, P.; Táborský, P.; Kotek, J.; Hermann, P.; Lukeš, I. *Dalton Trans.* **2007**, 535−549.
(58) (a) Kývala, M.; Lukeš, I. *International Conference Chemometrics '95*, Pardubice, Czech Republic, 1995, p 63. (b) Kývala, M.; Lubal, P.; Lukeš, I. IX. Spanish-Italian and Mediterranean Congress on Thermodynamics of Metal Complexes (SIMEC 98), Girona, Spain, 1998. The full version of OPIUM is available (free of charge) at http://www.natur.cuni.cz/_kyvala/opium.html.
(59) (a) Martell, A. E.; Smith, R. M. *Critical Stability Constants*; Plenum Press: New York, 1974−1989, Vols. *1*−*6*. (b) *NIST Standard Reference Database 46 (Critically Selected Stability Constants of Metal Complexes)*, version 7.0; National Institute of Standards and Technology: Gaithersburg, MD, 2003. (c) Baes, C. F., Jr.; Mesmer, R. E. *The Hydrolysis of Cations*; Wiley: New York, 1976.
(60) Frisch, M. J.; Trucks, G. W.; Schlegel, H. B.; Scuseria, G. E.; Robb, M. A.; Cheeseman, J. R.; Montgomery, J. J. A.; Vreven, T.; Kudin, K. N.; Burant, J. C.; Millam, J. M.; Iyengar, S. S.; Tomasi, J.; Barone, V.; Mennucci, B.; Cossi, M.; Scalmani, G.; Rega, N.; Petersson, G. A.; Nakatsuji, H.; Hada, M.; Ehara, M.; Toyota, K.; Fukuda, R.; Hasegawa, J.; Ishida, M.; Nakajima, T.; Honda, Y.; Kitao, O.; Nakai, H.; Klene, M.; Li, X.; Knox, J. E.; Hratchian, H. P.; Cross, J. B.; Bakken, V.; Adamo, C.; Jaramillo, J.; Gomperts, R.; Stratmann, R. E.; Yazyev, O.; Austin, A. J.; Cammi, R.; Pomelli, C.; Ochterski, J. W.; Ayala, P. Y.; Morokuma, K.; Voth, G. A.; Salvador, P.; Dannenberg, J. J.; Zakrzewski, V. G.; Dapprich, S.; Daniels, A. D.; Strain, M. C.; Farkas, O.; Malick, V.; Rabuck, A. D.; Raghavachari, K.; Foresman, J. B.; Ortiz, J. V.; Cui, Q.; Baboul, A. G.; Clifford, S.; Cioslowski, J.; Stefanov, B. B.; Liu, G.; Liashenko, A.; Piskorz, P.; Komaromi, I.; Martin, R. L.; Fox, D. J.; Keith, T.; Al-Laham, M. A.; Peng, C. Y.; Nanayakkara, A.; Challacombe, M.; Gill, M. W.; Johnson, B.; Chen, W.; Wong, M. W.; Gonzalez, C.; Pople, J. A. *Gaussian 03*, revision D.01; Gaussian Inc.: Wallingford, CT, 2004.
(61) Becke, A. D. *Phys. Rev. A* **1988**, *38*, 3098−3100.
(62) Perdew, J. P. *Phys. Rev. B* **1986**, *33*, 8822−8824.
(63) Schaefer, A.; Huber, C.; Ahlrichs, R. *J. Chem. Phys.* **1994**, *100*, 5829−5835.
(64) Barone, V.; Cossi, M. *J. Phys. Chem. A* **1998**, *102*, 1995−2001.
(65) Cossi, M.; Rega, N.; Scalmani, G.; Barone, V. *J. Comput. Chem.* **2003**, *24*, 669−681.
(66) Csaszar, P.; Pulay, P. *THEOCHEM* **1984**, *114*, 31−34.
(67) Farkas, Ö. Ph.D. Thesis, Eötvös Loránd University and Hungarian Academy of Sciences, Budapest, 1995.
(68) Farkas, Ö.; Schlegel, H. B. *J. Chem. Phys.* **1999**, *111*, 10806−10814.
(69) Frisch, M. J.; Trucks, G. W.; Schlegel, H. B.; Scuseria, G. E.; Robb, M. A.; Cheeseman, J. R.; Scalmani, G.; Barone, V.; Mennucci, B.; Petersson, G. A.; Nakatsuji, H.; Caricato, M.; Li, X.; Hratchian, H. P.; Izmaylov, A. F.; Bloino, J.; Zheng, G.; Sonnenberg, J. L.; Hada, M.; Ehara, M.; Toyota, K.; Fukuda, R.; Hasegawa, J.; Ishida, M.; Nakajima, T.; Honda, Y.; Kitao, O.; Nakai, H.; Vreven, T.; Montgomery, J. A., Jr.; Peralta, J. E.; Ogliaro, F.; Bearpark, M.; Heyd, J. J.; Brothers, E.; Kudin, K. N.; Staroverov, V. N.; Kobayashi, R.; Normand, J.; Raghavachari, K.; Rendell, A.; Burant, J. C.; Iyengar, S. S.; Tomasi, J.; Cossi, M.; Rega, N.; Millam, J. M.; Klene, M.; Knox, J. E.; Cross, J. B.; Bakken, V.; Adamo, C.; Jaramillo, J.; Gomperts, R.; Stratmann, R. E.; Yazyev, O.; Austin, A. J.; Cammi, R.; Pomelli, C.; Ochterski, J. W.; Martin, R. L.; Morokuma, K.; Zakrzewski, V. G.; Voth, G. A.; Salvador, P.; Dannenberg, J. J.; Dapprich, S.; Daniels, A. D.; Farkas, O.; Foresman, J. B.; Ortiz, J. V.; Cioslowski, J.; Fox, D. J. *Gaussian 09*, revision A.02; Gaussian, Inc.: Wallingford, CT, 2009.
(70) Zhao, Y.; Truhlar, D. G. *Theor. Chem. Acc.* **2008**, *120*, 215−241.
(71) Weigend, F.; Ahlrichs, R. *Phys. Chem. Chem. Phys.* **2005**, *7*, 3297−3305.
(72) Basis sets were obtained from the Extensible Computational Chemistry Environment Basis Set Database, version 02/02/06, as developed and distributed by the Molecular Science Computing Facility, Environmental and Molecular Sciences Laboratory, which is part of the Pacific Northwest National Laboratory (Richland, WA) and funded by the U.S. Department of Energy. The Pacific Northwest National Laboratory is a multiprogram laboratory operated by Battelle Memorial Institute for the U.S. Department of Energy under Contract DE-AC06-76RLO 1830. Contact Karen Schuchardt for further information.