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New Macrobicycles Containing a Tetralactam Moiety: Template Synthesis and Study of their Binding with Lanthanides

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Abstract—The synthesis of macrobicyclic cryptands **1**, **2** from an 18-membered diazatetralactam and 6,6′-bis(bromomethyl)-2,2′-bipyridine or 2,6-bis(3-bromomethyl-1-pyrazolyl)pyridine has been investigated using various metal carbonates. The analysis of the distribution products by gel permeation chromatography showed that macrocyclization process was markedly conditioned by cation template effects. Optimum yields were obtained for Li^+ and Na^+ in the preparation of **1** and **2**, respectively. The cryptate structure of the corresponding complexes of Ln(III) (Ln=Eu, Tb, Gd) is discussed on the basis of spectroscopic and photophysical data.

Introduction

The design and synthesis of macrocyclic ligands and their lanthanide complexes are the subject of intensive research owing to the paramagnetic, luminescence and Lewis acid properties of the lanthanide ions. Receptors which encapsulate these ions are widely used in biomedical applications as contrast agents in NMR imaging,[1] as luminescent probes,[2] and as catalysts in DNA or RNA hydrolysis.[3] A lanthanide complex suitable for these biomedical applications should be characterized by a high kinetic stability with respect to metal ion dissociation, even at a low concentration. Macrobicyclic ligands may fulfil this essential requirement. This is due to the three dimensional intramolecular cavity of the cage-type ligand, which provides an efficient shielding of the bound ion from interaction with other solute molecules.[4] At present, however, only a few cage-type lanthanide complexes of biochemical interest have been described.[5] The most salient example is the Eu(III) complex of the macrobicyclic tris(bipyridine) ligand[6] which is currently available for routine diagnostic use in homogeneous time-resolved fluoroimmunoassays (TR-FIA).[7]

Several synthetic paths may be followed for the construction of macrobicyclic ligands, namely the preparation of an intermediate macrocycle and its subsequent bridging (1:1

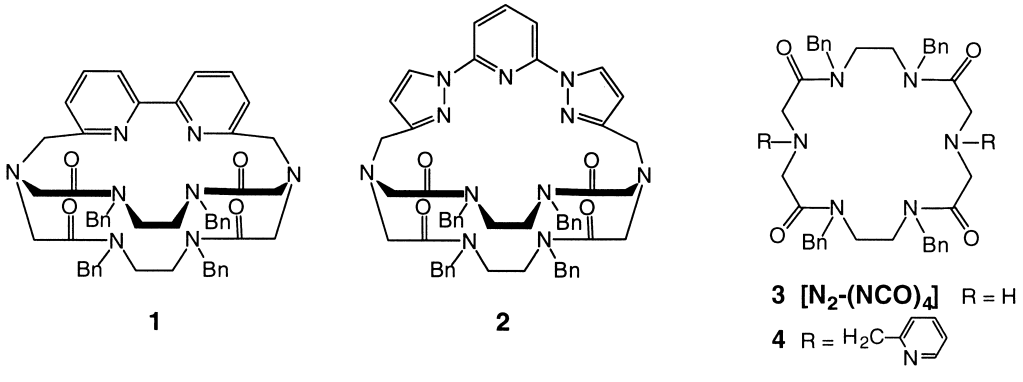

Figure 1. Schematic representation of the ligands.

Keywords: cryptands; template effects; amides; lanthanides.

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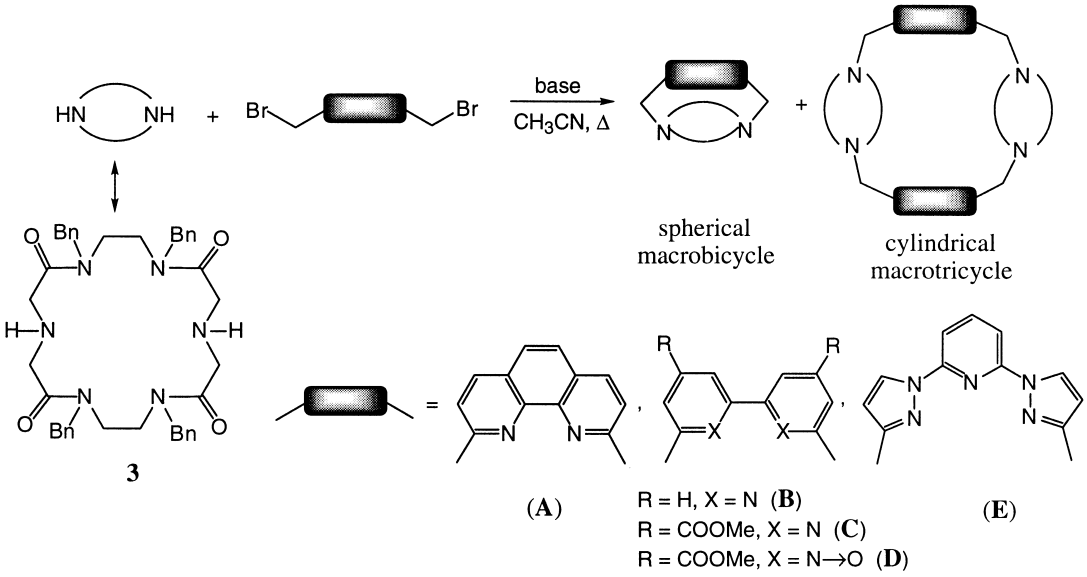

Scheme 1. Synthetic strategy for the obtention of tetralactam-based cryptands.

cyclization) or a direct macrobicyclization procedure from linear polyfunctional intermediates in 1:1, 1:2 or 2:3 cyclizations (see Fig. 3 in Ref. 8). The most frequently used approach relies on a stepwise procedure involving the treatment of azamacrocycles with bifunctional alkylating agents, in the presence of alkaline carbonates as bases. In numerous reports,[9] the final cyclization step gives the corresponding alkali–cation cryptates in high yields, without using high dilution techniques. It is suggested that an efficient template effect may occur in these reactions. However, conclusions have been drawn only from isolated yields and comparison with the effect of other cations, potentially able to increase the cyclization yield, has not been carried out.

In this context and in the course of our studies of luminescent lanthanide complexes,[10,11] we report here on the synthesis and some properties of lanthanide cryptates of the macrobicyclic ligands **1**, **2** (Fig. 1). They are built from a tetralactam [N_2-$(NCO)_4$] complexing moiety (**3** in Fig. 1) associated to a 2,2′-bipyridine (bpy) or a bis(*N*-pyrazolyl) pyridine (bpzpy) chromophoric unit. The selectivity of the macrobicyclization reaction was investigated using various metal ions as 'template' agents and gel permeation chromatography as analytical tool. Although seldom used in supramolecular chemistry, this analytical technique provided a direct evaluation of the distribution products from the crude reaction mixture. The spectroscopic and photophysical properties of the corresponding Ln(III) (Ln=Eu, Tb, Gd) complexes are also reported in the present work.

Results and Discussion

Cryptand synthesis

Three main methods have been applied to construct dissymetrical bicyclic cryptands starting from diazamacrocycles:

Table 1. Cyclization of 18-membered diaza-macrocycles with $BrCH_2$–(Arene)–CH_2Br in the presence of Na_2CO_3 as base ($1–5\times10^{-3}$ M dilution in CH_3CN)

Diaza-macrocycle[a]	Arene subunit[b]	Isolated yield[c] (%)		Reference
		Macrobicycle	Macrotricycle	
$[N_2O_4]$	**A**	50	–	9a
	B	40	–	17
	C	61	–	9d
$[N_2$–$(bpy)_2]$	**A**	71	–	9a
	B	62	–	9a
	C	40	–	9d
	D	45	–	18
$[N_2$–$(NCO)_4]$	**A**	7	54	16
	B	35[d]	21[d]	This work
	C	10	38	10b
	D	7	21	10b
	E	28	9	This work

[a] $[N_2O_4]$: 1,10-diaza-18-crown-6; $[N_2$-$(bpy)_2]$: 6,6′,6″,6‴-bis[iminodi(methylene)]bis(2,2′-bipyridine); $[N_2$-$(NCO)_4]$: see Fig. 1.
[b] See Scheme 1.
[c] Compounds isolated as their cryptates with Na^+ cation.
[d] In the presence of Li_2CO_3 as base; compounds isolated as their cryptates with Li^+ cation.

Table 2. Macrobicycle–macrotricycle distribution (MB/MT ratio) (MB/MT ratio as determined by GPC analysis; uncertainty ±10%) depending on the nature of the base in the reaction between [N_2-$(NCO)_4$] macrocycle and dibromomethyl heterocycles (see Scheme 1; reaction conditions: equimolar quantities of [N_2-$(NCO)_4$] (**3**) and dibromomethyl heterocycle (6×10^{-3} molar dilution and not high dilution procedures) and ten-fold excess of base in refluxing CH_3CN for 24 h)

Compound[a]	Base[b]						
	Li_2CO_3 (0.76)	Na_2CO_3 (1.02)	K_2CO_3 (1.38)	Cs_2CO_3 (1.67)	$CaCO_3$ (1.00)	$Eu_2(CO_3)_3$ (0.95)	iPr$_2$NEt
1[c]	2.50 (1.9)	0.25 (0.20)	0.60 (0.65)	0.80 (0.60)	1.15	1.60	0.85
2	3.0	3.80					

[a] See Fig. 1.
[b] Values in parentheses represent the six-coordinated cation radius in Å (taken from Ref. 22).
[c] Values in parentheses correspond to the analogous of compound **1** with arene subunit (**C**) of Scheme 1.

(i) condensation of diacid chlorides followed by a reduction step;[12,13] (ii) alkylation with bis halides or bis sulfonate esters[12,14] and, more recently; (iii) aminomethylation reaction using *N*,*N*′-bis(methoxymethyl)diazamacrocycles as reagents.[15] In these 1:1 cyclization processes, high dilution techniques or template synthesis are necessary to reduce polycondensations, which considerably decrease the yields of the final macrobicyclic compounds. We have previously reported[10,16] the synthesis of tetralactam-based cryptands by using an 18-membered tetralactam macrocycle [N_2-$(NCO)_4$] (**3** in Fig. 1) as starting material and 1,10-phenanthroline or 2,2′-bipyridine derivatives as cross-linkers (Scheme 1). The macrocyclization procedure using Na_2CO_3 as a base generated the sodium complex of both macrobicyclic and macrotricyclic structures, the former being obtained as secondary materials (Table 1). On the other hand, preliminary results[10b] indicated a strong dependence of the selectivity of the macrocyclization reaction, based on the tetralactam ring [N_2-$(NCO)_4$], according to the nature of the alkaline carbonate used. In contrast, the same reaction conditions yielded only macrobicyclic derivatives when analogous cross-linkers and [N_2O_4] or [N_2-(2,2′-bipyridine)$_2$] 18-membered macrocycles were used as building blocks (Table 1). A template effect of the sodium cation and a rigid-group effect of the bridging units introduced have been suggested to explain the selectivity of the last macrocyclization process favoring the monomeric structure.[9a]

In order to assess the true role of the cation in the macrobicyclization process and to improve its yield, the synthesis

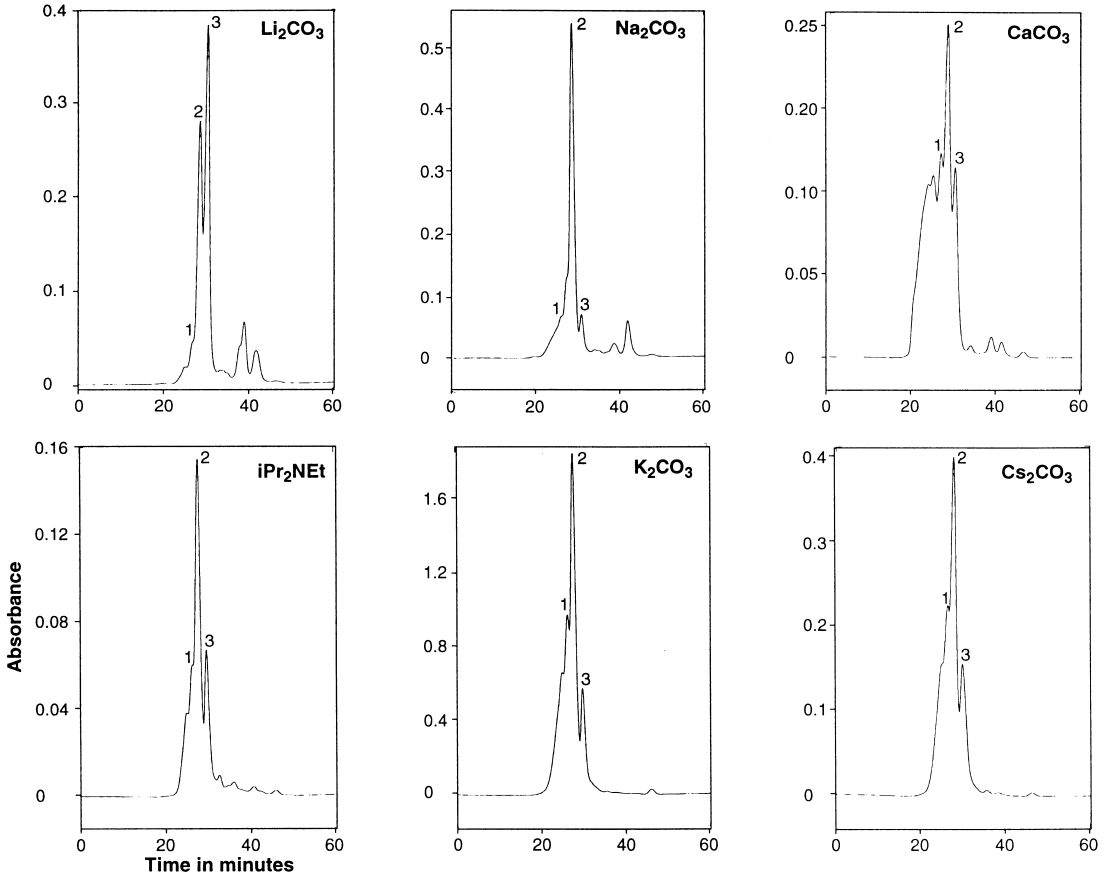

Figure 2. GPC chromatograms of the reaction mixtures obtained in experiments run in the presence of various bases for [N_2-$(NCO)_4$]=[6,6′-bis(bromomethyl)-2,2′-bipyridine]=6×10^{-3} M (see text). Conditions: PL gel column (5 mm, 100 Å), DMF as eluent. The higher oligomers (peaks 1) elute before the macrotricycle (peak 2) and macrobicyle (peak 3).

of cryptand **1** was tested with various metal carbonates [M_2CO_3 (M=Li, Na, K, Cs), $CaCO_3$ and $Eu_2(CO_3)_3$] in CH_3CN solution. Diisopropylethylamine was also used as a base to provide comparative experiments under a 'non-template' situation. Cyclization reactions were performed under identical conditions (see Table 2). To avoid material losses during the purification process, the reaction product distribution was directly evaluated from the crude reaction mixtures. The major problem in the analysis and the characterization of these macrobicycle, macrotricycle and higher derivatives resided in their almost identical spectroscopic features, including NMR spectra. Electrospray ionization mass spectrometry (ES-MS) has been recently used for the analysis of multiring macrocycles bearing diaza-18-crown-6 units.[19] However, quantitative speciation by ES-MS should be considered with caution owing to the large number of parameters influencing the response factors for different species.[20] Therefore, this technique may only be used in very specific cases.[21] Gel permeation chromatography (GPC) is much more reliable for this purpose. GPC provides a method for direct quantification of cyclic oligomers with the total exclusion of their linear counterparts and other undesired side-products. Although this technique is widely applied in polymer chemistry, its use for the analysis of relatively low-molecular organic compounds is little documented.

Fig. 2 shows the GPC chromatograms obtained from the crude reaction mixtures. Table 2 lists the macrobicycle/macrotricycle ratio (MB/MT) estimated from optical densities. The main trends found in Fig. 2 and Table 2 for **1** are:

1. A template effect appears to be particularly predominant when lithium carbonate is used. The selectivity ratio MB/MT is increased by a factor of 3 by replacing the organic base by lithium carbonate.
2. The other alkaline carbonates favor the production of macrotricycle. Among the alkaline metals, the sodium ion gives the lowest MB/MT ratio (0.25), in agreement with the results obtained from isolated yields reported for the other tetralactam-based cryptand series (Table 1).
3. Reactions with lithium and sodium bases are by far the cleanest (Fig. 2), with minor amounts of by-products (higher cyclic and linear oligomers). It is worth noting that the cesium ion[23] leads to no favorable effect on the competition between macrocyclization and polymerization.
4. A striking difference is noticed among sodium, calcium and europium bases. Although these ions share a similar size, the selectivity ratio (MB/MT) is increased by a factor of 4.5 for Ca^{2+} or 6.5 for Eu^{3+} vs. Na^+. Unfortunately, this favorable effect is compensated by a higher formation of by-products.

A similar trend is also seen in the macrocyclization reaction between [N_2-$(NCO)_4$] and dimethyl 6,6′-bis(bromomethyl)-2,2′-bipyridine-4,4′-dicarboxylate (see Table 2), namely the favored formation of the macrobicycle or the macrotricycle using lithium or sodium ions, respectively.

Alkaline salts (Li_2CO_3 and Na_2CO_3) were also tested in the synthesis of cryptand **2** (Table 2). In both cases, the formation of the macrobicyclic cryptand is favored. However, these cyclization reactions are accompanied by polymerization to a higher extent than in the preparation of **1.**

Taking into account these results, we scaled up the reactions leading to **1** and **2** using Li_2CO_3 and Na_2CO_3, respectively. The isolated yields are only modest (35% for **1** and 28 % for **2**), reflecting the difficulty of separating linear and cyclic oligomers in these reactions, even in relatively simple mixtures. However, the MB/MT ratio measured from the amounts of isolated products (Table 1) is consistent with the previous GPC analysis.

These results deserve further comment. The kinetic template effect[24] relies on molecular organization ('self-organization') induced by the metal ion which, by binding special sites, forces reactive groups to be properly oriented to give a predominant product. In order to determine if the metal ion really acts as a template, it appears essential to check if it can organize the available donor groups of the starting materials around itself. Thus, we have performed a simple experiment (Fig. 3) that gives additional support to the proposed template effect. Fig. 3 shows how the complexity of the proton-decoupled ^{13}C spectrum of the pyridine-armed tetralactam **4** (Fig. 1) was greatly simplified by adding one equivalent of lithium perchlorate. The large size of the ring should confer an important degree of conformational freedom to **4**. Besides, the relatively high-

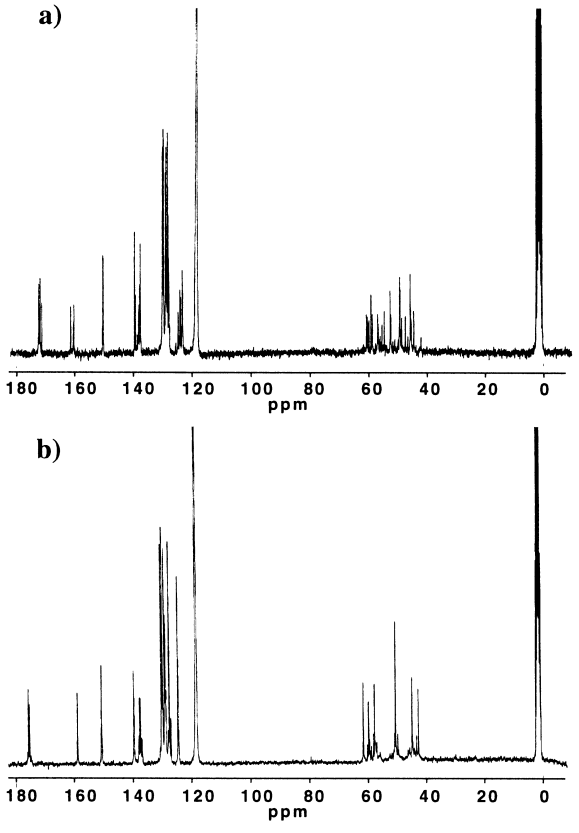

Figure 3. ^1H-decoupled ^{13}C NMR (75.5 MHz, CD_3CN, r.t.) spectra of pyridine-armed tetralactam **4** (Fig. 1): (a) free ligand, (b) $\mathbf{Li^+}$**.4** complex (1:1 stoichiometry).

energy barrier of rotation of the amide C–N bond[25] makes conformational interconversion to be slow in the NMR time scale at room temperature. These facts qualitatively explain the complex spectrum exhibited by **4** in which many conformations coexist at relatively slow equilibrium rates. Since Li^+ should not greatly affect the barrier of rotation of the amide C–N bond, the simplification exerted by the metal should be attributed to establishing new equilibria in which a species clearly dominates. The significant shifts observed upon complexation for amide carbonyl, pyridine arms and macrocycle ring carbons (see Fig. 3 and Experimental) suggest that pyridine-functionalized arms and tetramide ring co-operate in Li^+ binding. A reorganization of the conformation of the ligand **4** in CD_3CN solution also has been evidenced during the complexation process with the calcium ion.[26]

The strength of interactions between the metal and the substrates is also an important factor to be considered in the template effect: the more stable the complex, the higher the chemical yield of the pursued macroring should be. Best size fitting is usually the most common invoked factor. However, it is well established that the presence of amide functions in a complexone plays an important role in the enhancement of the selectivity in cations binding of higher charge density,[27] due to the high ground state dipole moments of these donor groups. Thus, the tendency to association with a tetralactam moiety should be higher for Li^+, Ca^{2+} and Eu^{3+} than for other alkaline ions. The results observed for the formation of 18-membered cryptand **1** may be rationalized by this way rather than by considering the best fit between the size of ion and the dimension of the molecular cavity. As a matter of fact, the ability of the compound **1** to cryptate the Eu^{3+} ion (vide infra) indicates that its macrobicyclic cavity may accommodate ions of similar size, especially Na^+ (see Table 2). The different orientation of the cyclization process observed in the preparation of cryptands of the same size (Table 1) arise certainly from a better affinity of the macrocyclic platform (e.g. $[N_2O_4]$) for sodium cation. Evidently, to fully rationalize these results, other considerations such as the base strength of the reaction medium and perhaps the interactions of the cation with the counterion must be taken into consideration.[28]

On the other hand, the metal ion size appears to be the predominant factor in the template effect observed for the preparation of cryptand **2** where a 21-membered ring is to be formed.

Complexation with lanthanides

The Ln^{3+} complexes (Ln=Eu, Tb, Gd) of the ligands **1** and **2** were prepared by ion exchange from $\mathbf{Li^+}$**.1** or $\mathbf{Na^+}$**.2** complexes in a manner similar to the preparation of the europium complexes of analogous cryptands.[10] Alkaline ions were displaced by simply refluxing for 24 h the corresponding complexes in the presence of a slight excess of $LnCl_3{\cdot}6H_2O$ in methanol solution. The resulting $\mathbf{Ln^{3+}}$**.1** and $\mathbf{Ln^{3+}}$**.2** complexes were isolated by precipitation with diethyl ether. They were soluble in MeOH and, unlike their parent alkaline counterparts, in H_2O.

Table 3 summarizes the MS, IR and UV spectral data of the alkaline and lanthanide complexes of ligands **1** and **2**. The luminescence maxima and lifetimes of the Eu^{3+} and Tb^{3+} complexes are also listed in Table 3.

These lanthanide complexes were characterized by electrospray mass spectrometry (ES-MS) recorded in methanol solutions (10^{-6}–10^{-7} M). In the ES-MS spectra of all complexes, the most abundant ion corresponds to $[(L-H)Ln]^{2+}$ species. This assignment was confirmed by the *m/z* 0.5 separation between adjacent peaks, indicating a doubly charged ion. In the case of europium and gadolinium complexes, we observed the characteristic isotopic abundance for these ions and the simulated isotopic patterns were superposable to the measured ones. Some less intense peaks are also detected and assigned to expected species with one or two associated chloride counterions. ES-MS study also evidenced a different binding ability of Ln^{3+} for these two ligands. Unlike $\mathbf{Ln^{3+}}$**.1**, a peak corresponding to the protonated free ligand was observed in the ES-MS mass spectra of $\mathbf{Ln^{3+}}$**.2**. The weaker ability of **2** to bind Ln^{3+} may arise from the expansion of the cavity size of this ligand as compared to **1**; this unfavorable factor overcame the presence of an additional coordination site in ligand **2**.

Table 3. MS, IR, UV and luminescence spectral data of the alkaline and lanthanide complexes derived from cryptands **1**, **2** (in methanol solution unless otherwise indicated)

	MS[a] (Base peak)	IR[b] ν(C=O) cm^{-1}	Absorption[c] λ_{max} (nm)	Emission[d,e] λ (nm)	Lifetime[d,f] τ (ms)
Li^+.**1**	861.4 $[L+Li]^+$	1643	296	347	[g]
Eu^{3+}.**1**	503.3 $[(L-H)Eu]^{2+}$	1610	307	616 (67%)	1.41
Tb^{3+}.**1**	506.4 $[(L-H)Tb]^{2+}$	1614	306	545 (56%)	1.77
Gd^{3+}.**1**	505.8 $[(L-H)Gd]^{2+}$	1614	306	340	[g]
Na^+.**2**	474.4 $[L+H+K]^{2+}$	1646	310	340	[g]
Eu^{3+}.**2**	530.3 $[(L-H)Eu]^{2+}$	1616	315	617 (58%)	0.84
Tb^{3+}.**2**	533.7 $[(L-H)Tb]^{2+}$	1615	317	545 (59%)	1.96
Gd^{3+}.**2**	533.2 $[(L-H)Gd]^{2+}$	1615	316	348	[g]

[a] ES^+ ionization mode.
[b] KBr discs.
[c] Wavelengths correspond to the λ_{exc} in the emission study.
[d] At 300 K.
[e] Assigned to the most intense emission band, $^5D_0 \rightarrow {}^7F_2$ and $^5D_4 \rightarrow {}^7F_5$ transitions for Eu^{3+} and Tb^{3+} complexes, respectively—the percentages of the total emission due to these transitions is given in parentheses.
[f] Experimental uncertainties: <10%.
[g] Short-lived fluorescence—lifetime not measured.

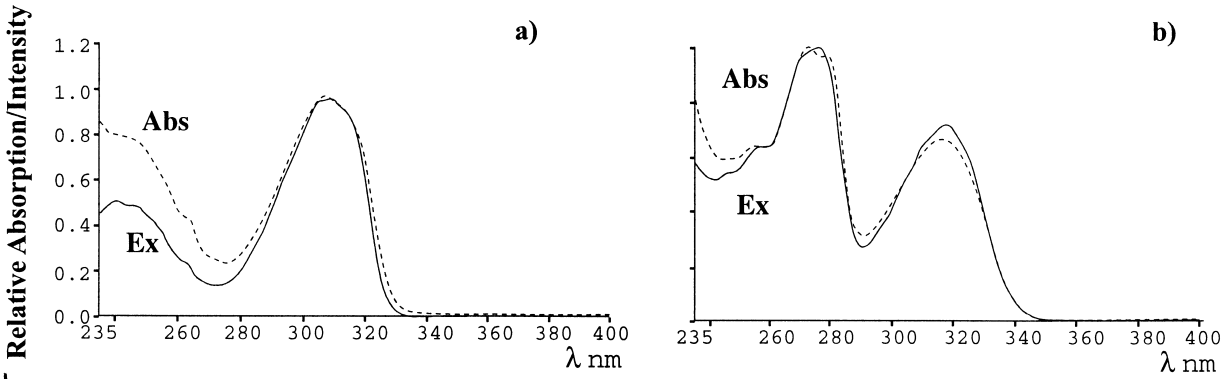

Figure 4. Normalised absorption (––) and excitation (—) spectra of Eu^{3+}**.1** (a) and Tb^{3+}**.2** (b) in methanol solution. Excitation spectra: λ_{obs}=616 (a), 545 (b) nm.

The IR spectra of the studied complexes are similar and display the characteristic vibration bands of their heterocyclic moieties in the 1570–600 cm^{-1} range,[29] together with a single and intense carbonyl stretching vibration assigned to the carboxamide group (~1615 cm^{-1}), which is shifted to lower wavenumbers with respect to the corresponding alkaline complexes (~1645 cm^{-1}). A variation of the same magnitude has been observed for lanthanide complexes with linear amides.[30] This clearly suggests the co-operative binding of all amide-carbonyl groups of the tetralactam ring.

The UV spectra and the luminescence properties of the complexes in methanol solution support the participation of the nitrogen heterocyclic atoms in the complexation. The UV spectra of all studied lanthanide complexes were very similar to those of their parent alkaline counterparts, but showed in all cases a bathochromic shift of ca. 5–10 nm. These are the expected shifts for a perturbation produced by the coordinated lanthanide ion which have been observed in other Ln(III) complexes with ligands derived from 2,2′-bipyridine[31] and 2,6-bis(*N*-pyrazolyl)pyridine.[32] In the case of Ln^{3+}**.2** complexes, the presence of two well-resolved absorption bands at 272 and 279 nm indicates that the two pyrazoles have a slightly different coordination mode.[33]

The characteristic metal-based luminescence was observed for europium and terbium complexes following excitation into the lowest-energy ligand-centered absorption band. The observed transitions were the $^5D_0 \rightarrow {}^7F_j$ (j=0, 1, 2, 3, 4) and $^5D_4 \rightarrow {}^7F_j$ (j=3, 4, 5, 6) in the case of Eu and Tb complexes, respectively. It can be seen in Fig. 4 that the metal luminescence corrected excitation spectra closely resemble the UV absorption profiles of the ligands. This suggests that energy transfer from the excited ligand to the emitting metal ion is effectively taking place and, therefore, the emitting states of the metal ion are being populated from the absorption of the bpy or the bpzpy chromophores.

All the measured luminescent decays are monoexponential, suggesting a unique chemical environment of the metal ion. On the other hand, luminescent lifetimes of these complexes are significantly larger than those of $EuCl_3$ (τ=0.26 ms) and $TbCl_3$ (τ=0.65 ms) in the same solvent.[34] This indicates[5c] that the ligands provide an efficient protection to the metal from hydroxylated solvent molecules in its close environment. To this effect, it is noteworthy that the lifetime of Eu^{3+}**.1** is twice as long as that of the Eu complex derived from the corresponding bilariat [N_2-$(NCO)_4$] ring, with bipyridine groups as side arms.[35]

These features observed by IR, UV and luminescence techniques can be explained by the chelation of the lanthanide ion at the same time by all the O-carbonyl atoms and the N-heterocyclic atoms, and confirm the expected cryptate structure of the Ln^{3+} complexes with macrobicyclic ligands **1** and **2**.

Conclusion

We present in this paper unambiguous evidence of the template effect exerted by metal cations in the preparation of macrobicyclic cryptands derived from a tetralactam moiety. The results are rationalized in terms of the interaction efficiency between the metal and the tetralactam ring, which is mainly governed, among other factors, by the charge/radius ratio of the metal ion. The Eu(III) and Tb(III) complexes derived from cryptands **1** and **2** are luminescent in methanol solution and spectral data evidence their cryptate structure, in which the ion is encapsulated in the intramolecular tridimensional cavity. The detailed photophysical study of these new complexes in aqueous solutions is now in progress, yet the preliminary results presented in this work seriously nominate these compounds as good candidates for luminescent labeling of biological materials.

Experimental

General

Melting points were determined on a Kofler apparatus. ^1H magnetic resonance spectra were recorded on Bruker AC-250, 400, 500 spectrometers. Data are reported in the following order: chemical shift δ in ppm, spin multiplicity, integration and assignment. ^{13}C magnetic resonance spectra were recorded on Bruker AC-250, 300 spectrometers. The

multiplicity of signals (*n*) is given under the interval. Infrared spectra were recorded on a Perkin–Elmer spectrometer (ν cm^{-1}) in potassium bromide. UV spectra (λ nm (ϵ M^{-1} cm^{-1})) were measured from solutions in CH_3OH on a Perkin–Elmer Lambda 17 spectrophotometer. Fluorescence spectra (λ nm (relative intensity %)) were obtained from solutions in CH_3OH with an LS-50B Perkin–Elmer spectrofluorimeter equipped with a Hamamatsu-R928 photomultiplier tube. Positive ES-MS spectra (mass range 400–2000, *m/z* (relative intensity %)) were recorded with a Perkin–Elmer SCIEX API 100 apparatus, in methanol solution (unless otherwise indicated). Elemental analyses were carried out by the 'Service Commun de Microanalyse élémentaire UPS-INP' in Toulouse. Chromatographic purifications were performed by high pressure liquid chromatography (Amicon silicagel 6–35 μm, Jobin-Yvon miniprep LC apparatus).

GPC analyses were performed on a Waters 600 instrument with a Waters 996 photodiode array detector. The wavelength used for the analyses were 290 or 310 nm. A PLgel column of 300×7.50 mm, with a 100 Å pore size and a 5 μm particle size was employed. The typical flow rate (DMF as eluent) was maintained at 0.25 ml min^{-1}. GPC retention times (GPC tr) are given in minutes.

The following compounds were prepared as described in the literature: 6,6′-bis(bromomethyl)-2,2′-bipyridine,[9a] dimethyl 6,6′-bis(bromomethyl)-2,2′-bipyridine-4,4′-dicarboxylate,[36] 2,6-bis(3-bromomethyl-1-pyrazolyl)pyridine.[33]

Diazatetralactam 3. This compound was prepared in five steps from iminodiacetic acid and *N*,*N*′-dibenzylethylenediamine, as reported previously.[16] ^1H NMR (250 MHz, CD_3CN): δ 3.26–4.06 (m, 16H,CH_2), 4.35–4.51 (m, 8H, CH_2Ph), 7.16–7.35 (m, 20H, Ar). ^{13}C NMR (62.9 MHz, CD_3CN): δ 42.2–45.9 ($n\geq7$, NCH_2), 48.8–52.2 ($n\geq9$, NCH_2CO, CH_2Ph), 127.9–130.0 ($n\geq8$, *C*H Ar), 137.2–138.3 ($n\geq4$, Cq Ar), 169.3–170.6 ($n\geq5$, *C*O).

***N*,*N*′-bis((2-Pyridyl) methyl) diazatetralactam 4.** This compound was prepared as described in the literature.[16] ^{13}C NMR (75.5 MHz, CD_3CN): δ 44.3–47.2 ($n\geq6$, NCH_2), 48.5–52.4 ($n\geq6$, CH_2Ph), 54.5–56.7 ($n\geq5$, CH_2CO), 58.4–60.4 ($n\geq5$, CH_2Py), 122.9, 123.1 (C_5, Py), 123.9, 124.1, 124.5 (C_3, Py), 127.4–128.0 ($n\geq5$, C_p, Ar), 128.4–128.8 ($n\geq4$, C_m, Ar), 129.1–129.8 ($n\geq6$, C_o, Ar), 137.3–137.9 ($n\geq4$, C_i, Ar), 138.9, 139.3 (C_4, Py), 149.9,150.0 (C_6, Py), 159.9, 160.1, 161.1 (C_2, Py), 171.1–172.2 ($n\geq6$, CO).

[Li.4]ClO_4. ^{13}C NMR (75.5 MHz, CD_3CN), preponderant signals: δ 42.7,44.9 (NCH_2), 50.5, 57.6, 59.6 (CH_2Ph, CH_2CO), 61.4 (CH_2Py), 124.4, 124.5 (C_3, C_5, Py), 127.4–130.2 ($C_{o,m,p}$ Ar), 137.2, 137.5 (C_i, Ar), 139.5 (C_4, Py), 150.4 (C_6, Py), 158.7 (C_2, Py), 175.2, 175.6 (CO).

Cryptand 1. A mixture of **3** (1 g, 1.5 mmol) and Li_2CO_3 (1.1 g, 14.8 mmol) in anhydre acetonitrile (300 ml) under Argon was heated to reflux for 1 h. Then, 6,6′-bis(bromomethyl)-2,2′-bipyridine (0.506 g, 1.48 mmol) was added in one portion. The resulting mixture was refluxed for further 24 h. After cooling to room temperature, the insoluble solid was filtered off and the filtrate evaporated to dryness. The solid residue was subjected to HPLC purification on silicagel eluting with dichloromethane-methanol (100:0→20:80) to give the LiBr complexes of macrobicycle **1** (0.51 g, 35% yield) and corresponding macrotricycle (0.33 g, 21% yield).

*Macrobicycle **1**:* white solid; mp 174–176°C. GPC tr=30. IR: ν 3400, 2927, 1643, 1575, 1472, 1452, 1431, 1361, 1278, 1232, 1169, 1131, 1080, 1029, 977, 874, 795, 736, 702, 632. UV: λ (ϵ) 245 (11800), 296 (11100).^1H NMR (500 MHz, $CDCl_3$): δ 1.96–5.28 (m, 28H, CH_2), 6.55, 6.81 (2d, 2H, J=7.5 Hz, $H_{5,5'}$ Bpy), 6.94–7.58 (m, 20H,C_6H_5), 7.79, 8.02 (2t, 2H, J=7.85 Hz, $H_{4,4'}$ Bpy), 8.17, 8.21 (2d, 2H, J=7.85 Hz, $H_{3,3'}$ Bpy). MS (CH_3CN): *m/z* 877.5 (27) $[L+Na]^+$, 861.4 (100) $[L+Li]^+$. Anal. Calcd For $C_{52}H_{54}N_8O_4$, LiBr, 2 H_2O (977.94): C, 63.87; H, 5.98; N, 11.46. Found: C, 64.08; H, 5.77; N, 11.19.

Corresponding macrotricycle: white solid, mp >230°C. GPC tr=28. IR: ν 3400, 2927, 1651, 1575, 1473, 1451, 1430, 1354, 1276, 1227, 1170, 1149, 1087, 1029, 978, 879, 796, 738, 702, 633. UV: λ (ϵ) 245 (21900), 295 (20700). ^1H NMR (400 MHz, $CDCl_3$): δ 2.10–5.34 (m, 56H,CH_2), 6.75–7.35 (m, 44H, C_6H_5, $H_{5,5'}$ Bpy), 7.62 (t, 4H, J=7.9 Hz, $H_{4,4'}$ Bpy), 7.85 (d, 4H, J=7.9 Hz, $H_{3,3'}$ Bpy). MS (CH_3CN): *m/z* 1748.9 (1) $[L+K]^+$, 1733.0 (4) $[L+Na]^+$, 1710.9 (2) $[L+H]^+$, 886.2 (15) $[L+Na+K]^{2+}$, 878.2 (100) $[L+2Na]^{2+}$, 867.3 (10) $[L+Na+H]^{2+}$. Anal. Calcd For $C_{104}H_{108}N_{16}O_8$, 3 LiBr·9H_2O (2132.8): C,58.57; H, 5.95; N, 10.51. Found: C,58.08; H, 5.48; N,10.35.

Cryptand 2. As described for **1**, using **3** (0.68 g, 1 mmol), 2,6-bis(3-bromomethyl-1-pyrazolyl)pyridine (0.4 g, 1 mmol) and Na_2CO_3 (1.06 g, 10 mmol). HPLC chromatography (dichloromethane–methanol 100:0→20:80) gave the NaBr complexes of macrobicyle **2** (0.295 g, 28% yield) and corresponding macrotricycle (0.106 g, 9% yield).

*Macrobicycle **2**:* white solid; mp 167–170°C. GPC tr=28. IR: ν 3420, 2926, 1646, 1585, 1532, 1472, 1455, 1385, 1360, 1311, 1232, 1174, 1103, 1065, 1026, 984, 878, 799, 741, 703, 614. UV: λ (ϵ) 252 (9700), 270 (12500), 310 (9800). ^1H NMR (500 MHz, $CDCl_3$): δ 2.80–5.45 (m, 28H, CH_2), 6.36 (d, 1H, J=2.7 Hz, H_4 Pz), 6.47 (d, 1H, J=2.7 Hz, $H_{4'}$ Pz), 6.76–7.35 (m, 20H, C_6H_5), 7.55 (d, 1H, J=8.0 Hz, H_5 Py), 7.72 (d, 1H, J=8.0 Hz, H_3 Py), 8.18 (t, 1H, J=8.0 Hz, H_4 Py),8.18 (d, 1H, J=2.7 Hz, H_5 Pz), 8.42 (d, 1H, J=2.7 Hz, $H_{5'}$ Pz). MS (CH_3CN): *m/z* 984.8 (32) $[L+K+2H_2O]^+$, 948.6 (28) $[L+K]^+$, 932.8 (8) $[L+Na]^+$, 474.4 (100) $[L+K+H]^{2+}$. Anal. Calcd For $C_{53}H_{55}N_{11}O_4$, NaBr, 3 H_2O (1067.05): C, 59.66; H, 5.76; N, 14.44. Found: C, 59.30; H, 5.62; N, 14.21.

Corresponding macrotricycle: white solid, mp >230°C. GPC tr=27. IR: ν 3440, 2925, 1649, 1605, 1586, 1531, 1496, 1473, 1451, 1387, 1357, 1214, 1144, 1078, 1042, 976, 946, 800, 775, 733, 699, 617. UV: λ (ϵ) 252 (22600), 270 (23700), 308 (20600).^1H NMR (400 MHz, $CDCl_3$): δ 1.78–5.34 (m, 56H,CH_2), 6.44 (d, 2H, J=2.7 Hz, H_4 Pz), 6.52 (d, 2H, J=2.7 Hz, $H_{4'}$ Pz), 6.94–7.50 (m, 42H, C_6H_5, H_5 Py), 7.51 (d, 2H, J=7.9 Hz, H_3 Py),

7.64 (t, 2H, J=7.9 Hz, H_4 Py), 8.07 (d, 2H, J=2.7 Hz, H_5 Pz), 8.54 (d, 2H, J=2.7 Hz, $H_{5'}$ Pz). MS (CH_3CN): m/z 1859.0 (6) $[L+K]^+$, 1843.1 (65) $[L+Na]^+$, 1821.2 (28) $[L+H]^+$, 933.1 (100) $[L+2Na]^{2+}$, 922.2 (28) $[L+H+Na]^{2+}$. Anal. Calcd. For $C_{106}H_{110}N_{22}O_8$, 3 NaBr, 9 H_2O (2291.04): C, 55.57; H, 5.63; N, 13.45. Found: C, 55.52; H, 5.13; N, 13.06.

General procedure for the preparation of lanthanides complexes

To a solution of the appropriate ligand (0.02 g, 1 equiv.) in methanol (10 ml) was added the lanthanide salt $LnCl_3{\cdot}6H_2O$ (1.1 equiv.). After 18 h of refluxing, the solvent was evaporated, and the residue dissolved in the minimum of methanol. Anhydre diethyl ether was added carefully until the apparition of a slight trouble. The mixture was cooled to 4°C, and the resulting precipitate was isolated after centrifugation.

[Eu.1]Cl_3. IR: ν 3396, 2936, 1610, 1496, 1455, 1431, 1380, 1360, 1308, 1280, 1243, 1179, 1103, 1080, 1016, 968, 884, 796, 741, 703, 661, 642. UV: λ 245, 307, 318sh. Luminescence (λ_{exc}=307 nm): λ 582 (2), 591 (10), 596 (12), 616 (100), 622sh (35), 650 (2), 655 (2), 691 (7), 695 (7), 703sh (4). MS: m/z1041.3 (61) $[(L-H)EuCl]^+$, 1005.4 (56) $[(L-2H)Eu]^+$, 503.3 (100) $[L-H)Eu]^{2+}$.

[Tb.1]Cl_3. IR: ν 3392, 2935, 1614, 1497, 1455, 1433, 1381, 1360, 1308, 1280, 1243, 1179, 1106, 1080, 1016, 969, 885, 795, 742, 703, 661, 642. UV: λ 245, 306, 317sh. Luminescence (λ_{exc}=306 nm): λ 491 (45), 545 (100), 585 (15), 590sh (10), 623 (8), 647 (1). MS: m/z 1047.5 (2) $[(L-H)TbCl]^+$, 1011.5 (2) $[(L-2H)Tb]^+$, 506.4 (100) $[(L-H)Tb]^{2+}$.

[Gd.1]Cl_3. IR: ν 3398, 2935, 1614, 1498, 1455, 1434, 1384, 1360, 1307, 1278, 1243, 1180, 1105, 1081, 1017, 970, 885, 796, 742, 704, 666, 643. UV: λ 245, 306, 317sh. MS: m/z1046.5 (9) $[(L-H)GdCl]^+$, 1010.4 (7) $[(L-2H)Gd]^+$, 505.8 (100) $[(L-H)Gd]^{2+}$.

[Eu.2]Cl_3. IR: ν 3400, 2927, 1616, 1534, 1482, 1462, 1385, 1357, 1320, 1236, 1176, 1069, 1033, 995, 890, 795, 740, 701, 665. UV: λ 253, 273, 278, 315. Luminescence (λ_{exc}=315 nm): λ 594 (24), 617 (100), 652 (3), 686sh (9), 699 (14). MS: m/z1132.3 (4) $[LEuCl_2]^+$, 1096.3 (3) $[(L-H)EuCl]^+$, 548.7 (36) $[LEuCl]^{2+}$, 530.3 (100) $[(L-H)Eu]^{2+}$.

[Tb.2]Cl_3. IR: ν 3400, 2928, 1615, 1534, 1481, 1466, 1455, 1384, 1355, 1317, 1234, 1176, 1068, 1028, 990, 892, 798, 741, 703, 666. UV: λ 255, 273, 279, 317. Luminescence (λ_{exc}=317 nm): λ 490 (43), 545 (100), 587 (15), 591sh (10), 622 (10), 646 (1). MS: m/z1138.4 (33) $[LTbCl_2]^+$, 1102.5 (40) $[(L-H)TbCl]^+$, 1066.5 (7) $[(L-2H)Tb]^+$, 551.8 (75) $[LTbCl]^{2+}$, 533.7 (100) $[(L-H)Tb]^{2+}$.

[Gd.2]Cl_3. IR: ν 3390, 2928, 1615, 1534, 1481, 1466, 1455, 1385, 1355, 1317, 1233, 1175, 1068, 1029, 989, 893, 797, 741, 703, 663. UV: λ 255, 273, 278, 316. MS: m/z1101.4 (8) $[(L-H)GdCl]^+$, 533.2 (100) $[(L-H)Gd]^{2+}$.

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