ARTICLE

OBC
www.rsc.org/obc

Design and synthesis of a novel family of semi-rigid ligands: versatile compounds for the preparation of ^{99m}Tc radiopharmaceuticals

Julien Le Gal,[a] Laure Latapie,[a] Marie Gressier,[a] Yvon Coulais,[b] Michèle Dartiguenave[a] and Eric Benoist *[a]

[a] *Laboratoire de Chimie Inorganique, EA 807, Université Paul Sabatier, Bat. IIR1, 118, route de Narbonne, 31062 Toulouse, France*

[b] *Laboratoire "Traceurs et traitement de l'image", Faculté de Médecine de Purpan, EA 3033, 133, route de Narbonne, 31062 Toulouse, France*

Received 4th November 2003, Accepted 22nd January 2004
First published as an Advance Article on the web 18th February 2004

Synthetic pathways to a range of novel semi-rigid tetradentate ligands, with sulfur, amido or amino donor groups, designed to coordinate technetium 99m have been developed. The technetium-99m complexes have been prepared and their stabilities in serum and by metathesis reaction *via* cysteine exchange reactions were compared. HPLC comparison of two ^{99m}Tc-complexes and their rhenium analogues led to the first proof of the nature of our radioactive complexes.

Introduction

Technetium chemistry attracts considerable research attention owing to the prominent use of the metastable ^{99m}Tc isomer (half life of 6.02 h, γ emitter with energy 140 KeV and convenient availability from the ^{99}Mo/^{99m}Tc generator) in diagnostic nuclear medicine.[1] The efforts made in this field in recent years have led to the synthesis of new and more efficient radiopharmaceuticals.[2–4] Several types of ligand frameworks have been developed. Among them, N_2S_2 or N_3S tetradentate ligands are known to form stable technetium(V) complexes, exemplified by compounds (**1**–**4**) shown in Fig. 1.[5–8]

Our research has focused on the development of a novel family of ^{99m}Tc-specific ligands whose design features, illustrated in Fig. 2, may be summarised as follows: (i) an aromatic ring to enhance the sp^2 character of the nitrogen atom bonded to the metal and promote rigidity of the square pyramidal base, to favour and stabilise the chelate ring by an entropic effect (even if in some cases the stiffening of the ligand should give an unfavourable energetic contribution for the process of complex formation); (ii) amide and thiol functions for ^{99m}Tc complexation; (iii) *ortho-N,O,S*-substitution to compare the stability of the corresponding technetium complexes. Moreover, another advantage of including an aromatic cycle in the chelate ring compared to the semi-rigid cyclohexyl moiety like in CDTA derivatives[9] is its ease of functionalisation in order to prepare bifunctional chelating agents.

In a previous communication,[10] we noticed that the presence of an hydroxyl group at the 2-position of the ring greatly

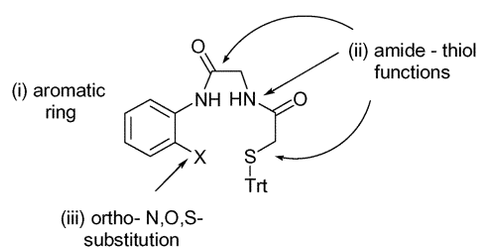


Fig. 2 Design features of novel semi-rigid ^{99m}Tc-ligand.

enhances the stability of the technetium complex compared to the compound bearing a methoxy moiety. To rationalize these first results, we decided to introduce at the 2-position a non-, partially- or fully-substituted amino group in order to check the influence of the aromatic group substitution on the stability of their corresponding ^{99m}Tc complexes and to determine the best candidates for a radiopharmaceutical use. In this paper, we describe the synthesis and characterisation of a range of new unsymmetrical tetradentate ligands, the ^{99m}Tc-labelling procedure and the stability of the different radioactive complexes. HPLC comparison of two ^{99m}Tc complexes and their rhenium analogues led to the first proof of the nature of our radioactive complexes.

Results and discussion

If it has been shown that the use of semi-rigid tetradentate ligands improved the stability of the resulting complexes,[11] only

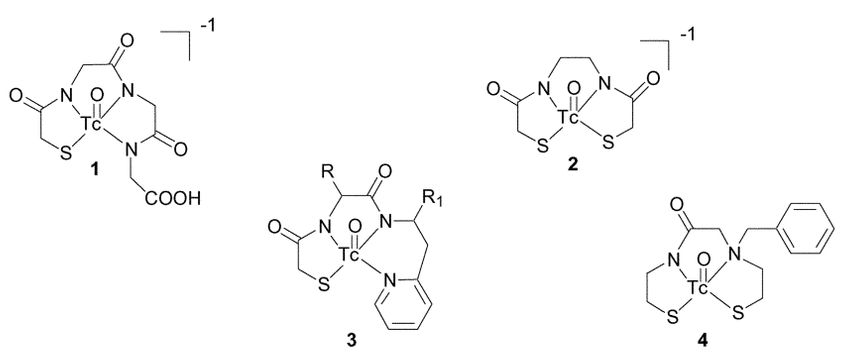
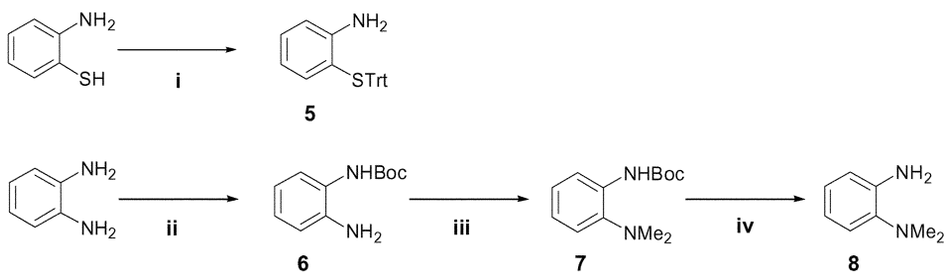


Fig. 1 Representative tetradentate N_2S_2– or N_3S–technetium(V) oxo complexes.

DOI: 10.1039/b313996d



Scheme 1 *ortho* Substituted aniline. *Reagents and conditions*: (i) TrtOH, TFA, CH_2Cl_2, r.t., 1 h, 86%; (ii) BOC-ON, DMF, 55 °C, 16 h, 66%; (iii) 37% aq. CH_2O, $B_{10}H_{14}$, MeOH, r.t., 4 h, 70%; (iv) CF_3COOH, CH_2Cl_2, r.t., 1 h, 68%.

a few examples of such systems have been described in the design of Tc(v) and Re(v) ligands. To our knowledge, only N_2S_2 ligands bearing an aromatic ring in a central position have been developed.[12,13] Our synthetic strategy offers a simple method for including an aromatic cycle in the chelate ring and generating a range of new unsymmetrical N_2S_2, N_2SO and N_3S ligands.

Synthesis and characterisation of ligands

The ligands reported herein have all been synthesised in the thiol-protected form. Triphenylmethyl (trityl) groups were preferred over other sulfur-protected groups (such as benzoyl, ethylethoxyethyl. . .) as they combine a good stability during the ligand synthesis with ease of removal during the ^{99m}Tc radiolabelling reaction.

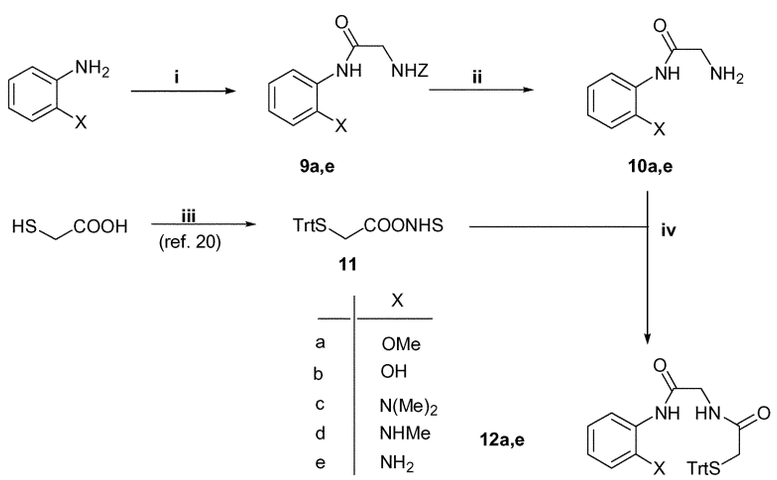
Our different synthetic pathways need commercial or synthesised aromatic diamines as starting materials. To avoid oxidation problems, the thiol function of the 2-aminothiophenol was protected by a trityl group according to the conditions described by Noveron[14] to give **5** in excellent yield. The preparation of **8** follows a three-step procedure as shown in Scheme 1. The monoprotection of the phenylenediamine, by BOC-ON in DMF, gave **6** with 66% yield.[15] The crucial step was the dimethylation of the aromatic nitrogen of **6**. Reductive methylation is known as one of the powerful methods for the methylation of amine. We tried different systems like sodium borohydride–trifluoroacetic acid[16] or sodium borohydride–zinc chloride–paraformaldehyde[17] but with limited success. The best results were obtained by reductive methylation using 37% aqueous formaldehyde and decaborane in methanol.[18] The corresponding tertiary amine **7** was synthesised in 70% yield. Deprotection of the *t*-butyl carbamate by acid hydrolysis yielded the amino precursor **8** in modest yield (after conversion of the amino salt into the free amine).

The ligands were prepared according to two different pathways, depending on the *ortho*-substitution of the aromatic ring. For N_3S or N_2SO ligands, a three-step route was investigated (Scheme 2). The first step involved a conventional carbodiimide amide coupling of *N*-carbobenzyloxyglycine and *ortho* non-sulfur substituted aniline in THF, followed by a classical catalytic hydrogenation using 10% palladium on charcoal as catalyst.[19] The intermediates **10a**–**e** were obtained in excellent yields (70–94% for the two steps). The synthetic key step was the acylation reaction of **10a**–**e** with the activated ester **11**, this latter compound being first prepared in two steps (protection of the thiol functionality with a trityl group followed by activation of the carboxylic acid with *N*-hydroxysuccinimide (NHS) in the presence of EDCI).[20]

While acylation of **10a**–**d** with **11** afforded the corresponding tetradentate ligands **12a**–**d** in 55–88% yield, a mixture of several compounds was obtained for **12e** using the same reaction conditions. To circumvent this problem, we adopted a different synthetic route, carrying out the reaction at room temperature instead of 60 °C and in the presence of Et_3N instead of DMAP. The ligand **12e** was obtained in 76% yield. In these conditions, no condensation between **11** and the aromatic amino group of **10e** was observed.

The preparation of the N_2S_2 ligands followed a four-step procedure as shown in Scheme 3. Peptidic coupling of **11** with ethyl glycinate in DMF, in the presence of Et_3N, gave **13** with an excellent yield (98%). Basic hydrolysis of the ethyl ester of **13** followed by the activation of the acid function of **14** with NHS–DCC afforded the corresponding *N*-hydroxysuccinimidyl ester **15** (83% for the two steps). The final step involved an amide coupling reaction. The compound **15** reacted with the commercial *o*-thiomethoxyaniline or the compound **5** in acetonitrile to give respectively the ligands **12f** (60%) or **12g** (50%).

All the purified ligands were characterised by MS, elemental analysis, proton and carbon NMR spectroscopies with assignments based on 2-dimensional spectra. This kind of ligand exhibits enhanced deshielding for the proton *ortho* to the acylamine group called the "*ortho* effect".[21] *ortho*-Substituents

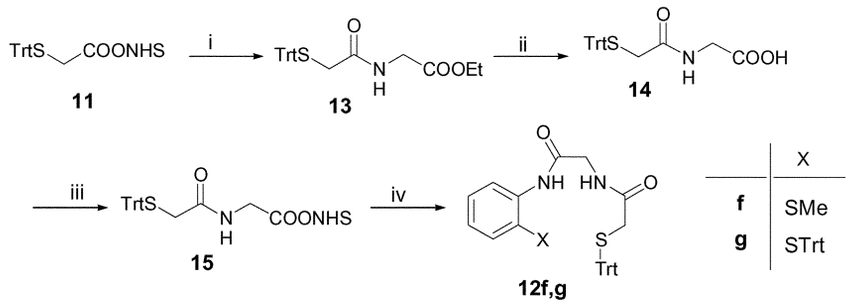


Scheme 2 Synthetic route for ligands **12a**–**e**. *Reagents and conditions*: (i) CbZ-glycine, DCC, 4 h, r.t., THF (71–97%); (ii) H_2, 10% Pd/C, 2 h, r.t., EtOH (81–100%); (iii) (a) TrtOH, AcOH, 2 h, r.t., CH_2Cl_2, 87% (b) EDCI, NHS, 4 h, r.t., ACN, 85%; (iv) **11**, DMAP or Et_3N, 3 h, 60 °C or r.t., ACN (55–88%).

Table 1 Retention time of ^{99m}Tc-complexes

Complex	^{99m}Tc-**12a**	^{99m}Tc-**12b**	^{99m}Tc-**12c**	^{99m}Tc-**12d**	^{99m}Tc-**12e**	^{99m}Tc-**12f**	^{99m}Tc-**12g**
t_r (min)[a]	5.10 5.60	4.98	—	14.10	4.10	6.80 7.19	13.60

[a] MeOH–H_2O–TFA 50 : 50 : 0.1.



Scheme 3 Synthesis of ligands **12f**,**g**. *Reagents and conditions*: (i) HCl·NH_2CH_2COOEt, Et_3N, 3 h, r.t., DMF, 98%; (ii) NaOH, 2 h, 40 °C, MeOH, 100%; (iii) NHS–DCC, 4 h, r.t., THF, 83%; (iv) **5** or *o*-$C_6H_4(SMe)NH_2$, DMAP, 6 h, 60 °C, ACN (50–60%).

capable of hydrogen bonding with the amide hydrogen are expected to stabilise planar conformations in which anisotropic deshielding of the remaining *ortho* hydrogen is maximised and in which the nitrogen, carbonyl carbon, oxygen, *ortho* proton, and ring carbon are locked into a six -membered ring through incipient H-bonding as illustrated in Fig. 3. The H_α shift values are consistent with the values obtained for similar compounds.[22]

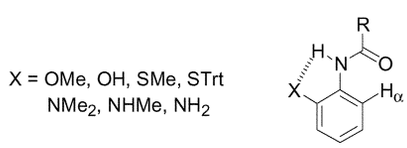


Fig. 3 Hydrogen bonding stabilisation of planar conformation.

^{99m}Tc radiolabelling

Trityl deprotection and ^{99m}Tc-labelling of the pure ligands were performed *in situ* in a methanol–buffer solution pH 8.6 (1 : 4 v/v) by direct reduction of sodium pertechnetate in the presence of an excess of tin chloride at 80 °C during 30 minutes.

For the tetraanionic ligands **12b**,**d**,**e**,**g**, under these conditions, the labelling reaction was quantitative and only negatively charged ^{99m}Tc-product was observed. The two analogous complexes ^{99m}Tc-**12b** and ^{99m}Tc-**12g** were obtained with a large difference in the retention time (Table 1). In a previous communication,[10] we suggest this difference results in the formation of an L_2M species for ^{99m}Tc-**12g**. As reported by Archer *et al.*,[23] it may be that the thiolate sulfurs are slightly better donors than the nitrogen for technetium in this complex. So the 2 : 1 (ligand : metal) complex may form with each ligand bound only through the sulfur atom when the ligand is present in large excess. The ligands **12a**,**12f** did not lead to a single complex as published before[4] but to a mixture of two complexes of technetium in an approximate 2 : 1 ratio. These two products seemed to be the *anti* and *syn* isomers with respect to the technetium oxo and the methyl of the methoxy group. Attempts to prepare a ^{99m}Tc complex with **12c** remained unsuccessful; the latter reaction afforded a mixture of several ^{99m}Tc-complexes.

Purification of ^{99m}Tc-complexes was accomplished by C-18 reverse phase HPLC and resulted in only one component (two isomers in the case of ^{99m}Tc-**12a**,**f**) with an excellent radiochemical yield.

Stability

The radiolabelled chelators were subject to metathesis in the presence of an excess of cysteine as an assay of label stability. After 12 hours (two half-lives of technetium 99m), more than 50% of dissociated ^{99m}Tc was observed for ^{99m}Tc-**12a**,**f** and 40% for ^{99m}Tc-**12g** while ^{99m}Tc-**12e** exhibited a good stability (less than 15% of ^{99m}Tc dissociated) and ^{99m}Tc-**12b**,**d** a great stability (less than 5% of ^{99m}Tc dissociated). The excellent behaviour for the two latter compounds was confirmed by a serum stability study. After 12 h of incubation in fresh human serum, only minor decomposition of the complexes or back oxidation of the metal center to $^{99m}TcO_4^-$ was observed (< 5%). For an eventual application in diagnostic nuclear medicine, this stability would be sufficient.

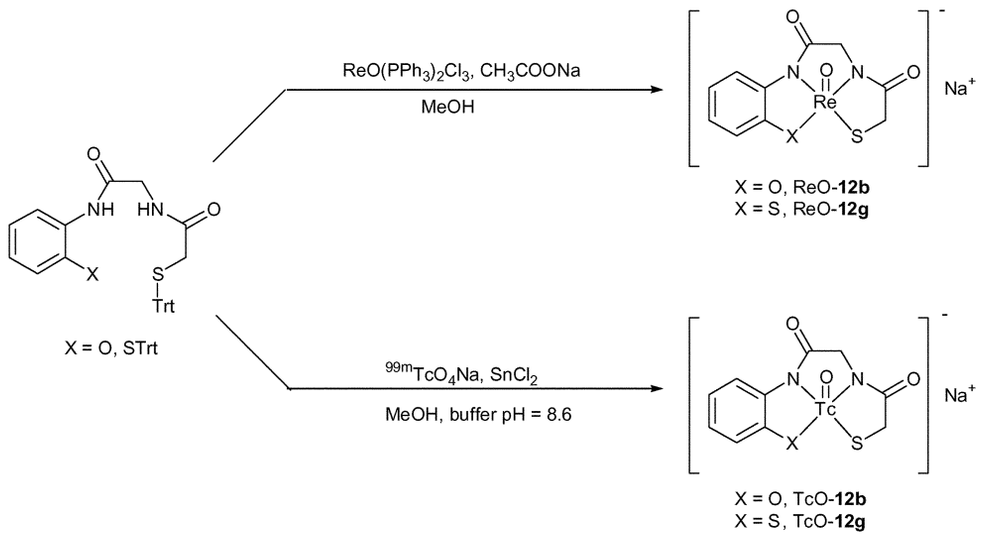
Rhenium chemistry

In order to determine the structure of our ^{99m}Tc-complexes, we synthesised the oxorhenium complexes of **12b** and **12g** using a ligand exchange reaction with $ReO(PPh_3)_2Cl_3$[24] in the presence of sodium acetate (Scheme 4).

The resultant rhenium complexes ReO-**12b** and ReO-**12g** were obtained by silica gel chromatography as the sodium salt respectively in 97% and 83% yield. In the free ligand, the methylene group next to the sulfur appears as a unique singlet (3.21 ppm for **12b** and 3.10 for **12g**). After complexation with $ReO(PPh_3)_2Cl_3$, the singlet splits into two doublets forming the pattern of two AB-spin systems at 3.81 and 4.15 ppm (J = 17.2 Hz) for ReO-**12b** and 3.71 and 3.90 ppm (J = 17.4 Hz) for ReO-**12g**. The two protons next to the chelator amides are also non-equivalent. Negative-ion electrospray of each complex presents two prominent ion peaks with an isotope distribution pattern consistent with the monomeric anion. The complexes were sodium salts as depicted by elemental analysis. The IR spectra showed an absorption at 968 or 964 cm^{-1} indicating the presence of a rhenium–oxo bond.

HPLC comparison by co-injection of the two complexes, ReO-**12b** and the corresponding ^{99m}Tc-**12b**, has shown only a minor difference of retention times (4.50 *versus* 4.98), which could be explained by the different polarities of the $[Tc{=}O]^{3+}$ and $[Re{=}O]^{3+}$ groups[25] (Fig. 4). These results indicate that ligand **12b** forms similar 1 : 1 complexes with $[Tc{=}O]^{3+}$ and $[Re{=}O]^{3+}$ groups.

A short difference of retention time for ^{99m}Tc-**12g** and ReO-**12g** was observed as well (13.60 *versus* 13.00). This surprising result proved that the two complexes are similar. ^{99m}Tc-**12g** seems to be a monomeric complex and not a dimeric structure as suggested previously. The reasons to explain the large difference of retention time between the two metallic complex analogues (Tc or Re) which differ only by one atom (an oxygen



Scheme 4 ReO and TcO complex synthesis.

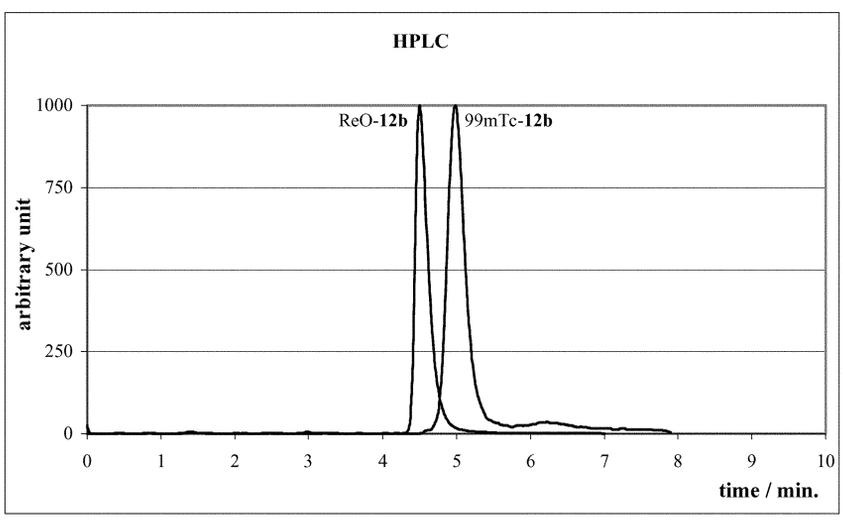


Fig. 4 HPLC comparison of ReO- and TcO-complexes of **12b**.

instead of a sulfur) are not entirely clear yet. To better understand this point and confirm the structure of our metallic complexes, ^{99}Tc coordination chemistry with the two ligands is under investigation.

Conclusion

In an effort to develop efficient ^{99m}Tc-radiopharmaceuticals based on tetradentate ligands, a high yielding route to a new class of semi-rigid tetradentate ligands, with sulfur, amido or amino donor groups has been developed. Their corresponding ^{99m}TcO-complexes were achieved with a good radiochemical yield except for one of them. Tests with technetium 99m (*in vitro* behavior and stability) showed the major influence of the ligand *ortho* substituent on the complex stability. Tetra-anionic ligands formed more stable complexes than trianionic ones with the following order for the *ortho* substituent: OH ~ NHMe > NH_2 > Strt >> OMe, SMe.

In addition, comparative HPLC analysis with prototypic TcO- and ReO-complexes confirms the TcO nature of the radiocomplexes.

Finally, the three ligands **12b**, **12d** and **12e** seem promising for a radiopharmaceutical use because they lead to pure, stable and unique radiocomplexes. Biodistribution studies and coordination chemistry with ^{99g}Tc and $^{185/187}$Re will be addressed in ongoing investigations.

Experimental

All chemicals were of the highest purity commercially available. Solvents were purified by standard methods before use and stored over 0.3 nm molecular sieves. Silica gel (0.060–0.200 nm) was purchased from Acros. TLC was performed using pre-coated Kieselgel 60 plates F_{254} (TLC plates, Merck) and was visualized by UV or iodine. Column chromatography was carried out using "gravity" silica (Merck). NMR spectra were recorded on a Bruker AC 200 (50.323 MHz for ^{13}C and 200.133 MHz for ^{1}H), 250 apparatus (62.896 MHz for ^{13}C and 250.133 MHz for ^{1}H) or 400 (100.63 MHz for ^{13}C and 400.133 MHz for ^{1}H). Chemical shifts are indicated in δ values (ppm) downfield from internal TMS, and coupling constants (J) are given in hertz (Hz). Multiplicities were recorded as s (singlet), d (doublet), t (triplet), q (quadruplet) and m (multiplet). Infrared spectra were recorded as KBr pellets on a Bruker Vector 22 spectrophotometer in the range 4000–400 cm^{-1}. Negative electrospray or DCI-Mass spectra were obtained on a NERMAG R 10–10 mass spectrometer. Microanalysis was performed by the Microanalytical Department of the Ecole Nationale Supérieure de Chimie de Toulouse. HPLC purifications were achieved on a Waters 600E gradient chromatograph with a Waters Lambda Max UV detector, an SAIP radioactivity detector and an ICS dual integrator for effluent monitoring. Radiochemical purity was assessed by thin layer chromato-

graphy on Nano-sil C18 plates (Macherey-Nagel) with an LB 2832 linear analyser (Berthold).

Literature methods were used to prepare *N*-hydroxysuccinimidyl 2-(triphenylmethylthio)ethanoate **11**,[20] monoprotected phenylenediamine **6**[15] and $ReO(PPh_3)_2Cl_3$.[24] Preparation of the intermediate ligands **9b** and **9e**, **10e** has been described previously.[19]

Synthesis of non-commercial *o*-substituted anilines

2-(Triphenylmethylthio)aniline 5. The reagent was prepared following a published procedure[14] using 2-aminothiophenol and triphenylmethanol. A little improvement of yield (86% *vs.* 78%) was obtained and the mass spectra was done.

^{1}H NMR (200 MHz, $CDCl_3$) δ_H(ppm): 3.64 (2H, br s, NH_2); 6.44 (2H, m, ArH); 7.00 (2H, m, ArH); 7.24 (9H, m, ArH_{Trt}); 7.34 (6H, m, ArH_{Trt}); ^{13}C {^{1}H} NMR (50.3 MHz, $CDCl_3$) δ_C(ppm): 70.9 (CPh_3); 115.2, 118.0, 130.8, 137.9 (4 × CH_{Ar}); 116.3, 151.4 (2 × C_{Ar}); 126.8, 127.7, 129.2 (15 × CH_{Trt}); 144.3 (3 × C_{Trt}); MS (DCI/NH_3): 368 ($M+H^+$).

***N*,*N*-Dimethyl-*N′*-(*tert*-butyloxycarbonyl)-1,2-diaminobenzene 7.** A solution of *N*-(*tert*-butyloxycarbonyl)-1,2-diaminobenzene **6** (2.08 g, 10 mmol) and 37% aqueous formaldehyde (1.73 mL, 60 mmol) in MeOH (40 mL) was stirred for 2 hours at room temperature. To the solution was added decaborane (732 mg, 6 mmol) then the resulting solution was stirred for 2 more hours. Solvents were concentrated and the residue was purified by column chromatography on silica gel (eluent: CH_2Cl_2) to give **7** as a pale yellow oil (1.65 g, 70%).

^{1}H NMR (250 MHz, $CDCl_3$) δ_H(ppm): 1.54 (9H, s, CH_3); 2.63 (6H, s, NCH_3); 6.93–7.16 (3H, m, ArH); 7.71 (1H, br s, NH); 8.07 (1H, d, J = 8.2 Hz, ArH); ^{13}C {^{1}H} NMR (62.9 MHz, $CDCl_3$) δ_C(ppm): 28.4 (3 × CH_3); 44.8 (2 × NCH_3); 71.8 (CMe_3); 117.7, 119.9, 122.3, 125.0 (4 × CH_{Ar}); 134.1, 142.3 (2 × C_{Ar}); 153.3 (CO); MS (DCI/NH_3): 237 ($M+H^+$); elemental analysis found C, 66.30; H, 8.82; N, 11.53; $C_{13}H_{20}N_2O_2$ requires C, 66.07; H, 8.53; N, 11.85%.

***N*,*N*-Dimethyl-1,2-diaminobenzene 8.** Trifluoroacetic acid (4.46 mL, 60 mmol) was added to a solution of **7** (1.00 g, 4.23 mmol) in CH_2Cl_2 (3 mL). The mixture was stirred at room temperature for one hour. The solution was concentrated to dryness and the residue was washed with 1 M HCl solution–EtOAc mixture (20 mL, 50 : 50 v/v). The pH of the aqueous layer was raised to 12 and the product was extracted using $CHCl_3$ (2 × 10 mL). The organic layer was dried, concentrated to dryness and the residue was purified by column chromatography on silica gel (eluent: CH_2Cl_2 then CH_2Cl_2–AcOEt 8 : 2) to give **8** as a yellow oil (0.40 g, 68%).

^{1}H NMR (400 MHz, $CDCl_3$) δ_H(ppm): 2.71 (6H, s, NCH_3); 3.98 (2H, br s, NH_2); 6.77 (2H, m, ArH); 6.95 (1H, m, ArH); 7.06 (1H, m, ArH); ^{13}C {^{1}H} NMR (100.6 MHz, $CDCl_3$) δ_C(ppm): 43.9 (2 × NCH_3); 115.4, 118.7, 119.5, 124.4 (4 × CH_{Ar}); 140.9, 141.6 (2 × C_{Ar}); MS (DCI/NH_3): 137 ($M+H^+$).

Synthesis of N_2SO or N_3S tetradentate ligands

N-(*o*-substituted phenyl)-2-[(phenylmethoxy)carbonylamino]-ethanamides

General procedure. To a solution of carbobenzyloxyglycine (2.09 g, 10 mmol) in THF (40 mL) were added *o*-substituted aniline (10 mmol) and a slight excess of dicyclohexylcarbodiimide (2.26 g, 11 mmol). The mixture was stirred under N_2 at room temperature for 6 hours. The insoluble dicyclohexylurea was removed by filtration and the solvent concentrated to dryness. Pure products were obtained after crystallisation from ethyl acetate–petroleum ether 40–60 °C 2 : 1.

***N*-(2-Methoxyphenyl)-2-[(phenylmethoxy)carbonylamino]-ethanamide 9a.** 1.23 g of *o*-anisidine gave **9a** as a white powder (2.95 g, 94%).

^{1}H NMR (250 MHz, $CDCl_3$) δ_H(ppm): 3.83 (3H, s, OCH_3); 4.04 (2H, d, J = 5.1 Hz, NCH_2); 5.16 (2H, s, OCH_2); 5.60 (1H, br s, NH); 6.95 (3H, m, ArH); 7.36 (5H, m, ArH_{Bz}); 8.21 (1H, br s, NH); 8.31 (1H, d, J = 7.9 Hz, ArH); ^{13}C {^{1}H} NMR (50.3 MHz, $CDCl_3$) δ_C(ppm): 45.6 (NCH_2); 55.8 (OCH_3); 67.3 (OCH_2); 110.1, 120.0, 124.3, 128.1 (4 × CH_{Ar}); 121.1, 128.3, 128.6 (3 × $CH_{Ar(Bz)}$); 127.0, 148.1 (2 × C_{Ar}); 136.3 ($C_{Ar(Bz)}$); 156.8, 167.1 (2 × CO); MS (DCI/NH_3): 315 ($M+H^+$), 332 ($M+NH_4^+$); elemental analysis found C, 65.21; H, 5.68; N, 9.05; $C_{17}H_{18}N_2O_4$ requires C, 64.96; H, 5.77; N, 8.91%.

***N*-[2-(*N*,*N*-Dimethylamino)phenyl]-2-[(phenylmethoxy)-carbonylamino]ethanamide 9c.** 1.36 g of **8** gave **9c** as a white powder (2.32 g, 71%).

^{1}H NMR (250 MHz, $CDCl_3$) δ_H(ppm): 2.57 (6H, s, 2 × NCH_3); 4.05 (2H, d, J = 5.8 Hz, NCH_2); 5.16 (2H, s, OCH_2); 5.65 (1H, br s, NH); 7.03–7.18 (3H, m, ArH); 7.37 (5H, m, ArH_{Bz}); 8.32 (1H, d, J = 7.3 Hz, ArH); 8.93 (1H, s, NH); ^{13}C {^{1}H} NMR (100.6 MHz, $CDCl_3$) δ_C(ppm): 44.9 (2 × NCH_3); 45.8 (NCH_2); 67.5 (OCH_2); 119.6, 120.2, 124.4, 125.3 (4 × CH_{Ar}); 132.9, 136.5 (2 × C_{Ar}); 128.4, 128.5, 128.8 (5 × $CH_{Ar(Bz)}$); 143.1 ($C_{Ar(Bz)}$); 167.0, 171.5 (2 × CO); MS (DCI/NH_3): 328 ($M+H^+$); elemental analysis found C, 66.42; H, 6.60; N, 12.72; $C_{18}H_{21}N_3O_3$ requires C, 66.04; H, 6.47; N, 12.84%.

***N*-[2-(*N*-Methylamino)phenyl]-2-[(phenylmethoxy)carbonyl-amino]ethanamide 9d.** 1.13 mL of *N*-methyl-1,2-phenylenediamine gave **9d** as a white powder (3.04 g, 97%).

^{1}H NMR (250 MHz, DMSO d_6) δ_H(ppm): 2.71 (3H, d, J = 5.0 Hz, NCH_3); 3.86 (2H, d, J = 5.9 Hz, NCH_2); 5.07 (3H, br s, NH + OCH_2); 6.58 (2H, m, ArH); 7.10 (2H, m, ArH); 7.38 (5H, m, ArH_{Bz}); 7.56 (1H, br s, NH); 9.13 (1H, s, NH); ^{13}C {^{1}H} NMR (100.6 MHz, DMSO d_6) δ_C(ppm): 30.5 (NCH_3); 44.5 (NCH_2); 66.2 (OCH_2); 110.7, 115.9, 126.7, 127.4 (4 × CH_{Ar}); 123.5, 137.7 (2 × C_{Ar}); 128.4, 128.5, 129.0 (5 × $CH_{Ar(Bz)}$); 144.6 ($C_{Ar(Bz)}$); 157.3, 169.0 (2 × CO); MS (DCI/NH_3): 314 ($M+H^+$) 331 ($M+NH_4^+$); elemental analysis found C, 65.38; H, 6.22; N, 13.09; $C_{17}H_{19}N_3O_3$ requires C, 65.16; H, 6.11; N, 13.41%.

2-Amino-*N*-(*o*-substituted phenyl)ethanamides

General procedure. Catalytic hydrogenation of *N*-(*o*-substituted phenyl)-2-[(phenylmethoxy)carbonylamino]ethanamides (10 mmol) in methanol (100 mL) over 10% Pd/C (20% w/w) was carried out at atmospheric pressure. The mixture was stirred for 2 h, then the catalyst was filtered off through Celite and the solvent was removed under reduced pressure to give the deprotected amine as an oil or a solid.

2-Amino-*N*-(2-methoxyphenyl)ethanamide 10a. 3.14 g of **9a** gave **10a** as white crystals (1.76 g, 98%).

^{1}H NMR (250 MHz, $CDCl_3$) δ_H(ppm): 1.61 (2H, br s, NH_2); 3.50 (2H, s, NCH_2); 3.89 (3H, s, OCH_3); 6.95 (3H, m, ArH); 8.42 (1H, dd, J = 1.8 and 7.8 Hz, ArH); 9.71 (1H, br s, NH); ^{13}C {^{1}H} NMR (50.3 MHz, $CDCl_3$) δ_C(ppm): 45.7 (NCH_2); 55.8 (OCH_3); 110.0, 119.6, 121.0, 123.8 (4 × CH_{Ar}); 127.4, 148.4 (2 × C_{Ar}); 170.9 (CO); MS (DCI/NH_3): 181 ($M+H^+$).

2-Amino-*N*-(2-hydroxyphenyl)ethanamide 10b. This reagent was first prepared by Bermejo *et al.*[19] using cyclohexene as hydrogen source but a quantitative yield was obtained with our method.

3.00 g of **9b** gave **10b** as green powder (1.66 g, 100%).

^{1}H NMR (250 MHz, DMSO d_6) δ_H(ppm): 3.25 (2H, s, NCH_2); 6.75 (1H, m, ArH); 6.87 (2H, m, ArH); 8.17 (1H, d, J = 8.0 Hz, ArH); MS (DCI/NH_3): 167 ($M+H^+$).

2-Amino-*N*-[2-(*N*,*N*-dimethylamino)phenyl]ethanamide 10c. 3.27 g of **9c** gave **10c** as a white powder (1.56 g, 81%).

^{1}H NMR (250 MHz, DMSO d_6) δ_H(ppm): 2.61 (6H, s, 2 × NCH_3); 3.26 (2H, s, NCH_2); 7.02 (2H, m, ArH); 7.19 (1H, m, ArH); 8.28 (1H, m, ArH); ^{13}C {^{1}H} NMR (50.3 MHz, DMSO d_6) δ_C(ppm): 44.6 (2 × NCH_3); 45.8 (NCH_2); 119.2, 120.2, 124.0, 124.4 (4 × CH_{Ar}); 133.0, 144.6 (2 × C_{Ar}); 171.9 (CO); MS (DCI/NH_3): 194 ($M+H^+$).

2-Amino-*N*-[2-(*N*-methylamino)phenyl]ethanamide 10d. 3.13 g of **9d** gave **10d** as a white powder (1.79 g, 100%).

^{1}H NMR (250 MHz, DMSO d_6) δ_H(ppm): 2.70 (3H, d, J = 4.8 Hz, NCH_3); 3.28 (2H, s, NCH_2); 6.60 (2H, m, ArH); 7.05 (1H, m, ArH); 7.24 (1H, m, ArH); ^{13}C {^{1}H} NMR (50.3 MHz, DMSO d_6) δ_C(ppm): 30.6 (NCH_3); 46.0 (NCH_2); 111.1, 116.4, 125.6, 126.9 (4 × CH_{Ar}); 124.3, 144.1 (2 × C_{Ar}); 172.8 (CO); MS (DCI/NH_3): 180 ($M+H^+$).

N-(*o*-substituted phenyl)-2-[(triphenylmethylthio)methylcarbonylamino]ethanamides; general procedure

Method A. To a solution of 2-amino-*N*-(*o*-substituted phenyl)-ethanamide (10 mmol) in acetonitrile (100 mL) were added **11** (4.31 g, 10 mmol) and DMAP (1.22 g, 10 mmol). The solution was heated at 60 °C, under nitrogen, for 3 hours. After cooling, the product was left to precipitate overnight at −4 °C. After filtration, the precipitate was washed with cold acetonitrile to lead to pure crystals.

Method B. As method A except the purification procedure. After cooling, the solution was concentrated to dryness and the crude was purified by column chromatography on silica gel.

Method C. To a solution of 2-amino-*N*-(*o*-substituted phenyl)-ethanamide (10 mmol) in acetonitrile (100 mL) were added **11** (4.31 g, 10 mmol) and Et_3N (1.01 g, 10 mmol). The solution was stirred at room temperature, under nitrogen, for 3 hours. After cooling, the product was left to precipitate overnight at −4 °C. After filtration, the precipitate was washed with cold acetonitrile to lead to pure crystals.

N-(2-Methoxyphenyl)-2-[(triphenylmethylthio)methylcarbonylamino]ethanamide 12a

Method A: 1.80 g of **10a** gave **12a** as pure white crystals (4.36 g, 88%).

^{1}H NMR (250 MHz, $CDCl_3$) δ_H(ppm): 3.20 (2H, s, SCH_2); 3.74 (2H, d, J = 5.5 Hz, CH_2N); 3.82 (3H, s, OCH_3); 6.60 (1H, br s, NH); 6.84–7.10 (3H, m, ArH); 7.20–7.32 (9H, m, ArH_{Trt}); 7.43–7.47 (6H, m, ArH_{Trt}); 8.27 (1H, br s, NH); 8.30 (1H, dd, J = 1.5 and 8.0 Hz, ArH); ^{13}C {^{1}H} NMR (50.3 MHz) δ_C(ppm): 35.7 (CH_2S); 44.5 (CH_2N); 55.7 (OCH_3); 68.0 (CPh_3); 110.1, 119.9, 121.1, 124.2 (4 × CH_{Ar}); 127.2, 128.3, 129.6 (15 × CH_{Trt}); 143.9 (3 × C_{Trt}); 125.0, 148.2 (2 × C_{Ar}); 166.2, 168.9 (2 × CO); ν_{max}/cm^{-1} (KBr): 3409, 3280 (NH) 1643 (CO); *m/z* (DCI NH_3): 497 ($M+H^+$), 514 ($M+NH_4^+$); elemental analysis found C, 72.31; H, 5.94; N, 5.77; $C_{30}H_{28}N_2O_3S$ requires C, 72.55; H, 5.68; N, 5.64%.

N-(2-Hydroxyphenyl)-2-[(triphenylmethylthio)methylcarbonylamino]ethanamide 12b

Method B: after purification by column chromatography on silica gel (eluent: CH_2Cl_2–AcOEt 9 : 1) 1.66 g of **10b** gave **12b** as a pale yellow powder (2.65 g, 55%).

^{1}H NMR (250 MHz, $CDCl_3$) δ_H(ppm): 3.21 (2H, s, SCH_2); 3.76 (2H, d, J = 5.2 Hz, CH_2N); 6.72 (1H, br s, NH); 6.80–7.31 (13H, m, 4ArH + 9ArH_{Trt}); 7.41–7.44 (6H, m, ArH_{Trt}); 8.68 (1H, s, OH); 8.83 (1H, br s, NH); ^{13}C {^{1}H} NMR (62.9 MHz) δ_C(ppm): 35.5 (CH_2S); 44.4 (CH_2N); 68.1 (CPh_3); 118.9, 120.4, 122.3, 126.8 (4 × CH_{Ar}); 125.3, 148.1 (2 × C_{Ar}); 127.2, 128.3, 129.5 (15 × CH_{Trt}); 143.7 (3 × C_{Trt}); 167.6, 169.9 (2 × CO); ν_{max}/cm^{-1} (KBr): 3409, 3347, 3292 (NH), 1700, 1627 (CO); *m/z* (DCI NH_3): 483 ($M+H^+$), 500 ($M+NH_4^+$); elemental analysis found C, 71.90; H, 5.67; N, 5.71; $C_{29}H_{26}N_2O_3S$ requires C, 72.17; H, 5.43; N, 5.80%.

N-[2-(*N*,*N*-Dimethylamino)phenyl]-2-[(triphenylmethylthio)-methylcarbonylamino]ethanamide 12c

Method A: 1.93 g of **10c** gave **12c** as a yellow powder (4.12 g, 81%).

^{1}H NMR (400 MHz, $CDCl_3$) δ_H(ppm): 2.61 (6H, s, NCH_3); 3.22 (2H, s, SCH_2); 3.80 (2H, d, J = 5.3 Hz, CH_2N); 6.65 (1H, br s, NH), 7.10–7.34 (12H, m, 3ArH + 9ArH_{Trt}); 7.46–7.49 (6H, m, ArH_{Trt}); 8.31 (1H, d, J = 8.0 Hz, ArH); 8.75 (1H, br s, NH); ^{13}C {^{1}H} NMR (100.6 MHz) δ_C(ppm): 35.9 (CH_2S); 44.6 (CH_2N); 45.0 (2 × NCH_3); 68.2 (CPh_3); 119.6, 120.3, 124.3, 125.3 (4 × CH_{Ar}); 127.3, 128.4, 129.7 (15 × CH_{Trt}); 133.0, 143.1 (2 × C_{Ar}); 144.0 (3 × C_{Trt}); 166.2, 168.8 (2 × CO); ν_{max}/cm^{-1} (KBr): 3330, 3287 (NH), 1691, 1640 (CO); *m/z* (DCI NH_3): 510 ($M+H^+$), elemental analysis found C, 72.76; H, 6.24; N, 8.04%; $C_{31}H_{31}N_3O_2S$ requires C, 73.05; H, 6.13; N, 8.24%.

N-[2-(*N*-Methylamino)phenyl]-2-[(triphenylmethylthio)methylcarbonylamino]ethanamide 12d

Method A: 1.79 g of **10d** gave **12d** as a grey powder (2.77 g, 56%).

^{1}H NMR (250 MHz, $CDCl_3$) δ_H(ppm): 2.73 (3H, s, NCH_3); 3.15 (2H, s, SCH_2); 3.72 (2H, d, J = 5.2 Hz, CH_2N); 6.69 (2H, m, ArH); 6.80 (1H, br s, NH); 7.11–7.30 (12H, m, NH + 2ArH + 9ArH_{Trt}); 7.40–7.44 (6H, m, ArH_{Trt}); 7.93 (1H, br s, NH); ^{13}C {^{1}H} NMR (62.9 MHz) δ_C(ppm): 30.5 (NCH_3); 35.6 (CH_2S); 44.5 (CH_2N); 67.9 (CPh_3); 111.4, 117.1, 125.3, 127.5 (4 × CH_{Ar}); 127.1, 128.2, 129.5 (15 × CH_{Trt}); 122.5, 143.2 (2 × C_{Ar}); 143.8 (3 × C_{Trt}); 167.2, 169.7 (2 × CO); ν_{max}/cm^{-1} (KBr): 3409, 3390, 3260 (NH), 1673, 1645 (CO); *m/z* (DCI NH_3): 496 ($M+H^+$), elemental analysis found C, 72.61; H, 5.87; N, 8.40; $C_{30}H_{29}N_3O_2S$ requires C, 72.70; H, 5.90; N, 8.48%.

N-(2-Aminophenyl)-2-[(triphenylmethylthio)methylcarbonylamino]ethanamide 12e

Method C: 1.65 g of **10e** gave **12e** as a white powder (3.66 g, 76%).

^{1}H NMR (250 MHz, DMSO d_6) δ_H(ppm): 2.89 (2H, s, CH_2S); 3.80 (2H, d, J = 5.6 Hz, CH_2N); 4.88 (2H, br s, NH_2); 6.52 (1H, m, ArH); 6.70 (1H, m, ArH); 6.91 (1H, m, ArH); 7.07 (1H, m, ArH); 7.22–7.35 (15H, m, ArH_{Tr}); 8.27 (1H, br s, NH); 9.12 (1H, s, NH); ^{13}C {^{1}H} NMR (100.6 MHz, DMSO d_6) δ_C(ppm): 35.8 (SCH_2); 42.7 (NCH_2); 65.9 (CPh_3); 115.4, 115.9, 125.8, 126.2 (4 × CH_{Ar}); 126.7, 128.0, 129.0 (15 × CH_{Trt}); 122.5, 142.4 (2 × C_{Ar}); 143.9 (3 × C_{Trt}); 167.4, 167.8 (2 × CO); ν_{max}/cm^{-1} (KBr): 3454, 3365, 3275 (NH), 1736, 1672 (CO); *m/z* (DCI NH_3): 482 ($M+H^+$), elemental analysis found C, 72.00; H, 5.69; N, 9.02%; $C_{29}H_{27}N_3O_2S$ requires C, 72.32; H, 5.65; N, 8.72%.

Synthesis of N_2S_2 tetradentate ligands

Ethyl 2-[(triphenylmethylthio)methylcarbonylamino]ethanoate 13. A solution of *N*-hydroxysuccinimidyl 2-(triphenylmethylthio)ethanoate **11** (7.46 g, 17.3 mmol) in dry THF (25 mL) was added dropwise, using a syringe, to a stirred solution of ethyl glycinate hydrochloride (2.67 g, 19 mmol) and dry triethylamine (2.67 mL, 19 mmol) in fresh distillated DMF (25 mL). The mixture was stirred at room temperature for 3 h. Solvents were removed under pressure and the residue was washed with 1 M HCl–$CHCl_3$ mixture (100 mL, 50 : 50 v/v). The organic layer was separated, dried over sodium sulfate, filtered off and concentrated to dryness under reduced pressure to give **13** as a pure white powder (7.10 g, 98%).

^{1}H NMR (250 MHz, $CDCl_3$) δ_H(ppm): 1.26 (3H, t, J = 7.0 Hz, CH_3); 3.15 (2H, s, CH_2S); 3.69 (2H, d, J = 5.1 Hz,

CH_2N); 4.18 (2H, q, J = 7.0 Hz, CH_2O); 6.51 (1H, br s, NH); 7.18–7.31 (9H, m, ArH); 7.41–7.44 (6H, m, ArH); ^{13}C {^1H} NMR (50.3 MHz, $CDCl_3$) δ_C(ppm): 14.2 (CH_3); 35.7 (CH_2S); 41.6 (CH_2N); 61.5 (CH_2O); 67.8 (CPh_3); 127.1, 128.2, 129.5 (15 × CH_{Trt}); 143.9 (3 × C_{Trt}); 168.1, 169.4 (2 × CO); MS (DCI/NH_3): 420 ($M+H^+$), 437 ($M+NH_4^+$); elemental analysis found C, 71.86; H, 6.15; $C_{25}H_{25}O_3S$ requires C, 71.57; H, 6.01%.

2-[(Triphenylmethylthio)methylcarbonylamino]ethanoic acid 14. A solution of **13** (10.20 g, 24.3 mmol) in a 1 M NaOH–MeOH mixture (225 mL, 67 : 33, v/v) was boiled at 40 °C for 2 h. The solution was then cooled and acidified with 6 M HCl (15 mL). The obtained white precipitate was filtered off, washed with cooled water and dried under low pressure to give **14** as a white powder (9.85 g, 100%).

^1H NMR (250 MHz, $CDCl_3$) δ_H(ppm): 3.20 (2H, s, CH_2S); 3.68 (2H, d, J = 4.8 Hz, CH_2N); 6.51 (1H, br s, NH); 7.24–7.28 (9H, m, ArH); 7.41–7.45 (6H, m, ArH); 10.49 (1H, br s, OH); ^{13}C {^1H} NMR (50.3 MHz, $CDCl_3$) δ_C(ppm): 35.4 (CH_2S); 41.6 (CH_2N); 67.9 (CPh_3); 127.1, 128.2, 129.5 (15 × CH_{Trt}); 143.9 (3 × C_{Trt}); 169.1, 172.7 (2 × CO); MS (DCI/NH_3): 409 ($M+NH_4^+$), 391 ($M+NH_4^+-H_2O$).

***N*-Hydroxysuccinimidyl 2-[(triphenylmethylthio)methylcarbonylamino]ethanoate 15. 14** (4.00 g, 10.22 mmol) and *N*-hydroxysuccinimide (1.18 g, 10.22 mmol) were dissolved in dry THF (50 mL). Dicyclohexylcarbodiimide (1.32 g, 11.26 mmol) was added and the mixture was stirred at room temperature under nitrogen for 4 h. The reaction mixture was then filtered and the filter cake extracted twice with THF. All filtrates were combined and evaporated to give a solid which was recrystallised from ethyl acetate–petroleum ether (v/v: 2 : 1) to afford **15** as a white powder (4.14 g, 83%).

^1H NMR (250 MHz, $CDCl_3$) δ_H(ppm): 2.81 (4H, s, CH_2); 3.20 (2H, s, CH_2S); 4.01 (2H, d, J = 5.5 Hz, CH_2N); 6.45 (1H, br s, NH); 7.23–7.29 (9H, m, ArH); 7.42–7.44 (6H, m, ArH); ^{13}C {^1H} NMR (50.3 MHz, $CDCl_3$) δ_C(ppm): 25.6 (2 × CH_2); 35.3 (CH_2S); 39.2 (CH_2N); 69.2 (CPh_3); 127.1, 128.3, 129.4 (15 × CH_{Trt}); 143.8 (3 × C_{Trt}); 165.4, 168.3 (2 × CO); 168.5 (2 × CO_{NHS}); MS (DCI/NH_3): 506 ($M+NH_4^+$); elemental analysis found C, 66.50; H, 5.06; N, 5.45; $C_{27}H_{24}N_2O_5S$ requires C, 66.38; H, 4.95; N, 5.73%.

***N*-(2-Thiomethoxyphenyl)-2-[(triphenylmethylthio)methylcarbonylamino]ethanamide 12f.** To a solution of 2-(methylthio)-aniline (1.00 g, 7.18 mmol) in acetonitrile (30 mL) were added **15** (3.51 g, 7.18 mmol) and DMAP (0.88 g, 7.18 mmol). The mixture was heated at 60 °C for six hours then cooled to room temperature and concentrated. Purification by column chromatography on silica gel (eluent: CH_2Cl_2 then CH_2Cl_2–AcOEt: 95 : 5) gave **12f** as a white powder (2.21 g, 60%).

^1H NMR (250 MHz, $CDCl_3$) δ_H(ppm): 2.31 (3H, s, SCH_3); 3.25 (2H, s, SCH_2); 3.75 (2H, d, J = 5.5 Hz, CH_2N); 6.68 (1H, br s, NH); 7.08–7.30 (12H, m, 3ArH + 9ArH_{Trt}); 7.43–7.48 (6H, m, ArH_{Trt}); 8.23 (1H, d, J = 8.0 Hz, ArH); 8.63 (1H, br s, NH); ^{13}C {^1H} NMR (50.3 MHz) δ_C(ppm): 18.9 (SCH_3); 35.7 (CH_2S); 44.6 (CH_2N); 68.0 (CPh_3); 120.8, 124.9, 128.8, 132.8 (4 × CH_{Ar}); 127.2, 128.3, 129.5 (15 × CH_{Trt}); 126.1, 137.6 (2 × C_{Ar}); 143.9 (3 × C_{Trt}); 166.7, 169.0 (2 × CO); ν_{max}/cm^{-1} (KBr): 3356, 3267 (NH), 1699, 1651 (CO); *m*/*z* (DCI NH_3): 513 ($M+H^+$), 530 ($M+NH_4^+$); elemental analysis found C, 69.85; H, 5.59; N, 5.72; $C_{30}H_{28}N_2O_2S_2$ requires C, 70.28; H, 5.50; N, 5.46%.

***N*-[2-(Triphenylmethylthio)phenyl]-2-[(triphenylmethylthio)-methylcarbonylamino]ethanamide 12g.** The same procedure described for compound **12f** was used (with **5** instead of 2-(methylthio)aniline as starting material). **12g** was obtained as a white powder (2.60 g, 50%).

^1H NMR (250 MHz, $CDCl_3$) δ_H(ppm): 3.10 (2H, s, SCH_2); 3.29 (2H, d, J = 5.2 Hz, CH_2N); 6.42 (1H, br s, NH); 6.79 (1H, m, ArH); 7.22–7.29 (26H, m, ArH); 7.42–7.45 (6H, m, ArH); 8.09 (1H, br s, NH); 8.17 (1H, d, J = 7.5 Hz, ArH); ^{13}C {^1H} NMR (50.3 MHz) δ_C(ppm): 35.5 (CH_2S); 43.7 (CH_2N); 67.8, 71.7 (2 × CPh_3); 119.4, 123.6, 131.2, 137.4 (4 × CH_{Ar}); 127.1, 127.2, 127.9, 128.3, 129.5, 129.6 (30 × CH_{Trt}); 120.4, 141.5 (2 × C_{Ar}); 143.6, 143.9 (6 × C_{Trt}); 165.5, 168.4 (2 × CO); ν_{max}/cm^{-1} (KBr): 3325 (NH), 1679 (CO); *m*/*z* (DCI NH_3): 741 ($M+H^+$), 758 ($M+NH_4^+$); elemental analysis found C, 77.00; H, 5.32; N, 3.50; $C_{48}H_{40}N_2O_2S_2$ requires C, 77.80; H, 5.44; N, 3.78%.

Rhenium complex synthesis

***N*-(2-Hydroxyphenyl)-2-(thiomethylcarbonylamino)ethanamide oxorhenate(v), sodium salt: ReO-12b.** To **12b** (144.6 mg, 0.3 mmol) and sodium acetate (163.2 mg, 1.2 mmol) dissolved in 40 mL of dry methanol, was added $ReO(PPh_3)_2Cl_3$ (324.5 mg, 0.39 mmol). After refluxing for 4 hours, the solution turned brown and clear. After cooling, the solution was filtered and evaporated to dryness. The residue was purified by column chromatography on silica gel (eluent: CH_2Cl_2–MeOH: 9 : 1) to yield the complex as an orange-red powder (134.2 mg, 97%).

^1H NMR (400 MHz, DMSO d_6) δ_H(ppm): 3.81 (1H, d, J = 17.2 Hz, CH_2S); 4.15 (1H, d, J = 17.2 Hz, CH_2S); 4.54 (1H, d, J = 18.3 Hz, CH_2N); 5.46 (1H, d, J = 18.3 Hz, CH_2N); 6.78 (1H, t, J = 7.7 Hz, CH_{Ar}); 6.90 (1H, t, J = 7.7 Hz, CH_{Ar}); 7.06 (1H, d, J = 7.7 Hz, CH_{Ar}); 8.25 (1H, d, J = 7.9 Hz, CH_{Ar}); ^{13}C {^1H} NMR (100.63 MHz; MeOD) δ_C(ppm): 40.3 (1C, CH_2S); 61.0 (1C, CH_2N); 115.1, 118.1, 119.5, 123.4 (4C, CH_{Ar}); 140.2 (1C, C_{Ar}); 169.5 (1C, C_{Ar}); 187.3, 194.5 (2C, C=O); ν_{max}/cm^{-1} (KBr): 1624 (CO) 968 (ReO); *m*/*z* (ES^-): 437 (60), 439 (100) [M^-]; elemental analysis found C, 25.74; H, 2.06; N, 5.62; $C_{10}H_8N_2O_4SReNa$ requires C, 26.00; H, 1.73; N, 6.07%.

***N*-(2-Thiophenyl)-2-(thiomethylcarbonylamino)ethanamide oxorhenate(v), sodium salt: ReO-12g.** The same procedure described for compound ReO-**12b** was used (with **12g** instead of **12b**). After purification by column chromatography on silica gel (eluent: CH_2Cl_2 then CH_2Cl_2–MeOH: 9 : 1) ReO-**12g** was obtained as an orange-red powder (197.5 mg, 83%).

^1H NMR (300 MHz, DMSO d_6) δ_H(ppm): 3.71 (1H, d, J = 17.4 Hz, CH_2S); 3.90 (1H, d, J = 17.4 Hz, CH_2S); 4.54 (1H, d, J = 18.9 Hz, CH_2N); 4.94 (1H, d, J = 18.9 Hz, CH_2N); 6.88 (2H, m, CH_{Ar}); 7.54 (1H, m, CH_{Ar}); 8.72 (1H, m, CH_{Ar}); ^{13}C {^1H} NMR (75.5 MHz; MeOD) δ_C(ppm): 40.7 (1C, CH_2S); 61.2 (1C, CH_2N); 126.1, 124.7, 125.3, 129.1 (4C, CH_{Ar}); 148.8 (1C, C_{Ar}); 152.6 (1C, C_{Ar}); 190.5, 194.8 (2C, C=O); ν_{max}/cm^{-1} (KBr): 1620 (CO), 964 (ReO); *m*/*z* (ES^-): 453 (60), 455 (100) [M^-]; elemental analysis found C, 25.08; H, 2.10; N, 5.48; $C_{10}H_8N_2O_3S_2ReNa$ requires C, 25.10; H, 1.67; N, 5.85%.

^{99m}Tc labelling. Into a borosilicated vial containing buffer (pH = 8.6; 200 µl), were added a solution of ligand in methanol (1 mg ml^{-1}, 100 µl) and $Na^{99m}TcO_4$ generator eluate (100 µl, 2 mCi). After addition of a fresh $SnCl_2$ solution in MeOH (75 µl, 2.25 mg ml^{-1}), the vial was sealed with a Teflon-lined cap and the mixture was heated at 80 °C for 30 minutes. After cooling, the resulting complexes were purified by HPLC, using a Satisfaction RP18AB column (eluent: MeOH–H_2O–TFA 50 : 50 : 0.1) at a flow rate of 1 ml min^{-1}.

Stability *versus* cysteine. The complex after purification by HPLC under the conditions described above was incubated at 37 °C, under a nitrogen atmosphere with a freshly prepared solution of L-cysteine at 1 mg ml^{-1} in a 500 : 1 cysteine/complex molar ratio. All solutions were purged with nitrogen prior to use. Incubate aliquots at 2, 6 and 12 hours intervals were analyzed by thin layer chromatography on nano-sil C18 plates by elution with MeOH–CH_3CN–H_2O–TFA 20 : 15 : 65 : 0.1. R_f (free technetium) = 1; R_f (^{99m}Tc-complex) = between 0.25 and 0.60.

Acknowledgements

We are grateful to the French Ministère de l'Education Nationale, de la Recherche et de la Technologie.

References

1 (*a*) J. R. Dilworth and S. J. Parrot, *Chem. Soc. Rev.*, 1998, **27**, 43–55; (*b*) K. Schwochau, *Angew. Chem., Int. Ed. Engl.*, 1994, **33**, 2258–2267.
2 (*a*) F. Mevellec, A. Roucoux, N. Noiret, A. Moisan, H. Patin and A. Duatti, *J. Labelled Compd. Radiopharm.*, 2003, **46**, 319–331; (*b*) F. Mevellec, F. Tisato, F. Refosco, A. Roucoux, N. Noiret, H. Patin and G. Bandoli, *Inorg. Chem.*, 2002, **41**, 598–601.
3 B. J. Cleynhens, G. M. Bormans, H. P. Vanbilloen, D. V. Vanderghinste, D. M. Kieffer, T. J. de Groot and A. M. Verbruggen, *Tetrahedron Lett.*, 2003, **44**, 2597–2600.
4 L. Fourteau, E. Benoist and M. Dartiguenave, *Synlett*, 2001, **1**, 126–128.
5 S. Jurisson and J. D. Lydon, *Chem. Rev.*, 1999, **99**, 2205–2218 and references therein.
6 A. Davison, A. G. Jones, C. Orvig and M. Sohn, *Inorg. Chem.*, 1981, **20**, 1629–1632.
7 R. Rajagopolan, G. D. Grummon, J. Bugaj, L. S. Halleman, E. G. Webb, M. E. Marmion, J. L. Vanderheyden and A. Srinivasan, *Bioconjugate Chem.*, 1997, **8**, 407–415.
8 O'J. P. Neil, S. R. Wilson and J. A. Katzenellenbogen, *Inorg. Chem.*, 1994, **33**, 319–323.
9 A. Loussouarn, M. Duflos, E. Benoist, J.-F. Chatal, G. Le Baut and J.-F. Gestin, *J. Chem. Soc., Perkin Trans. 1*, 1998, 237.
10 J. Le Gal, E. Benoist, M. Gressier, Y. Coulais and M. Dartiguenave, *Tetrahedron Lett.*, 2002, **43**, 9295–9297.
11 (*a*) A. S. De Sousa, G. J. B. Croft, C. A. Wagner, J. P. Mickael and R. D. Hancock, *Inorg. Chem.*, 1991, **30**, 3525–3529; (*b*) A. S. De Sousa and R. D. Hancock, *J. Chem. Soc., Chem. Commun.*, 1995, 415–416.
12 S. Kasina, A. R. Fritzberg, D. L. Johnson and D. Eshima, *J. Med. Chem.*, 1986, **29**, 1933–1940.
13 (*a*) B. J. McBride, R. M. Baldwin, J. M. Kerr, J. L. Wu, L. M. Schultze, N. E. Salazar, J. M. Chinitz and E. F. Byrne, *J. Med. Chem.*, 1993, **36**, 81–86; (*b*) S. Liu, D. S. Edwards, in *Technetium, Rhenium and Other Metals in Chemistry and Nuclear Medicine Vol. 4*, Eds M. Nicolini, G. Bandoli, U. Mazzi, SGEditoriali, Padova, Italy, 1994, pp. 383–393.
14 J. C. Noveron, M. M. Olmstead and P. K. Mascharak, *J. Am. Chem. Soc.*, 2001, **123**, 3242–3259.
15 C. T. Seto, J. P. Mathias and G. M. Whitesides, *J. Am. Chem. Soc.*, 1993, **115**, 1321–1329.
16 G. W. Gribble and C. F. Nutaitis, *Synthesis*, 1987, 709–711.
17 S. Bhattacharyya, *Synth. Commun.*, 1995, **25**, 2061–2069.
18 Y. J. Jung, J. W. Bae, C. O. M. Yoon, B. W. Yoo and C. M. Yoon, *Synth. Commun.*, 2001, **31**, 3417–3421.
19 (*a*) M. R. Bermejo, A. M. Gonzalez, M. Fondo, A. Garcia-Deibe, M. Maneiro, J. Sanmartin, O. L. Hoyos and M. Watkinson, *New J. Chem.*, 2000, **24**, 235–241; (*b*) O. L. Hoyos, M. R. Bermejo, M. Fondo, A. Garcia-Deibe, A. M. Gonzalez, M. Maneiro and R. Pedrido, *J. Chem. Soc., Dalton Trans.*, 2000, 3122–3127.
20 R. A. Bell, D. W. Hughes, C. J. L. Lock and J. F. Valliant, *Can. J. Chem.*, 1996, **74**, 1503–1511.
21 M. Zanger, W. W. Simons and A. R. Gennaro, *J. Org. Chem.*, 1968, **33**, 3673–3675.
22 J. P. Hagemann and P. T. Kaye, *Synth. Commun.*, 1999, **29**, 303–310.
23 C. M. Archer, J. R. Dilworth, D. Vaughan Griffiths, Al-M. J. Jeboori, J. Duncan Kelly, C. Lu, M. J. Rosser and Y. Zheng, *J. Chem. Soc., Dalton Trans.*, 1997, 1403–1410.
24 J. Chatt and G. A. Rowe, *J. Chem. Soc.*, 1962, 4019–4033.
25 C. S. Hilger, B. Noll, F. Blume, P. Leibnitz, B. Johannsen, in *Technetium, Rhenium and Other Metals in Chemistry and Nuclear Medicine Vol. 5*, Eds M. Nicolini, U. Mazzi, SGEditoriali, Padova, Italy, 1999, pp. 221–224.