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Characterization of $^{111}In^{3+}$ Complexes of DTPA Amide Derivatives: Biodistribution and Clearance Studied by Gamma Imaging

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ABSTRACT. A large series of structurally related diethylenetriaminepentaacetic acid amide derivatives with different structures and lipophilic properties were synthesized and radiolabeled with $^{111}In^{3+}$. Two of the more hydrophobic compounds studied ($[^{111}In]L^9$ and $[^{111}In]L^{10}$) showed high affinity for human serum albumin (HSA). The biodistribution and clearance properties shown by all complexes upon injection in Wistar rats were followed by gamma imaging. The blood retention time of the chelates correlates better with their binding to HSA than with their hydrophilic/lipophilic ratio. Hydrophilic and negatively charged complexes undergo renal retention, while the majority of the lipophilic complexes are retained in the blood for a longer period of time and are cleared through the liver. NUCL MED BIOL 27;6:605–610, 2000. **© 2000 Elsevier Science Inc. All rights reserved.**

KEY WORDS. Albumin, Gamma imaging, $^{111}In^{3+}$ complexes, DTPA derivatives

INTRODUCTION

Among the chelating agents that have found applications in medicine, $DTPAH_5$ (diethylenetriaminepentaacetic acid) remains one of the most commonly used (14) because it forms stable complexes with many cations (12) and allows the preparation of bifunctional chelating derivatives, which can easily be linked to high molecular weight compounds. $[[^{111}In](DTPA)]^{2-}$ has been described as an ideal agent for scintigraphic studies of the cerebrospinal fluid pathway (10, 14). $DTPAH_5$ is also the ligand of choice in one of the widely used magnetic resonance imaging (MRI) contrast agents $[Gd(III)(DTPA)]^{2-}$ (Magnevist: Berlex; Wayne, NJ, USA) (31). One of the important characteristics of this ligand is that it allows functionalization via the carboxylic functional groups simply by using the DTPA-bisanhydride as a precursor (15, 23). The DTPA amide derivatives (either bisamides or monoamide) possess the same number of donor atoms (three nitrogens and five oxygens) as $DTPAH_5$, with a reduction of ionizable groups. These acyclic ligands have some advantages relative to macrocyclic analogues in biomedical applications because of their much higher complexation rates and their higher flexibility in coordinating cations compared to the macrocyclic compounds, although their generally faster dissociation kinetics and lower thermodynamic stability can constitute serious disadvantages (27).

$DTPAH_5$ is an octadentate ligand that can bind to a metal ion through five deprotonated carboxylate groups and the three tertiary amino groups (21). In the past, we had prepared a series of new DTPA mono and bis-amides (Fig. 1) by derivatizing one or two of the $DTPAH_5$ carboxylate groups to amide functionalities with a variety of substituents (2, 8). For some of them, the solution structure and dynamics of their In^{3+} complexes and their thermodynamic stability constants were obtained, with a small decrease in the thermodynamic stability of those chelates relative to $[In^{3+}DTPA]^{2-}$ (8).

In this work we report some *in vitro* and *in vivo* properties of this series of $^{111}In^{3+}$ complexes, comparing them with the known behavior of $[[^{111}In](DTPA)]^{2-}$ (10, 11), relevant for the assessment of the potential applications of the DTPA amide chelates in nuclear medicine. The ligands (L^1 to L^{13}) form a series of neutral or negatively charged complexes with In^{3+} with varying molecular properties, in particular, with a different lipophilic/hydrophilic character. An increased hydrophobicity may lead to stronger interactions of these chelates, for example, with proteins such as human serum albumin (HSA) or lipophilic entities, potentially altering their biodistribution. The factors determining biliary versus urinary excretion are chiefly influenced by three main chelate properties: polarity, molecular size and weight, and structure (4, 5, 18, 19). Here we also assess the correlation of the observed *in vitro* and *in vivo* behavior of these $^{111}In^{3+}$ complexes with those molecular properties, contributing to the design of new and more specific radiopharmaceuticals.

MATERIALS AND METHODS

Reagents and Apparatus

$[^{111}In]Cl_3$ was obtained from CIS-Biointernational (Sarclay, France); instant thin layer chromatography-silica gel (ITLC-SG) was from Gelman Sciences, Inc. (Ann Arbor, MI, USA); HSA (crystallized and lyophilized) was from Sigma (Madrid, Spain). All the ligands used in this work were synthesized and characterized as described elsewhere (1, 2, 8, 9, 16). Other reagents and solvents were obtained from either Aldrich or Sigma and were used as

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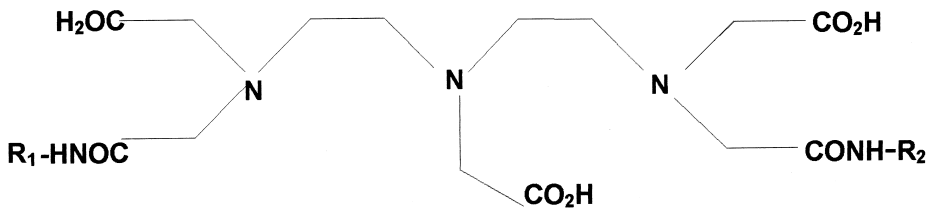

FIG. 1. Chemical structures of the DTPA amide derivatives. Bisamides, L^1 to L^{12}, $R_1 = R_2$ = methylenephenyl (L^1); ethylenephenyl (L^2); n-propyl (L^3); n-butyl (L^4); cyclohexyl (L^5); methylenecyclohexyl (L^6); 2-norbornyl (L^7); methinediphenyl (L^8); 2-ethylhexyl (L^9); n-heptyl (L^{10}); glucosyl (L^{11}); methoxyethyl (L^{12}); monoamide, R_2 = OH and R_1 = n-propyl (L^{13}).

received. All reagents for *in vivo* use were prepared sterile and pyrogen-free. A gamma camera-computer system (GE 400 GenieAcq, General Electric, Milwaukee, WI, USA) was used for acquisition and preprocessing. Data processing and display were performed on a personal computer using homemade software developed for the IDL 5.2 computer tool. A γ well-counter (DPC-Gamma C_{12}) with a Compaq DeskPro compatible computer was used for activity counting in the biodistribution studies.

Determination of 1-Octanol/Saline Partition Coefficients

$^{111}In^{3+}$ complexes were prepared in distilled water by mixing the ligand with [^{111}In]Cl_3 (in a 10:1 ratio) and adjusting the pH to approximately 7 with 0.1 M NaOH. The partition coefficients were determined by addition of 25 μL of $^{111}In^{3+}$-chelate solution in a test tube containing 1 mL of saline solution and 1 mL of 1-octanol. The tubes were vortexed and centrifuged for 3 min at 3,000 rpm. Then, aliquots of 100 μL of each phase were taken for radioactivity counting. The partition coefficients were calculated by dividing the net radioactivity counting of the organic phase by that of the aqueous phase. The results presented are the means of five determinations (with an SD of <0.01).

Calculation of 1-Octanol/Water Partition Coefficients

The 1-octanol/water partition coefficients for the free ligands (L^1 to L^{10}) were calculated using the log KOW program from SRC (Syracuse Research Corporation, New York, NY, USA) using a SMILE (Simplified Molecular Input Line Entry System) notation to represent each molecule. The calculations were based upon the additive-constitutive character of the partition coefficient (P) and the use of a substituent constant π_x, which is defined as Eq. (1) (7, 20):

$$\pi_x = \log P_x - \log P_H \quad (1)$$

where P_x is the partition coefficient of the derivative of a parent molecule with a partition coefficient P_H.

Interaction of Human Serum Albumin with $^{111}In^{3+}$ Chelates

Albumin solutions were freshly prepared by dissolving HSA in HEPES 0.1 M buffer to obtain 0.6 mM concentration (assuming 69 KDa as the HSA molecular weight). 0.2-mM L^9 and L^{10} water solutions were labeled with a small amount of [^{111}In]Cl_3 (in 0.1 N HCl), and afterwards $InCl_3$ (nonradioactive indium chloride) was added to obtain a 1:1 complex. These solutions were mixed with 2 mL of the HSA solution and were continuously stirred for 6 h and then left overnight at room temperature. A 1 mL sample was then dialyzed for 24 h, through membranes retaining molecules above 12 KDa at 281 K (30). Aliquots were collected inside and outside the membrane and counted in a γ well-counter. Total protein binding of InL chelates was calculated as percent of the applied InL solution that was retained inside the membrane (30).

Gamma Imaging

Stock solutions of the ligands were prepared in isotonic HEPES 0.1 M buffer and mixed with [^{111}In]Cl_3 (0.1 N HCl) to obtain 1:1 $^{111}In^{3+}$ complexes (with 10% ligand excess). The efficiency of labeling of the ligands with $^{111}In^{3+}$ was checked by chromatography (an ITLC-SG/butanone system), indicating that an approximately 100% radiochemically pure preparation of the complex was achieved. Gamma images and biological distributions for the $^{111}In^{3+}$ complexes were determined using 300 g Wistar rats. All animal studies were carried out in compliance with procedures approved by the appropriate institutional review committees. Conscious rats were allowed free access of food and water *ad libitum*. Groups of four animals (one group for each complex) were anaesthetized with Ketamine (50 mg/mL)/chloropromazine (2.5%) (10:3) and injected in the tail vein with approximately 150 μCi of the respective $^{111}In^{3+}$ chelate. The animals were then positioned in dorsal decubitus over the detector. Image acquisition was initiated immediately before radiotracer injection. Sequences of 180 images (of 10 s each), were acquired to 64 × 64 matrices. Blood samples were taken during the dynamic acquisition (at 5, 15, and 30 min) and subsequently counted in a γ well-counter. The clearance of radiotracer over time was followed by time-activity curves on images obtained from three regions of interest, thorax, liver, and left kidney. In addition, static data were acquired at 24, 48, and 72 h after the radiotracer injection.

Biodistribution Studies

Groups of four animals were injected with approximately 100 μCi of the complexes and sacrificed 2 h later. The major organs (liver, spleen, left kidney, heart, lung, intestines, brain) were removed, weighed, and counted in a γ well-counter.

RESULTS AND DISCUSSION

Partition Coefficients

The binding of a wide variety of nonionic organic compounds of miscellaneous structures to both bovine serum albumin (BSA) and bovine hemoglobin appears to depend almost exclusively on hydrophobic interactions and is linearly related to their octanol-water partition coefficient (13). The partitioning between octanol and water has been used to predict the ability of a compound to be absorbed and to distribute out of the vascular compartment into

TABLE 1. Measured 1-Octanol/Saline Partition Coefficients for $^{111}In^{3+}$ Complexes and Calculated Values for the Free Ligands

Ligand	Log P[a]	[^{111}In]L	Log P[b]
L^1	−4.26	[^{111}In]L^1	–
L^2	−3.27	[^{111}In]L^2	−1.23
L^3	−5.71	[^{111}In]L^3	–
L^4	−4.73	[^{111}In]L^4	–
L^5	−3.13	[^{111}In]L^5	−0.4
L^6	−2.15	[^{111}In]L^6	0.07
L^7	−1.27	[^{111}In]L^7	−1.28
L^8	−0.99	[^{111}In]L^8	0.31
L^9	−0.94	[^{111}In]L^9	1.08
L^{10}	−1.78	[^{111}In]L^{10}	−0.33
DTPA	–	[^{111}In]DTPA	−2.86[c]

[a] Calculated.
[b] Experimental.
[c] (24).

intracellular sites (17). In fact, this partition should model the lipid solubility of metal complexes, allowing them to remain within hepatocytes.

The octanol-saline partition coefficients determined for the $^{111}In^{3+}$ complexes of L^2 and L^5 to L^{10} are summarized in Table 1, together with the computed values for the free ligands. Values were compared with log P for [^{111}InDTPA] taken from literature (24). The log KOW program only allows calculations for covalent molecules, which is not, obviously, the case of the $^{111}In^{3+}$ chelates. Due to a poor extraction of the more hydrophilic $^{111}In^{3+}$ complexes by 1-octanol, this type of determination only gave accurate results for the less hydrophilic chelates. A good correlation between the calculated log P values for the ligands and the experimental data for the corresponding complexes was obtained. The only exception was [^{111}In]L^7, possibly due to steric hindrance resulting from the presence of the bulky norbornyl substituents, making this chelate not isostructural with the others.

In Vitro *Studies*

The present HSA binding study was carried out only for some of the more lipophilic In^{3+} chelates that gave quite efficient binding: 80% for [^{111}In]L^9 and 72% for [^{111}In]L^{10}. Some HSA binding studies of Gd^{3+}-DTPA-bisamides have been published previously using water proton relaxivity measurements, namely of GdL^4 and GdL^{10}, qualitatively demonstrating some degree of HSA interaction for GdL^{10} (9). A more recent relaxivity study with the Gd^{3+} complexes of some of the presently studied ligands, GdL^i (i = 1, 2, 5, 7, 9) (2), has suggested that the bisamide derivatives with greater hydrophobic character may originate stronger interactions with HSA, but the protein binding sites did not recognize aryl or norbonyl side chains, as well as short substituent groups. The only quantitative binding study for this series was for GdL^9, with an association constant $K_A = (5.7 \pm 1.3) \times 10^3\ M^{-1}$, giving a value of 72% for the bound fraction of GdL^9 (0.1 mM) in the presence of 0.6 mM HSA. Thus, the 80% HSA-bound fraction for InL^9 measured in the same conditions in the present study is comparable to that of GdL^9.

In Vivo *Studies*

Figure 2 summarizes the percentage of injected dose/g of tissue obtained from blood samples collected at 30 min after injection of some of the most representative $^{111}In^{3+}$ complexes. While the hydrophilic chelates are rapidly cleared, long retention times are observed for the more hydrophobic chelates, [^{111}In]L^{10} > [^{111}In]L^9. [^{111}In]L^9 has a strong interaction with HSA, stronger than that of [^{111}In]L^{10}, as evaluated from our studies, which agrees with qualitative binding studies with GdL^{10} (9), and its hydrophobicity as measured by the log P values (Table 1) is higher than that of $^{111}InL^{10}$. Its shorter retention in blood compared to [^{111}In]L^{10} indicates that this parameter may not be exclusively related with HSA binding.

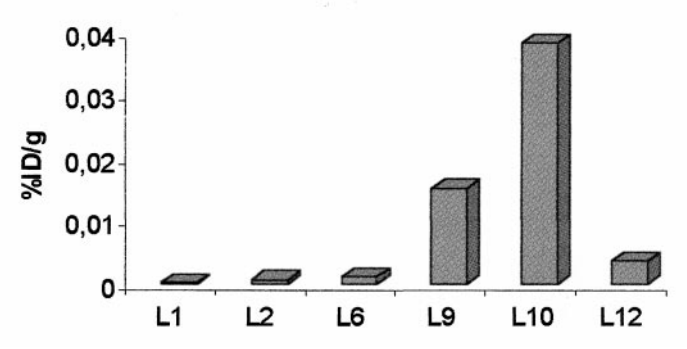

FIG. 2. Mean total %ID/g blood at 30 min after IV administration of some of the [^{111}In] chelates studied.

Gamma Imaging

Figure 3 shows the averaged time-activity curves obtained from dynamic acquisitions up to 30 min for two regions of interest (left kidney and liver) using some representative complexes. The mean thorax activity/pixel was considered as the background activity. The values of mean activity/pixel for each region of interest (RI), after background deduction, were used to obtain regional time/activity curves. The curves were normalized relative to the maximum activity obtained for each complex.

We can clearly divide the chelates studied into two main groups according to their clearance pathway. For the more hydrophilic $^{111}In^{3+}$ chelates (with the ligands, L^3, L^4, and L^{11}-L^{13}), the kidney activity/time curves, obtained from the dynamic acquisitions, generally show two different portions: an initial sharper one corresponding to uptake and a slower second one corresponding to slower clearance. This type of profile, exemplified in Figure 3 for [^{111}In]L^{12} and [^{111}In]L^{13}, indicates that those complexes undergo some renal retention. We would expect a more pronounced retention of [^{111}In]L^{13} (Fig. 3) when compared with the neutral hydrophilic complexes due to its negative charge. Instead, we can notice from Fig. 3 that these chelates have a very similar renal behavior. Some of these hydrophilic complexes show a small amount of activity at the liver-spleen region, which can be related with colloidal particles of indium hydroxide trapped by the reticuloendothelial system. However, the high thermodynamic stability of this type of complexes [*e.g.*, log K (InL^4) = 20.4] (8) seems to discard the hypothesis of premature dissociation of these $^{111}In^{3+}$ chelates. In a previous study (25), we reported the *in vitro* stability of $^{111}In^{3+}$ complexes of L^4 in blood serum and concluded that it has low dissociation (0.30% after 24 h), comparable to $[[^{111}In]DTPA]^{2-}$ (1.5% after 24 h) (26), indicating that conditional stability (defined as stability of the chelates at physiological conditions) is more important than thermodynamic stability to rationalize *in vivo*

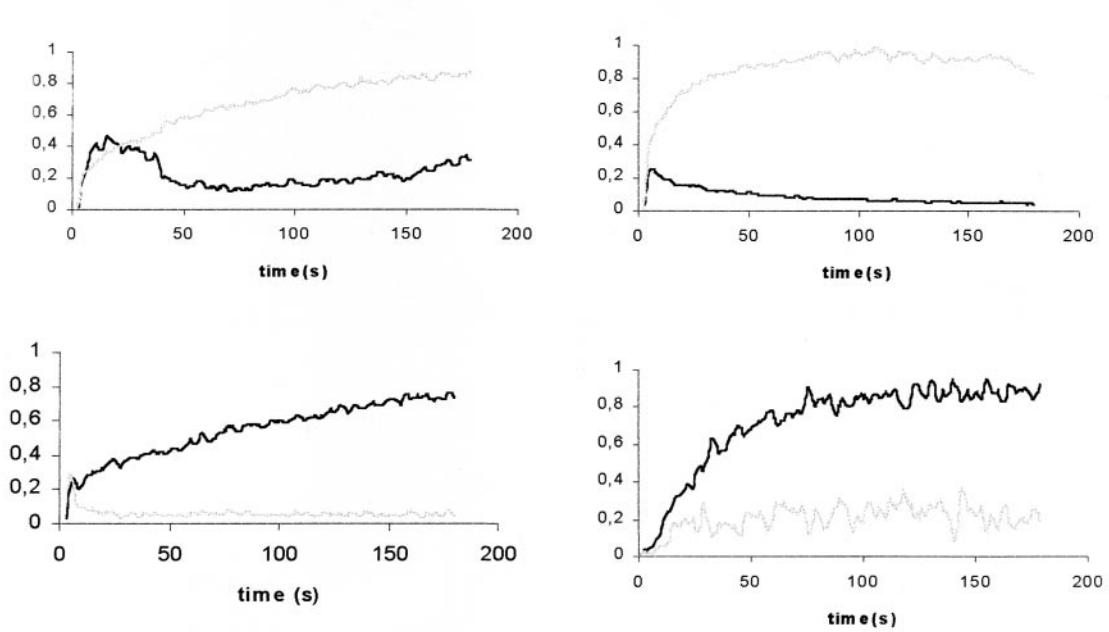

FIG. 3. Time-activity curves at the various regions of interest for (from left to right and top to bottom) [^{111}In]L^{6}, [^{111}In]L^{9}, [^{111}In]L^{12}, [^{111}In]L^{13}. Grey line: activity/time curve for liver; black line: activity/time curve for left kidney.

stability in blood serum. The thorax curve corresponds, in all the cases, only to blood activity.

The $^{111}In^{3+}$ complexes obtained with the ligands L^1, L^2, L^6, L^9, and L^{10} show a very different clearance pathway, which may be related to their increased lipophilicity. For the $^{111}In^{3+}$ complexes of L^1, L^2, L^6 (Fig. 3), L^9 (Fig. 3), and L^{10}, a pronounced uptake in the first few seconds was followed by a period of slow elimination of the radiotracer. This behavior suggests the implication of the liver in the metabolism of these complexes. Figure 4 shows the scintigraphic images of rats at 30 min after the injection of [^{111}In]L^{6} and [^{111}In]L^{12}. For the more lipophilic complexes, almost all the radioactivity was at the intestines, corresponding to the excretory phase of the chelates. However, a small activity in the bladder is also observed for most of these chelates, indicating that renal clearance is also present.

In the images obtained at 24 and 48 h after injection of all the $^{111}In^{3+}$ complexes (data not shown), a residual activity in the liver-spleen region was observed, possibly due to decomplexation and precipitation of indium hydroxides. As the complexes are thermodynamically quite stable (8), this fact should be related to

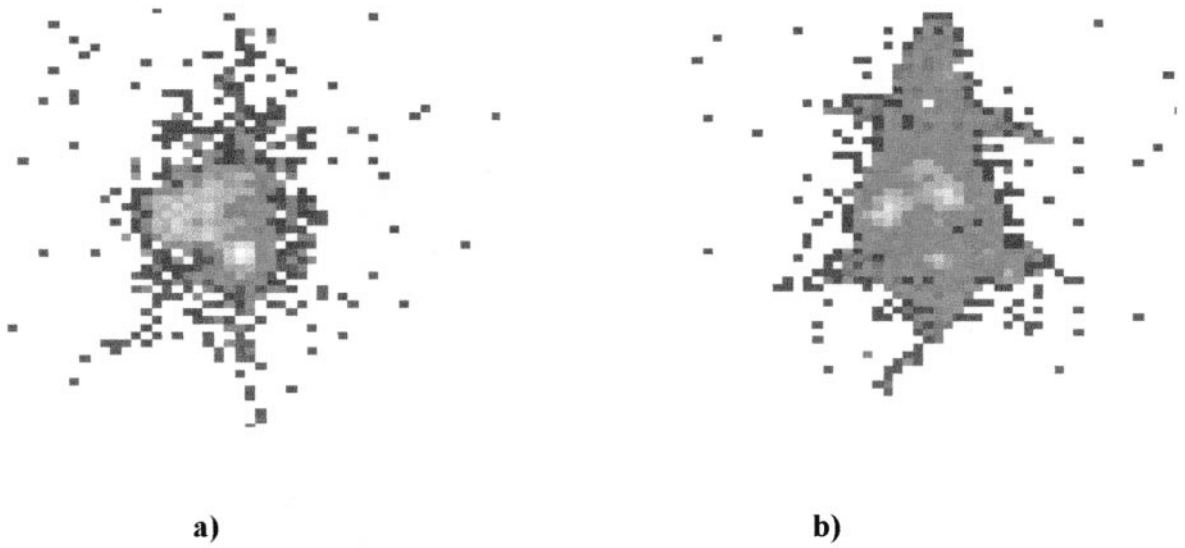

FIG. 4. Scintigraphic images at 30 min after injection with (a) [^{111}In]L^{6} and (b) [^{111}]InL^{12}.

TABLE 2. Mean Total %ID/g Tissue 2 h after IV Administration of [^{111}In] Chelates

	DTPA-amide ligand									
Tissue	**L^1**	**L^2**	**L^4**	**L^6**	**L^9**	**L^{10}**	**L^{11}**	**L^{12}**	**L^{13}**	**DTPA**
Liver	0.001	0.023	0.095	0.02	0.0012	0.021	0.013	0.024	0.036	0.01
Spleen	0.00025	0.001	0.016	0.0009	0.000085	0.024	0.028	0.023	0.023	0.006
Kidney	0.002	0.015	0.33	0.012	0.00061	0.010	0.129	0.24	0.18	0.126
Heart	0.00012	0.0011	0.006	0.00067	0.000039	0.001	0.0034	0.0027	0.023	0.005
Lung	0.00036	0.0038	0.009	0.0019	0.000094	0.0017	0.0058	0.0056	0.099	0.003
Small intestine	0.013	0.063	0.001	0.0001	0.044	0.050	0.011	0.012	0.025	0.0016
Large intestine	0.00019	0.006	0.04	0.022	0.000014	0.006	0.0032	0.004	0.01	0.0014
Brain	0.000001	0.00025	0.00076	0.00033	0.000008	0.00029	0.00067	0.00049	0.0036	0.0012

kinetic instability, which should influence their *in vivo* behavior (27).

Biodistribution Studies

The results of animal biodistribution studies at 2 h (in percent of injected dose per gram of organ or tissue) for the chelates studied are summarized in Table 2, along with data for $[[^{111}In]DTPA]^{2-}$, obtained in our laboratory, for comparative purposes. $[[^{111}In]DTPA]^{2-}$ is an extracellular complex and is known to be eliminated by glomerular filtration (10). Comparison of the images obtained at 30 min with the biodistribution data at 2 h shows that the unexpected low activity values for liver and gastrointestinal tract for L^1, L^2, L^6, L^9, and L^{10} were due to the rapid clearance of these chelates. Particularly noticeable from Table 2 are the low values of kidney activity for the latter chelates when compared with the hydrophilic ones. Based on the values obtained for the partition coefficients between octanol and water, we would not expect such a pronounced liver uptake for $[^{111}In]L^2$, $[^{111}In]L^6$, $[^{111}In]L^9$, or $^{111}[In]L^{10}$. This fact indicates that liver uptake is not only a function of lipophilicity. Conversely, other less obvious parameters govern the amount of radioactivity clearing through the liver as it has been pointed out by other previous studies with $^{67}Ga^{3+}$ and $^{111}In^{3+}$ chelates (3, 22, 23, 24).

We found no evidence of bone uptake, which would reflect uptake in the bone marrow that is seen when the In^{3+}-transferrin complex is formed (28, 29). Thus, this is evidence that the chelates remained intact for the time they stayed in blood. This agrees with thermodynamic stability constants and pM values reported elsewhere for some of these bis-amides chelates, which were found to be higher than that determined for the In^{3+}-transferrin complex (8), the main competitor for In^{3+} and Ga^{3+} in blood (28, 29).

None of the complexes passed through the blood-brain barrier, as expected for high molecular weight and/or low lipophilic complexes. In fact, several authors have estimated the molecular weight cut-off for brain extraction to be between 400 and 600 for substances with log $P > 2$ (6, 18).

CONCLUSION

In this work, we have used a large series of bisamide ligands (and one monoamide), previously synthesized, based on the structure of $DTPAH_5$, and have shown that simple substitutions in this ligand can cause major alterations in the biodistribution and clearance of their $^{111}In^{3+}$ chelates. While the more hydrophilic complexes clear by the kidneys, the more lipophilic chelates clear by the liver and gastrointestinal tract, thus demonstrating the influence of lipophilicity in the clearance of these $^{111}In^{3+}$ chelates. However, within this later group, the observed liver uptake is much more pronounced than what would be expected based on their experimental log P values. This clearly illustrates that liver uptake is not just a simple function of lipophilicity, but other, less obvious parameters govern the amount of radioactivity clearing through the liver (3, 22, 23, 24). On other hand, the complex charge does not seem to be very relevant in their *in vivo* behavior, at least in what concerns low negative-charged complexes. The blood retention of the chelates also correlates better with their binding to HSA than with their hydrophilic/lipophilic ratio.

The results of the present systematic structure-biodistribution study may contribute to the optimization of the properties of the potential radiopharmaceuticals derived from the $^{111}In^{3+}$ chelates of DTPA.

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References

1. Annie Bligh S. W., Chowdhury A. H. M. S., McPartlin M. and Scowen I. J. (1995) Neutral gadolinium (III) complexes of bulky octadentate DTPA derivatives as potential contrast agents for magnetic resonance imaging. *Polyhedron* **14,** 567–569.
2. Annie Bligh S. W., Chowdhury A. H. M. S., Kennedy D., Luchinat C. and Parigi G. (1999) Non-ionic bulky Gd(III) DTPA-bisamide complexes as potential contrast agents for magnetic resonance imaging. *Magn. Res. Med.* **41,** 767–773.
3. Bollinger J. E., Banks W. A., Welch M. J. and Roundhill D. M. (1996) Biodistribution of lipophilic complexes $^{59}Fe(RsalH_2)tach$ (R=H, NO_2 and OMe) and $^{68}Ga(NO_2salH_2)_3tach$. *Nucl. Med. Biol.* **23,** 645–652.
4. Chervu L. R., Nunn A. D. and Loberg M. D. (1982) Hepatobiliary radiopharmaceuticals. *Sem. Nucl. Med.* **12,** 5–17.
5. Chervu L. R. and Blaufox M. D. (1982) Renal radiopharmaceuticals—An update. *Sem. Nucl. Med.* **12,** 224–239.
6. Dischino D. D., Welch M. J., Lilbourn M. R. and Raichle M. E. (1983) Relationship between lipophilicity and brain extraction of C-11 labeled radiopharmaceuticals. *J. Nucl. Med.* **24,** 1030–1038.
7. Fujitec T., Iwasa J. and Hansh C. (1964) A new substituent constant π, derived from partition coefficients. *J. Am. Chem. Soc.* **86,** 5175–5183.
8. Geraldes C. F. G. C., Delgado R., Urbano A. M., Costa J., Jasanada F. and Nepveu F. (1995) Complexes of Ga^{3+} and In^{3+} with the N,N′′-bis(butylamide) derivative of diethylenetriaminepentaacetic acid: Stability constants and nuclear magnetic resonance studies in aqueous solution. *J. Chem. Soc. Dalton Trans.* 327–335.
9. Geraldes C. F. G. C., Urbano A. M., Alpoim M. C., Sherry A. D., Kuan K. T., Rajagolapan R., Maton F. and Muller R. N. (1995) Preparation, physico-chemical characterization, and relaxometry studies of various gadolinium(III)-DTPA-bis(amide) derivatives as potential magnetic resonance contrast agents. *Magn. Res. Imag.* **13,** 401–420.

10. Goodwin D. A., Song C. H., Finston R. and Martin P. (1973). Preparation, physiology and dosimetry of ^{111}In labelled radiopharmaceuticals for cisternography. *Radiology* **108,** 91–98.
11. Goodwin D. A., Goode R., Brown L. and Imborne C. J. (1971) ^{111}In-labeled transferrine for the detection of tumors. *Radiology* **100,** 175–179.
12. Hancock R. D. and Martell A. E. (1989) Ligand design for selective complexation of metal ions in aqueous solution. *Chem. Rev.* **89,** 1875–1914.
13. Helmer F., Kiehs K. and Hansch C. (1968) The linear free-energy relationship between partition coefficients and the binding and conformational perturbation of macromolecules by small organic compounds. *Biochemistry* **7,** 2858–2863.
14. Jurisson S., Berning D., Jia W. and Ma D. (1993) Coordination compounds in nuclear medicine. *Chem. Rev.* **93,** 1137–1156.
15. Khaw B. A., Cooney J., Edginton T. and Strauss H. W. (1986) Differences in experimental tumor localization of dual-labelled monoclonal antibody. *J. Nucl. Med.* **27,** 1293–1299.
16. Lammers H., Maton F., Pubanz D., van Laren M. W., van Bekkum H., Merbach A. E., Muller R. N. and Peters J. (1997) Structures and dynamics of lanthanide(III) complexes of sugar-based DTPA-bis (amides) in aqueous solution: A multinuclear NMR study. *Inorg. Chem.* **36,** 2527–2538.
17. Leo A., Hansh C. and Elkins D. (1971) Partition coefficients and their uses. *Chem. Rev.* **71,** 525–559.
18. Levin V. A. (1980) Relationship of octanol/water partition coefficients and molecular weight to rat brain capillary. *J. Med. Chem.* **23,** 682–684.
19. Levine W. G. (1978) Biliary excretion of drugs and other xenobiotics. *Annu. Rev. Pharmacol. Toxicol.* **18,** 81–96.
20. Lewis D. F. V. (1989) The calculation of molar polarizabilities by the CNDO/2 method: Correlation with the hydrophobic parameter, log P. *J. Comp. Chem.* **10,** 1145–1151.
21. Maecke H. R., Riesen A. and Ritter W. (1989) The molecular structure of Indium DTPA. *J. Nucl. Med.* **30,** 1235–1239.
22. Madsen S. L., Bannochie C. J., Martell A. E., Mathias C. J. and Welch M. J. (1990) Investigation of physicochemical and in vivo behavior of diastereomeric iron-59, gallium-68 and indium-111-EHPG trivalent metal complexes. *J. Nucl. Med.* **31,** 1662–1668.
23. Meares C. F. (1986) Chelating agents for the binding of metal ions to antibodies. *Nucl. Med. Biol.* **13,** 311–318.
24. Mathias C. J., Sun Y., Welch M. J., Green M. A., Thomas J. A., Wade K. R. and Martell A. E. (1988) Targeting radiopharmaceuticals: Comparative biodistribution studies of gallium and indium complexes of multidentate ligands. *Nucl. Med. Biol.* **15,** 69–81.
25. Prata M. I. M., Ribeiro M. J., Santos A. C., Peters J. A., Nepveu F., Geraldes C. F. C. G. C. and de Lima J. J. P. (1998) *In Vitro* and *in vivo* behaviour of ^{111}In complexes of polyaminocarboxylate ligands: Stability, biodistribution and excretion studied by gamma imaging. In: *Modern Trends in Radiopharmaceuticals for Diagnostic and Therapy*, Proceedings of a symposium IAEA, pp. 505–511.
26. Riesen A., Kaden T. A., Ritter W. and Maecke H. (1989) Synthesis and X-ray structural charactherisation of seven co-ordinate macrocyclic In^{3+} complexes with relevance to radiopharmaceuticals applications. *J. Chem. Soc. Chem. Comm.* 460–462.
27. Rocklage S. M., Watson A. D. and Carvlin M. J. (1992) *Magnetic Resonance Imaging*, Vol. 1. Mosby Year Book, St. Louis.
28. Thakur M. L. (1977) Gallium-67 and Indium-111 radiopharmaceuticals. *Int. J. Appl. Rad. Isotopes* **28,** 183–201.
29. Thomas E. (1986) Exchange of iron by gallium in siderophores. *Biochemistry* **25,** 4629–4633.
30. Vanlic-Razumenic N., Joksimovic J., Ristic B., Tonic M., Beatovó S. and Ajdinovic B. (1993) Interaction of ^{99m}Tc-radiopharmaceuticals with transport proteins in human blood. *Nucl. Med. Biol.* **20,** 363–365.
31. Weinman H. J., Brash R. C., Press W. R. and Wesbey G. E. (1984) Characteristics of gadolinium-DTPA complex: A potential NMR contrast agent. *Am. J. Roentgenol.* **1142,** 619–624.