Inorg. Chem. **2005**, *44*, 6763–6770

Thiol- and Thioether-Based Bifunctional Chelates for the $\{M(CO)_3\}^+$ Core (M = Tc, Re)

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Received May 10, 2005

By analogy to the recently described single amino acid chelate (SAAC) technology for complexation of the $\{M(CO)_3\}^+$ core (M = Tc, Re), a series of tridentate ligands containing thiolate and thioether groups, as well as amino and pyridyl nitrogen donors, have been prepared: $(NC_5H_4CH_2)_2NCH_2CH_2SEt$ (L1); $(NC_5H_4CH_2)_2NCH_2CH_2SH$ (L2); $NC_5H_4CH_2N(CH_2CH_2SH)_2$ (L3); $(NC_5H_4CH_2)N(CH_2CH_2SH)(CH_2CO_2R)$ [R = H (L4); R = $-C_2H_5$ (L5). The $\{Re(CO)_3\}^+$ core complexes of L1–L5 were prepared by the reaction of $[Re(CO)_3(H_2O)_3]Br$ or $[NEt_4]_2[Re(CO)_3Br_3]$ with the appropriate ligand in methanol and characterized by infrared spectroscopy, ^1H and ^{13}C NMR spectroscopy, mass spectrometry, and in the case of $[Re(CO)_3(L2)]$ (**Re-2**) and $[Re(CO)_3(L1)Re(CO)_3Br_2]$ (**Re-1a**) by X-ray crystallography. The structure of **Re-2** consists of discrete neutral monomers with a *fac*-$Re(CO)_3$ coordination unit and the remaining coordination sites occupied by the amine, pyridyl, and thiolate donors of L2, leaving a pendant pyridyl arm. In contrast, the structure of **Re-1a** consists of discrete binuclear units, constructed from a $\{Re(CO)_3(L1)\}^+$ subunit linked to a $\{Re(CO)_3Br_2\}^-$ group through the sulfur donor of the pendant thioether arm. The series of complexes establishes that thiolate donors are effective ligands for the $\{M(CO)_3\}^+$ core and that a qualitative ordering of the coordination preferences of the core may be proposed: pyridyl nitrogen ~ thiolate > carboxylate > thioether sulfur > thiophene sulfur. The ligands L1 and L2 react cleanly with $[^{99m}Tc(CO)_3(H_2O)_3]^+$ in H_2O/DMSO to give $[^{99m}Tc(CO)_3(L1)]^+$ (**^{99m}Tc-1**) and $[^{99m}Tc(CO)_3(L2)]$ (**^{99m}Tc-2**), respectively, in ca. 90% yield after HPLC purification. The Tc analogues **^{99m}Tc-1** and **^{99m}Tc-2** were subjected to ligand challenges by incubating each in the presence of 1000-fold excesses of both cysteine and histidine. The radiochromatograms showed greater than 95% recovery of the complexes.

Introduction

The significant contemporary interest in the chemistry of technetium and rhenium reflects the importance of the radioisotopes ^{99m}Tc and $^{186,188}Re$ in the development of diagnostic and therapeutic radiopharmaceuticals, respectively.[1−5] The established methods for radiolabeling of target-specific biomolecules with ^{99m}Tc or ^{188}Re exploit the coordination preferences of the metals in specific oxidation states and ligand environments. The organometallic complex $[M(H_2O)_3(CO)_3]^+$ (M = Re or Tc)[6,7] has recently attracted significant attention in radiopharmaceutical chemistry for several reasons: (i) The small size of the $\{M(CO)_3\}^+$ core allows labeling of low molecular weight biomolecules with high specific activites with retention of biological activity

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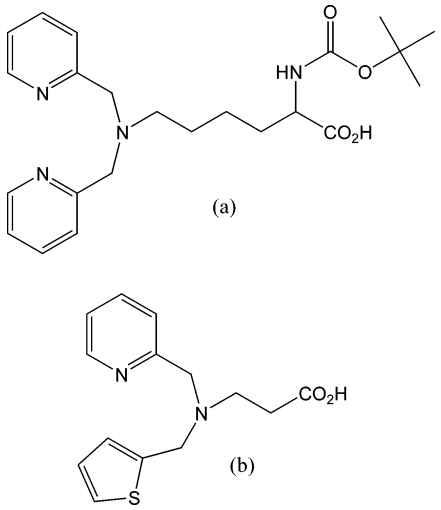
(1) (a) Nicolini, M., Bandoli, G., Mazzi, U., Eds. *Technetium, Rhenium and Other Metals in Chemistry and Nuclear Medicine*; Servici Grafici Editoriali: Padova, 2000; Vol. 5. (b) Nicolini, M., Bandoli, G., Mazzi, U., Eds. *Technetium and Rhenium in Chemistry and Nuclear Medicine*; Raven Press: New York, 1990. (c) Jurisson, S. S.; Lyden, J. D. *Chem. Rev.* **1999**, *99*, 2205. (d) Liu, S.; Edwards, D. S. *Chem. Rev.* **1999**, *99*, 2235. (e) Dilworth, J. R.; Parrott, S. J. *Chem. Soc. Rev.* **1998**, *27*, 43. (f) Hom, R. K.; Katzenellenbogen, J. A. *Nucl. Med. Biol.* **1997**, *24*, 485. (g) Eckelman, W. C. *Eur. J. Nucl. Med.* **1995**, *22*, 249. (h) Schwochan, K. *Angew. Chem., Int. Ed. Engl.* **1994**, *33*, 2258.
(2) (a) Blower, P. J.; Prakash, S. *Perspect. Bioinorg. Chem.* **1999**, *4*, 91. (b) Volkert, W. A.; Hoffman, T. J. *Chem. Rev.* **1999**, *99*, 2269. (c) Palmedo, H.; Guhlke, S.; Bender, H.; Sartor, J.; Schoeneich, G.; Risse, J.; Grunwalk, F.; Knapp, F. F., Jr.; Biersack, H. J. *Eur. J. Nucl. Med.* **2000**, *27*, 123.
(3) Alberto, R.; Abram, U. *Handb. Nucl. Chem.* **2003**, *4*, 211.

10.1021/ic050735a CCC: $30.25
Published on Web 08/17/2005

andspecificity. (ii) The core has a high affinity for a large variety of donor atoms. (iii) The precursor complexes for the radioconjugates, *fac*-$[M(CO)_3(H_2O)_3]^+$, may be readily prepared in aqueous based kit formulations.[8] (iv) The water molecules of the precursor complex are readily substituted by a variety of functional groups, including amines, imines, thioethers, thiols, and phosphines. (v) The *fac*-$\{M(CO)_3\}^+$ core is chemically robust, low-spin d^6, providing a convenient platform for drug development.

We have reported on a series of bifunctional single amino acid chelates (SAAC) for labeling of biomolecules with the $\{Tc(CO)_3\}^+$ and $\{Re(CO)_3\}^+$ cores.[9−15] These amino acid analogues incorporate a tridentate donor set[16] for chelation and an amino acid functionality for attachment to biomolecules. Furthermore, the SAACs can be readily incorporated into peptides using conventional solid phase synthesis. The approach also affords significant flexibility in the choice of donors for Tc and Re coordination. Initially, our efforts focused on the design of tridentate chelates with aromatic amines such as pyridine, imidazole, and quinoline on the arms of the chelate, radiating from the central aliphatic nitrogen residue. Subsequently, we developed less sterically demanding aliphatic amine chelators,[17] as well as bifunctional chelates with mixed aromatic and aliphatic arms projecting from the bridge-head nitrogen of the amino acid or amino acid analogue (Chart 1).

Chart 1. Representative Amino Acid Chelates (SAAC) Based On Natural Amino Acids, (a) *N*-α-(*tert*-Butoxycarbonyl)-*N*-ω-bis(2-pyridylmethyl)-L-lysine, and on Synthetic Amino Acids, (b) *N*-(Pyridylmethyl)-*N*-(2-thiophenemethyl)aminopropionic Acid



These and related studies on the coordination chemistry of the *fac*-$\{M(CO)_3\}^+$ core have established that tridentate ligands with amine, N-heterocyclic, and carboxylate donors are effective in forming robust complexes, as well as exhibiting improved clearance characteristics in vivo compared to the mono- or bidentate ligands.[18] Although chelates incorporating thiolate groups would offer considerable versatility in the design of complexes with neutral, anionic, or cationic charge, depending on the identity of the donor sets, the coordination chemistry of the $\{M(CO)_3\}^+$ core with tridentate chelating reagents incorporating thiolate or thioether donors remains relatively unexplored.[15,19]

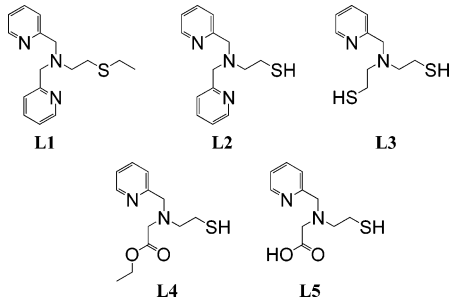
Consequently, the preparations of a series of sulfur-containing tridentate ligands and their complexation with the $\{Re(CO)_3\}^+$ core were undertaken. The N, S donor ligands of this study satisfy the following conditions: (i) The donor groups are appropriately oriented to provide *fac*-coordination in the $\{Re(CO)_3L\}^{n+}$ complex. (ii) The ligand set is sufficiently versatile to produce different overall charges on the complex. (iii) The ligands may be modified to provide a linker to a biomolecule. The preparations and characterizations of the ligands shown in Chart 2 are discussed, as well as the structures of $[Re(CO)_3\{(2\text{-pyridyl-}CH_2)_2N(CH_2)_2S)]$ and $[Re(CO)_3\{(2\text{-pyridyl})(CH_2)_2N(CH_2)_2SEt\}Re(CO)_3Br_2]$.

Experimental Section

General Methods. All reagents and organic solvents were ACS grade or higher and used without further purification. $[Re(CO)_3(H_2O)_3]Br$[20] and $[NEt_4]_2[ReBr_3(CO)_3]$[8] were prepared according to the literature methods. ^1H and ^{13}C NMR spectra were recorded on

(4) Schibli, R.; Schubiger, P. A. *Eur. J. Nucl. Med.* **2002**, *29*, 1529.
(5) Alberto, R. In *Comprehensive Coordination Chemistry II*; McCleverty, J., Meyer, T., Eds.; Elsevier: Oxford, U.K., 2004; Vol. 5, p 127.
(6) (a) Alberto, R.; Schibli, R.; Angst, D.; Schubiger, P. A.; Abram, U.; Abram, S.; Kaden, T. L. A. *Transition Met. Chem.* **1997**, *22*, 597. (b) Alberto, R.; Schibli, R.; Egli, A.; Schubiger, P. A. *J. Am. Chem. Soc.* **1998**, *120*, 7987. (c) Alberto, R.; Schibli, R.; Schubiger, P. A. *J. Am. Chem. Soc.* **1999**, *121*, 6076. (d) Waibei, R.; Alberto, R.; Willuda, J.; Finnern, R.; Schibli, R. Stichelberger, A.; Egli, A.; Abram, U.; Mach, J. P.; Pluckthun, A.; Schubiger, P. A. *Nat. Biotechnol.* **1999**, *17*, 897.
(7) (a) Amann, A.; Decristoforo, C.; Ott, I.; Wenger, M.; Bader, D.; Alberto, R.; Putz, G. *Nucl. Med. Biol.* **2001**, *28*, 243. (b) Schibli, R.; LaBella, R.; Alberto, R.; Garcia-Garayoa, L.; Ortner, K.; Abram, U.; Schubiger, P. A. *Bioconjugate Chem.* **2000**, *11*, 345. (c) Wald, J.; Alberto, R.; Ortner, K.; Candreia, L. *Angew. Chem., Int. Ed.* **2001**, *40*, 3062. (d) Petrig, R.; Schibli, R.; Dumas, C.; Alberto, R.; Schubiger, P. A. *Chem.*—Eur. J. **2001**, *7*, 1868.
(8) Alberto, R.; Egli, A.; Hegetschweiler, K.; Gramlich, V.; Schubiger, P.A. *J. Chem. Soc., Dalton Trans.* **1994**, 2815.
(9) Alberto, R.; Ortner, K.; Wheatley, N.; Schibli, R.; Schubiger, A. P. *J. Am. Chem. Soc.* **2001**, *123*, 3135−3136.
(10) Banerjee, S. R.; Levadala, M. K.; Lazarova, N.; Wei, L.; Valliant, J. F.; Stephenson, K. A.; Babich, J. W.; Maresca, K. P.; Zubieta, J. *Inorg. Chem.* **2002**, *41*, 6417.
(11) Stephenson, K. A.; Valliant, J. F.; Zubieta, J.; Banerjee, S. R.; Levadala, M. K.; Taggart, L.; Ryan, L.; MaFarlane, N.; Boreham, D. R.; Babich, J. W.; Maresca, K. P. Presented at the 50th Annual SNM Meeting at New Orleans, June 22−25, No. 154; *J. Nucl. Med.* **2003**, *44*, 48 p.
(12) Stephenson, K. A.; Zubieta, J.; Banerjee, S. R.; Levadala, M. K.; Taggart, L.; Ryan, L.; McFarlane, N.; Boreham, D. R.; Maresca, K. P.; Babich, J. W.; Valliant, J. F. *Bioconjugate Chem.* **2004**, *15*, 128.
(13) Levadala, M. K.; Banerjee, S. R.; Maresca, K. P.; Babich, J. W.; Zubieta, J. *Synthesis* **2004**, 1759.
(14) Stephenson, K. A.; Banerjee, S. R.; Besanger, T.; Sogbein, O. O.; Levadala, M. K.; McFarlane, N.; Lemon, J. A.; Boreham, D. R.; Maresca, K. P.; Brennan, J. D.; Babich, J. W.; Zubieta, J.; Valliant, J. F. *J. Am. Chem. Soc.* **2004**, *126*, 8598.
(15) Banerjee, S. R.; Francesconi, L.; Valliant, J. F.; Babich, J. W.; Zubieta, J. *Nucl. Med. Biol.* **2005**, *32*, 1.
(16) Schibli, R.; La Bella, R.; Alberto, R.; Garcia-Garayoa, E.; Ortner, K.; Abram, U.; Schubiger, P. A. *Bioconjugate Chem.* **2000**, *11*, 345.
(17) Banerjee, S. R.; Babich, J. W.; Zubieta, J. *Inorg. Chem. Commun.* **2004**, *7*, 481.
(18) (a) Schibli, R.; Alberto, R.; Abram, U.; Abram, S.; Egli, A.; Schubiger, P. A.; Kaden, T. A. *Inorg. Chem.* **1998**, *37*, 3509. (b) Pietzsch, H.-J.; Gupta, A.; Reigys, M.; Drew, A.; Seibert, S.; Sephie, R.; Spies, H.; Alberto, R.; Abram, U.; Schubiger, P. A.; Johannsen, B. *Bioconjugate Chem.* **2000**, *11*, 414. (c) Alves, S.; Paula, A.; Correia, J. D. G.; Domingos, A.; Santos, I. *J. Chem. Soc., Dalton Trans.* **2002**, 4714.
(19) Kramer, D. J.; Davison, A.; Davis, W. M.; Jones, A. G. *Inorg. Chem.* **2002**, *41*, 6181−6183.
(20) Lazarova, N.; James, S.; Babich, J.; Zubieta, J. *Inorg. Chem. Commun.* **2004**, *7*, 1023−1026.

Chart 2



a Bruker DPX 300 spectrometer. Proton chemical shifts are expressed as parts per million using tetramethylsilane or deuterated solvent as a reference standard. IR spectra were recorded as either KBr pallets or as solutions in methanol and run on a Perkin-Elmer Paragon 1000 FT-IR spectrophotometer. HRMS data were performed on a 3 T Fourier transform mass spectrometer with a ESI source in positive ion mode. All sulfur-containing materials were handled and stored under an atmosphere of argon. Most reactions were performed under argon atmosphere by using standard Schlenk techniques.

Ligand Syntheses. *N,N*-Bis(2-pyridylmethyl)((aminoethyl)-thio)ethane, L1. Sodium hydroxide (0.56 g, 14.12 mmol) was added to 2-(ethylthio)ethylamine (2 g, 14.12 mmol) in a mixture of methanol (5 mL) and 1,2-dichloroethane (20 mL). 2-Pyridinecarboxaldehyde (3.02 g, 28.24 mmol) was introduced to the reaction mixture followed by the addition of sodium triacetoxyborohydride (6.88 g, 32.47 mmol). After the mixture was stirred under Ar for 20 h, the crude product was purified by silica gel column chromatography using methanol−chloroform (3:97) to yield L1 as a foul-smelling dark green oil (3.524 g, 12.28 mmol, 87%). ^{1}H NMR (δ (ppm), $CDCl_3$): 8.50 (d, $J = 5.06$ Hz, 2H, PyH), 7.65 (t, $J = 8.23$ Hz, 2H, PyH), 7.56 (d, $J = 8.23$ Hz, 2H, PyH), 7.13 (t, $J = 6.96$ Hz, 2H, PyH), 3.85 (s, 4H, $PyCH_2$), 2.79 (m, 2H, NCH_2), 2.69 (m, 2H, CH_2S), 2.43 (q, $J = 7.60$ Hz, 2H, SCH_2), 1.17 (t, $J = 6.96$ Hz, 3H, CH_3). ^{13}C NMR (δ (ppm), $CDCl_3$): 160.15 (2C, Py), 148.29 (2CH, Py), 133.81 (2CH, Py), 121.13 (2CH, Py), 119.99 (2CH, Py), 61.22 (2C, CH_2Py), 54.22 (C, NCH_2), 37.45 (C, CH_2SH), 24.03 (C, SCH_2), 16.24 (C, CH_3). Anal. Calcd (found) for $C_{16}H_{21}N_3S$: C, 66.9 (66.8); H, 7.32 (7.41); N, 14.6 (14.4).

***N,N*-Bis(2-pyridylmethyl)aminoethanethiol, L2.** The preparation of L2 was based on a literature procedure[21] with some modifications. 2,2′-Dipicolylamine (5 g, 25.10 mmol) in benzene (5 mL) was placed in a Schlenk tube flushed with Ar. Ethylene sulfide (2.99 mL, 50.2 mmol) in benzene (5 mL) was added dropwise and the resulting solution stirred at 65 °C. After 48 h, analysis by ^{1}H and ^{13}C NMR spectroscopy confirmed that the reaction had gone to completion. The bulk solution was filtered and the excess solvent removed in vacuo to yield a foul-smelling yellow oil. The oil was redissolved in dicholoromethane, the solution filtered through a plug of silica to remove polymeric impurities, and the excess solvent removed in vacuo to yield the pure product as a yellow oil (5.79 g, 22.3 mmol, 89%). ^{1}H NMR (δ (ppm), $CDCl_3$): 8.47 (d, $J = 4.90$ Hz, 2H, PyH), 7.61 (t, $J = 7.47$ Hz, 2H, PyH), 7.51 (d, $J = 7.47$ Hz, 2H, PyH), 7.11 (t, $J = 6.23$ Hz, 2H, PyH), 3.85 (s, 4H, $PyCH_2$), 2.80 ($J = 7.47$ Hz, NCH_2), 2.63 (t, $J = 6.85$ Hz, CH_2SH). ^{13}C NMR (δ (ppm), $CDCl_3$): 159.26 (2C, Py), 149.07 (2CH, Py), 136.79 (2CH, Py), 123.43 (2CH, Py), 122.28 (2CH, Py), 60.16 (2C, CH_2Py), 53.42 (C, NCH_2), 36.66 (C, CH_2SH). Anal. Calcd (found) for $C_{14}H_{17}N_3S$: C, 64.9 (64.8); H, 6.56 (6.47); N, 16.2 (16.3).

***N,N*-Bis(mercaptoethyl)(aminomethyl)-2-pyridine, L3.** 2-(Aminomethyl)pyridine (1.62 g, 1.49 mmol) in benzene (5 mL) was reacted with ethylene sulfide (3.57 mL, 5.99 mmol) in benzene (5 mL). The crude product was purified by silica gel column chromatography using methanol−chloroform (5:95) to yield L3 as a foul-smelling yellow oil (0.3113 g, 1.36 mmol, 91%). ^{1}H NMR (δ (ppm), $CDCl_3$): 8.48 (d, $J = 4.51$ Hz, 1H, PyH), 7.63 (t, $J = 7.89$ Hz, 1H, PyH), 7.47 (d, $J = 7.33$ Hz, 1H, PyH), 7.13 (t, $J = 6.77$ Hz, 1H, PyH), 3.75 (s, 2H, $PyCH_2$), 2.73 (m, 4H, NCH_2), 2.61 (m, 4H, CH_2S), 1.71 (m, 2H, SH). ^{13}C NMR (δ (ppm), $CDCl_3$): 158.50 (1C, Py), 151.09 (1CH, Py), 135.69 (1CH, Py), 124.12 (1CH, Py), 120.29 (1CH, Py), 59.33 (1C, CH_2Py), 56.62 (2C, NCH_2), 29.79 (2C, CH_2SH).

Ethyl *N,N*-(2-Pyridylmethyl)(mercaptoethyl)aminoacetate, L4. *N*-(2-Pyridylmethyl)glycine ethyl ester (1 g, 5.15 mol) in benzene (5 mL) was reacted with ethylene sulfide (0.6129 mL, 1.03 mmol) in benzene (5 mL). The crude product was purified by silica gel column chromatography using methanol− chloroform (4:96) to yield L4 as a foul-smelling brown oil (0.0661 g, 0.26 mmol, 53%). ^{1}H NMR (δ (ppm), $CDCl_3$): 8.52 (d, $J = 4.76$ Hz, 1H, PyH), 7.66 (t, $J = 7.93$ Hz, 1H, PyH), 7.52 (d, $J = 7.14$ Hz, 1H, PyH), 7.16 (t, $J = 6.35$ Hz, 1H, PyH), 4.16 (q, $J = 6.35$ Hz, 2H, OCH_2), 3.98 (s, 2H, $PyCH_2$), 3.44 (s, 2H, NCH_2CO), 3.03 (t, $J = 7.14$ Hz, 2H, NCH_2), 2.78 (t, $J = 7.14$ Hz, 2H, CH_2S) 1.27 (t, $J = 7.14$ Hz, 3H, CH_3). ^{13}C NMR (δ (ppm), $CDCl_3$): 169.72 (C, CO), 159.42 (C, Py), 149.30 (CH, Py), 136.80 (CH, Py), 123.26 (CH, Py), 122.35 (CH, Py), 60.66 (C, OCH_2), 60.25 (C, CH_2N), 55.26 (C, NCH_2-CH_2), 53.93 (C, NCH_2CO), 37.32 (C, CH_2SH), 14.47 (C, CH_3).

***N,N*-(2-Pyridylmethyl)(mercaptoethyl)aminoacetic Acid, L5.** The preparation of the intermediate L5a and the product L5 were based on a literature procedure[22] with some modifications.

2-(Aminomethyl)pyridine (1 g, 9.25 mmol) and ethylene sulfide (0.55 mL, 9.25 mmol) in toluene (5 mL) were placed in a borosilicate tube with 5/8 in. outside diameter, 3/32 in. wall thickness, and 6 in length. The sealed tube was heated at 115 °C for 44 h. After being cooled to room temperature, the solution was filtered and loaded onto a silica gel column. The intermediate L5a was eluted with ethyl acetate−ethanol (1:1) as a yellow oil (1.485 g, 8.79 mmol, 95%).

To a mixture of L5a (0.7 g, 4.14 mmol) and triethylamine (0.57 mL, 4.14 mmol) in THF (50 mL) was added chlorotrimethylsilane (0.52 mL, 4.14 mmol) dropwise at 0 °C; a white solid began to form during the addition. The mixture was stirred at 0 °C for 1 h, allowed to warm to room temperature, and stirred for another 12 h. A second portion of triethylamine (0.57 mL, 4.14 mmol) and bromoacetic acid (0.58 g, 4.14 mmol) in THF (5 mL) were added. After the mixture was heated under reflux for 20 h, the resulting white solid was filtered off and discarded. The crude organic mixture was then hydrolyzed and concentrated in vacuo. The residue was chromatographed on a silica gel column using dichloromethane followed by ethyl acetate as eluent to yield the pure product of L5 as a viscous liquid (0.4512 g, 1.9872 mmol, 48%). ^{1}H NMR (δ (ppm), $CDCl_3$): 8.46 (d, $J = 4.69$ Hz, 1H, PyH), 7.60 (t, $J = 7.62$ Hz, 1H, PyH), 7.26 (d, $J = 8.20$ Hz, 1H, PyH), 7.13 (t, $J = 6.44$ Hz, 1H, PyH), 4.67 (s, 2H, $PyCH_2$), 3.66 (t, $J = 5.86$ Hz, 2H, NCH_2), 3.31 (s, 2H, NCH_2CO), 2.77 (t, $J = 5.86$ Hz, 2H, CH_2S). ^{13}C NMR (δ (ppm), $CDCl_3$): 168.83 (C, CO), 161.22 (C, Py), 151.36 (CH, Py), 138.97 (CH, Py), 124.56 (CH, Py), 123.45 (CH,

(21) Spencer, D. J. E.; Blake, A. J.; Parsons, S.; Schroder, M. *J. Chem. Soc., Dalton Trans.* **1999**, 1041−1042.

(22) Wong, Y.-L.; Dilworth, J. R. *J. Chem. Soc., Dalton Trans.* **2002**, 2366−2370.

Py), 60.14 (C, CH_2N), 57.16 (C, NCH_2CH_2), 52.85 (C, NCH_2CO), 29.31 (C, CH_2SH).

Rhenium Complex Syntheses. All complexation reactions were run in a 1:1 ratio of ligand to rhenium starting material.

[Re(CO)$_3$)(L1)]Br, Re-1, and [Re(CO)$_3$(L1)Re(CO)$_3$Br$_2$], Re-1a. L1 (0.038 g. 0.1947 mmol) in 2 mL of methanol was added to a stirred solution of $[Re(CO)_3(H_2O)_3]Br$ (0.0525 g, 0.130 mmol) or $[NEt_4]_2[Re(CO)_3Br_3]$ (0.150 g, 0.1947 mmol) in 10 mL of methanol. The solution was refluxed for 4 h, whereupon a light green precipitate formed. This precipitate was filtered out and washed with 1 mL of methanol, and the mother liquor was evaporated to dryness to give a green, oily residue. The oily residue was purified by basic alumina column chromatography using 5:95 MeOH−CH_2Cl_2. ^{1}H NMR, ^{13}C NMR, and MS analysis of the purified oily residue and the precipitate identified the formation of two rhenium complexes, **Re-1** and **Re-1a**, respectively.

Re-1. This complex was obtained as brown oil. Yield: 73%. ^{1}H NMR (δ (ppm), MeOH): 8.89 (d, J = 6.06 Hz, 2H, PyH), 7.98 (t, J = 8.08 Hz, 2H, PyH), 7.64 (d, J = 8.08 Hz, 2H, PyH), 7.41 (t, J = 6.06 Hz, 2H, PyH), 5.06 (d, J = 16.16 Hz, 2H, $PyCH_2$), 4.92 (d, J = 16.16 Hz, 2H, $PyCH_2$), 4.02 (m, 2H, NCH_2), 3.16 (m, 2H, CH_2S), 2.75 (q, J = 8.08 Hz, 2H, SCH_2), 1.37 (t, J = 6.74 Hz, 3H, CH_3). ^{13}C NMR (δ (ppm), $CDCl_3$): 196.56, 195.60 (*fac*-$Re(CO)_3$), 160.38 (2C, Py), 151.89 (2CH, Py), 140.85 (2CH, Py), 125.96 (2CH, Py), 123.56 (2CH, Py), 69.04 (C, NCH_2), 67.12 (2C, CH_2-Py), 26.29 (C, CH_2S), 25.87 (C, SCH_2), 17.24 (C, CH_3). Mass spectrum (ESIMS): m/z = 533.7, $[Re(CO)_3(L1)]^+$. IR (NaCl, ν/cm^{-1}): 2031, 1908 (*fac*-$Re(CO)_3$).

Re-1a. This complex was obtained as green solid. Yield: 27%. ^{1}H NMR (δ (ppm), $CDCl_3$): 8.94 (d, J = 7.02 Hz, 2H, PyH), 8.15 (t, J = 7.45 Hz, 2H, PyH), 7.87 (d, J = 8.02 Hz, 2H, PyH), 7.61 (t, J = 6.26 Hz, 2H, PyH), 5.78 (d, J = 17.04 Hz, 2H, $PyCH_2$), 5.53 (d, J = 16.75 Hz, 2H, $PyCH_2$), 5.01 (m, 2H, NCH_2), 4.17 (m, 2H, CH_2S), 3.55 (m, 2H, SCH_2), 2.48 (m, 3H, CH_3). ^{13}C NMR (δ (ppm), $CDCl_3$): 197.34, 196.40, 196.03, 195.41 (*fac*-$Re(CO)_3$), 161.72 (2C, Py), 157.57 (2CH, Py), 150.12 (2CH, Py), 132.96 (2CH, Py), 129.76 (2CH, Py), 71.23 (C, NCH_2), 65.42 (2C, CH_2-Py), 32.19 (C, CH_2S), 23.67 (C, SCH_2), 15.38 (C, CH_3). Mass spectrum (ESIMS): m/z = 988.69, $[Re_2(CO)_6(L\text{-}1a)Br_2H]^+$. IR (NaCl, ν/cm^{-1}): 2034, 2027, 1911, 1905 (*fac*-$Re(CO)_3$).

[Re(CO)$_3$(L2)], Re-2. L2 (0.0337 g, 0.1298 mmol) in 3 mL of methanol was added to a stirred solution of $[Re(CO)_3(H_2O)_3]Br$ (0.0525 g, 0.1298 mmol) or $[NEt_4]_2[Re(CO)_3Br_3]$ (0.100 g, 0.1298 mmol) in 10 mL of methanol. The solution was refluxed for 6 h to yield the desired product as yellow oil. Recrystallization from chloroform gave yellow crystals suitable for X-ray crystallography. Yield: 98%. ^{1}H NMR (δ (ppm), MeOH): 8.927 (d, J = 5.90 Hz, 2H, PyH), 8.00 (t, J = 7.86 Hz, 2H, PyH), 7.70 (d, J = 8.51 Hz, 2H, PyH), 7.43 (t, J = 5.84 Hz, 2H, PyH), 5.22 (d, 2H, J = 17.03 $PyCH_2$), 4.99 (d, 2H, J = 17.03 $PyCH_2$), 4.26 (m, 2H, NCH_2), 3.54 (m, 2H, CH_2S). ^{13}C NMR (δ (ppm), MeOH-d_4): 199.17, 198.32, 197.26 (*fac*-$Re(CO)_3$), 162.11 (1C, Py), 161.04 (1C, Py), 153.42 (1CH, Py), 151.98 (1CH, Py), 141.99 (2CH, Py), 127.16 (2CH, Py), 125.04 (1CH, Py), 124.23 (1CH, Py), 71.03 (C, NCH_2), 69.34 (2C, CH_2Py), 33.75 (C, CH_2S). Mass spectrum (ESIMS): m/z = 528.49, $[Re(CO)_3(L2)]$. IR (NaCl, ν/cm^{-1}): 2026, 1907 (*fac*-$Re(CO)_3$). Anal. Calcd (found) for $C_{17}H_{16}N_3O_3ReS$: C, 38.6 (38.4); H, 3.03 (3.11); N, 7.95 (8.10).

[Re(CO)$_3$(L3)], Re-3. L3 (0.0296 g. 0.1298 mmol) in 2 mL of methanol was added to a stirred solution of $[Re(CO)_3(H_2O)_3]Br$ (0.0525 g, 0.1298 mmol) or $[NEt_4]_2[Re(CO)_3Br_3]$ (0.100 g, 0.1298 mmol) in 10 mL of methanol. Purification was performed by silica gel chromatography using 1:99 MeOH−$CHCl_3$. Yield: 95%. ^{1}H NMR (δ (ppm), MeOH-d_4): 8.87 (d, J = 5.30 Hz, 1H, PyH), 8.35 (t, J = 7.90 Hz, 1H, PyH), 8.05 (d, J = 7.26 Hz, 1H, PyH), 7.64 (t, J = 5.54 Hz, 1H, PyH), 3.95 (m, 2H, $PyCH_2$), 3.65 (m, 2H, NCH_2), 3.13 (m, 2H, NCH_2), 3.01 (m, 2H, CH_2S), 2.82 (m, 2H, CH_2S). ^{13}C NMR (δ (ppm), $CDCl_3$): 199.35, 198.21, 194.10 (*fac*-$Re(CO)_3$), 152.50 (1C, Py), 148.15 (1CH, Py), 138.29 (1CH, Py), 138.01 (1CH, Py), 125.51 (1CH, Py), 67.02 (1C, CH_2Py), 55.89 (2C, NCH_2), 34.51 (1C, CH_2SH), 27.17 (1C, CH_2SH). IR (NaCl, ν/cm^{-1}): 2025, 1905 (*fac*-$Re(CO)_3$).

[Re(CO)$_3$(L4)], Re-4. L4 (0.033 g, 0.1298 mmol) in 2 mL of methanol was added to a stirred solution of $[Re(CO)_3(H_2O)_3]Br$ (0.0525 g, 0.1298 mmol) or $[NEt_4]_2[Re(CO)_3Br_3]$ (0.100 g, 0.1298 mmol) in 10 mL of methanol. Purification was effected by silica gel chromatography using 3:97 MeOH−CH_2Cl_2. Yield: 90%. ^{1}H NMR (δ (ppm), $CDCl_3$): 8.86 (d, J = 5.26 Hz, 1H, PyH), 7.88 (t, J = 7.54 Hz, 1H, PyH), 7.41 (m, 2H, PyH), 4.31 (q, J = 6.01 Hz, 2H, OCH_2), 3.86 (s, 2H, $PyCH_2$), 3.50 (s, 2H, NCH_2CO), 2.20 (m, 4H, NCH_2CH_2S), 1.35 (t, J = 6.77 Hz, 3H, CH_3). ^{13}C NMR (δ (ppm), MeOH-d_4): 198.34, 197.97, 196.79, (*fac*-$Re(CO)_3$), 170.20 (C, CO), 158.13 (C, Py), 141.42 (CH, Py), 134.36 (CH, Py), 123.55 (CH, Py), 122.02 (CH, Py), 61.54 (CH, OCH_2), 58.57 (C, CH_2N), 49.98 (C, NCH_2CH_2), 46.13 (C, NCH_2CO), 38.14 (C, CH_2SH), 15.12 (C, CH_3). Mass spectrum (ESIMS): m/z = 496.0, $[Re(CO)_3$-(L4)]. IR (NaCl, ν/cm^{-1}): 2035, 1924 (*fac*-$Re(CO)_3$), 1710 (ν_{as}-(C═O)), 1200 (ν_{sym}(C═O)) of the ester group.

[Re(CO)$_3$(L5)], Re-5. L5 (0.0294 g, 0.1298 mmol) in 2 mL of methanol was added to a stirred solution of $[Re(CO)_3(H_2O)_3]Br$ (0.0525 g, 0.1298 mmol) or $[NEt_4]_2[Re(CO)_3Br_3]$ (0.100 g, 0.1298 mmol) in 10 mL of methanol and stirred at room temperture for 72 h. Silica gel chromatography using 8:92 MeOH−CH_2Cl_2 provided pure **Re-5** in greater than 90% yield. Recrystallization from MeOH/CH_2Cl_2 provided microcrystalline **Re-5** in 70% yield. ^{1}H NMR (δ (ppm), MeOH-d_4): 8.77 (d, J = 4.55 Hz, 1H, PyH), 7.89 (t, J = 7.80 Hz, 1H, PyH), 7.66 (d, J = 7.80 Hz, 1H, PyH), 7.70 (t, J = 5.85 Hz, 1H, PyH), 4.74 (s, 2H, $PyCH_2$), 3.73 (t, J = 5.86 Hz, 2H, NCH_2), 3.40 (s, 2H, NCH_2CO), 2.92 (t, J = 5.86 Hz, 2H, CH_2S). ^{13}C NMR (δ (ppm), MeOH-d_4): 199.21, 198.36, 196.88 (*fac*-$Re(CO)_3$), 174.40 (C, CO), 150.13 (C, Py), 139.20 (CH, Py), 124.23 (CH, Py), 123.49 (2CH, Py), 53.56 (C, NCH_2CO)), 51.17 (C, CH_2N), 30.92 (C, NCH_2CH_2), 27.14 (C, CH_2SH). Mass spectrum (ESIMS): m/z = 524.0, $[Re(CO)_3(L5)]$. IR (NaCl, ν/cm^{-1}): 2027, 1905 (*fac*-$Re(CO)_3$), 1610 (ν_{as}(C═O)), 1160 (ν_{sym} (C═O)) of the acid group.

X-ray Crystal Structure Determinations. The selected crystals of the complexes **Re-1a** and **Re-2** were studied on a Bruker diffractometer equipped with the SMART CCD system,[23] using graphite-monochromated Mo Kα radiation (λ = 0.710 73 Å). The data collections were carried out at 90(5) K. The data were corrected for Lorentz polarization effects, and absorption corrections were made using SADABS.[24] All calculations were performed using SHELXTL.[25] The structures were solved by direct methods, and all of the non-hydrogen atoms were located from the initial solution. After location of all the non-hydrogen atoms in the structure, the model was refined against F^2, initially using isotropic and later anisotropic thermal displacement parameters until the final value

(23) *Smart Software Reference Manual*; Siemens Analytical X-ray Instruments, Inc.: Madison, WI, 1994.
(24) Sheldrick, G. M. *SADABS: Program for Empirical Absorption Correction*; University of Göttingen: Göttingen, Germany, 1996.
(25) Sheldrick, G. M. *SHELXL96: Program for Refinement of Crystal Structures*; University of Göttingen: Göttingen, Germany, 1996.
(26) Hawkes, M. J.; Ginsberg, A. P. *Inorg. Chem.* **1969**, *8*, 2189.
(27) Abel, E. W.; Butler, I. S.; Ganorkar, M. C.; Jenkins, C. R.; Stiddard, M. H. B. *Inorg. Chem.* **1966**, *5*, 2589.

Table 1. Crystallographic Data for the Structures of **Re-2** and **Re-1a**

param	**Re-2**	**Re-1a**
empirical formula	$C_{17}H_{16}N_3O_3ReS$	$C_{22}H_{21}Br_2N_3O_6Re_2S$
fw	528.59	987.70
space group	$P2_1/c$	$P2_1/c$
T, K	81(2)	101(2)
a, Å	15.846(2)	11.2217(7)
b, Å	9.5722(12)	19.7326(12)
c, Å	11.6240(15)	12.3746(7)
β, deg	104.272(2)	100.000(1)
V, Å^3	1708.7(4)	2698.5(3)
Z	4	4
D_{calc}, Mg cm^{-3}	2.055	2.431
μ, mm^{-1}	7.255	12.044
R1[a] (all data)	0.0197	0.1091
wR2[b]	0.0457	0.1924

[a] $R1 = \sum||F_o| - |F_c||/\sum|F_o|$. [b] $wR2 = [\sum w(F_o^2 - F_c^2)^2/\sum w(F_o^2)^2]^{1/2}$.

Table 2. Selected Bond Lengths (Å) and Angles (deg) for the Structures of $[Re(CO)_3\{(2\text{-pyridyl-}CH_2)_2N(CH_2)_2S)]$, **Re-2**, and $[Re(CO)_3\{(2\text{-pyridyl-})(CH_2)_2N(CH_2)_2SEt\}Re(CO)_3Br_2]$, **Re-1a**

param	**Re-2**	**Re-1a**
Re−C, range	1.9062(19)−1.9291(19)	1.884(16)−1.939(18)
Re−N amine	2.2704(16)	2.245(15)
Re−N pyridine	2.1814(15)	2.184(14)
Re−S	2.4666(5)	2.561(6)
Re−Br, range	N/A	2.621(3)−2.616(2)
C−Re−X trans angles	171.03(7)−177.09(6)	171.7(7)−179(2)

of Δ/σ_{max} was less than 0.001. At this point the hydrogen atoms were located from the electron density difference map and a final cycle of refinements was performed, until the final value of Δ/σ_{max} was again less than 0.001. No anomalies were encountered in the refinement of the structure. The relevant parameters for crystal data, data collection, structure solution, and refinement are summarized in Table 1, and important bond lengths and bond angles are presented in Table 2.

Technetium-99m Labeling. $Na[^{99m}Tc(CO)_3(H_2O)_3]$ was prepared according to the literature method.[9] $Na[^{99m}TcO_4]$ was sonicated for 5 min to facilitate the removal of dissolved O_2(g). Approximately 1.0 mL of generator eluate containing 16.30 mCi of $Na[^{99m}TcO_4]$ was added to the mixture consisting of $K_2[BH_3{\cdot}CO_2]$ (0.0085 g, 0.063 mmol), $Na_2B_4O_7{\cdot}10H_2O$ (0.0030 g, 0.0079 mmol), Na/K-tartratrate (0.0150 g, 0.053 mmol), and Na_2CO_3 (0.0040 g, 0.038 mmol). $Na[^{99m}Tc(CO)_3(H_2O)_3]$ was obtained after 30 min at 95 °C, with the final solution pH of ca. 12.0. Reaction progress was monitored by HPLC.

All technetium compounds were purified by Sep-Pak (Waters) methods. The columns were conditioned with 10 mL of absolute ethanol (2−3 drops/s) followed by 10 mL of 0.01 M HCl (2−3 drops/s). The samples were loaded with a 3 mL syringe and eluted using HCl/ACN solutions. The elution ratios are summarized in the Supporting Information.

Synthesis of $[^{99m}Tc(CO)_3$(L1)]Br, ^{99m}Tc-1. L1 (1.0 mg, 0.0035 mmol) was dissolved in 200 μL of deionized H_2O and 10 μL of DMSO. The ligand solution was flushed with N_2 for 30 min. $[^{99m}Tc(CO)_3(H_2O)_3]^+$ (500 μL, 85 μCi) was introduced to the solution mixture which was incubated at 95 °C for 45 min. Purity of the complex **^{99m}Tc-1** was assessed by HPLC.

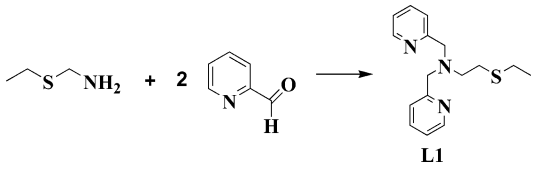
Synthesis of $[^{99m}Tc(CO)_3$(L2)], ^{99m}Tc-2. L2 (1.0 mg, 0.0039 mmol) was dissolved in 200 μL of deionized H_2O and 10 μL of DMSO. The ligand solution was flushed with N_2 for 30 min, whereupon $[^{99m}Tc(CO)_3(H_2O)_3]^+$ (500 μL, 82 μCi) was introduced to the solution mixture which was incubated at 95 °C for 45 min.

Syntheses of $[^{99m}Tc(CO)_3$(L3)], ^{99}Tc-3, and $[^{99m}Tc(CO)_3$(L4)], ^{99}Tc-4. The complexes were prepared in a fashion similar to that for **^{99m}Tc-2** from the appropriate ligand L*n* and $[^{99m}Tc(CO)_3(H_2O)_3]^+$. Purities of the complexes were assessed by HPLC.

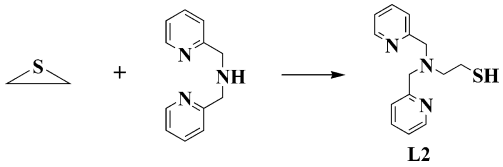
Synthesis of $[^{99m}Tc(CO)_3$(L5)], ^{99m}Tc-5. L5 (1.0 mg, 0.0044 mmol) was dissolved in 200 μL of deionized H_2O and 10 μL of DMSO. The ligand solution was flushed with N_2 for 30 min, whereupon $[^{99m}Tc(CO)_3(H_2O)_3]^+$ (500 μL, 76 μCi) was introduced to the solution mixture which was incubated at 95 °C for 45 min.

Analysis of the reaction products using C18 HPLC showed greater than 90% radiochemical purity for the **^{99m}Tc-1** and **^{99m}Tc-2** complexes. Similarly, Cl8 HPLC analyses of **^{99m}Tc-3** and **^{99m}Tc-4** exhibited greater than 95% radiochemical purity. However, the complexation of L5 resulted in multiple product formation. The HPLC analysis was performed using a C18 semipreparative column, 25 cm × 4.6 mm (5 μm pore size), equipped with a 2 cm guard column. Solvent A was 0.1% TFA in ddH_2O, and solvent B was 0.1% TFA in CH_3CN. The method employed a gradient run over 30 min at a flow rate of 4.0 mL/min. The gradient was ramped from 75:25 A/B to 20:80 A/B for 30 min.

Scheme 1



Scheme 2

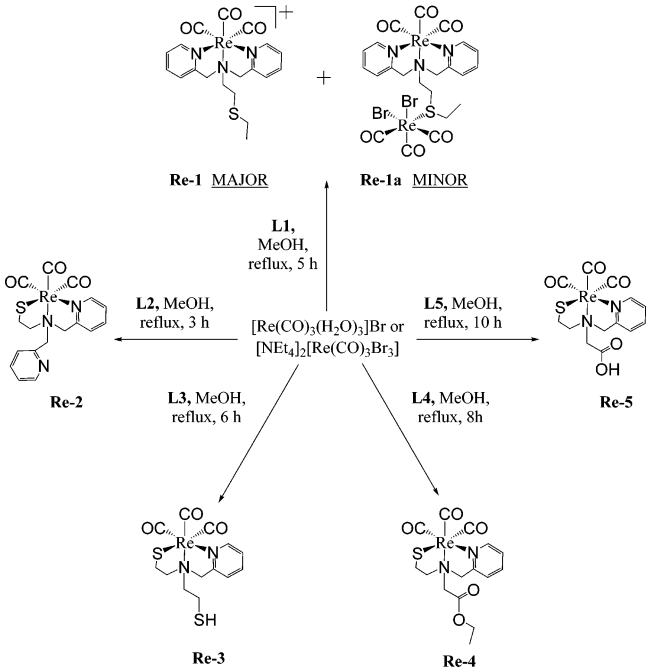


Results and Discussion

Syntheses and Spectroscopic Properties. The reductive alkylation of 1 equiv of 2-(ethylthio)ethylamine with 2 equiv of pyridinicarboxaldehyde in 1,2-dichloroethane in the presence of $NaBH(OAc)_3$ provided L1 in excellent yield (Scheme 1). Ligands L2−L5 were prepared by the reactions of ethylene sulfide with the appropriate amine starting material (Scheme 2). Thus, the reaction of 1 equiv of 2,2′-dipicolylamine with 1 equiv of ethylene sulfide in benzene gave L2, while the reaction of 1 equiv of 2-(aminomethyl)pyridine with 2 equiv of ethylene sulfide in benzene gave L3 in greater than 90% yield. The ligand L4 was prepared by reacting ethylene sulfide with *N*-(2-pyridylmethyl)glycine ethyl ester. The reaction of 2-(aminomethyl)pyridine with ethylene sulfide in a sealed borosilicate tube at 115 °C yielded *N*,*N*-(2-pyridylmethyl)(mercaptoethyl)amine L5a, which upon reaction with bromoacetic acid in the presence of triethylamine and chlorotrimethylsilane provided L5. The ligands were characterized by ^{1}H and ^{13}C NMR spectroscopy.

Complexes **Re-1**−**Re-4** were prepared in excellent yields by refluxing equivalent amounts of each ligand and $[Re(CO)_3(H_2O)_3]Br$ and/or $[NEt_4]_2[Re(CO)_3Br_3]$ in methanol for a few hours (Scheme 3). The two rhenium precursors were used for comparison purposes. Although both materials proved to be effective, complexation of the ligands with [Re-

Scheme 3



$(CO)_3(H_2O)_3]Br$ afforded the final products in much higher yield and greater purity of the crude reaction mixtures.[20] The disadvantages of the starting material $[NEt_4]_2[Re(CO)_3Br_3]$ include arduous preparation from the hydrolysis of $[Re(CO)_5Br]$ with NEt_4Br, the production of unwanted byproducts such as $(NEt_4)[Re(CO)_4Br_2]$ and $(NEt_4)_2[Re_2(CO)_6Br_4]$,[26,27] and the need to remove excess NEt_4Br. In contrast, $[Re(CO)_3(H_2O)_3]Br$ may be prepared directly from $[Re(CO)_5]Br$ by refluxing in water. Furthermore, the undesirable byproduct NEt_4Br from reactions with $(NEt_4)_2[Re(CO)_3Br_3]$ has polarity similar to that of the $[Re(CO)_3L]^+$ complexes, increasing the difficulties of purification by chromatography. In addition, NEt_4Br preferentially cocrystallizes with $[Re(CO)_3L]^+$ complexes. As summarized in eq 1, the use of $[Re(CO)_3(H_2O)_3]Br$ avoids these difficulties.

$$[Re(CO)_3(H_2O)_3]Br + Ln \rightarrow [Re(CO)_3(Ln)]^m \textbf{(Re-}\boldsymbol{n}\textbf{)} + H_2O \quad m = 0, +1;\ n = 1-5$$

$$[NEt_4]_2[Re(CO)_3Br_3] + Ln \rightarrow [Re(CO)_3(Ln)]^m \textbf{(Re-}\boldsymbol{n}\textbf{)} + NEt_4Br \text{ (undesired byproduct)} \quad (1)$$

In the case of **Re-1** and **Re-1a**, the complexes were obtained as a 70%:30% mixture from the reaction of $[Re(CO)_3(H_2O)_3]Br$ or $(NEt_4)_2[Re(CO)_3Br_3]$ and L1. The minor product precipitated directly from methanol, whereas the major product **Re-1** was obtained as brown oil following evaporation of the solvent.

In the case of **Re-5**, the reactants were stirred at room temperature rather than subjected to reflux due to the thermal instability of the ligand. Heating of solutions of **L5** generally resulted in varying degrees of decomposition and disulfide formation, providing reduced yields of the product **Re-5**. Under less forcing conditions, **Re-5** is obtained in greater than 90%. Furthermore, there is no evidence for the formation of a complex bound to the carboxylate oxygen donor rather than to the thiolate sulfur donor.

The IR spectra of these complexes contain strong bands in the region 2035−1924 cm^{-1} indicating the presence of the *fac*-$\{Re(CO)_3\}^+$ core. **Re-4** and **Re-5** exhibit two strong absorptions in the regions 1610−1710 and 1160−1200 cm^{-1}, corresponding to the two vibrations of the carboxylate group, $\nu_{as}(C{=}O)$ and $\nu_{sym}(C{=}O)$, respectively.

^1H and ^{13}C NMR spectroscopy indicates that the complexes retain their solid-state structure in solution. The assignments of all protons in all the ligands and the corresponding complexes are based on intensity and spin−spin splitting structure and are presented in the Experimental Section. The cationic species **Re-1** exhibits mirror symmetry along one of the three facial carbonyl groups and the aliphatic amine side chain, an observation reflected in the ^1H NMR spectrum where the proton signals of the methylene group adjacent to the pyridine are split into two sets of doublets with germinal coupling constants (17−20 Hz). Also, only one set of pyridine protons is observed. The ^{13}C NMR spectrum of **Re-1** displays two Re−CO peaks at 194−198 ppm with a 2:1 peak ratio, indicative of two CO groups that are magnetically equivalent due to the mirror symmetry (Figure 1). Likewise, the ^{13}C NMR spectrum showed only one set of pyridine carbons, a feature also consistent with mirror symmetry along an axis formed by the Re and one CO group. In contrast, the neutral species **Re-2**−**Re-5** do not possess a mirror phase, and the ^{13}C NMR reveals three, nonequivalent carbonyl peaks in the ratio 1:1:1 in the range 194−199 ppm (Figure 2).

The mass spectra of the complexes are consistent with the formulations from elemental analysis and from the spectroscopy. For example, the ESMS mass spectrum of **Re-1** exhibits the most prominent *m*/*z* peak at 533.7, which is assigned to $[Re(CO)_3L1]^+$ on the basis of the isotopic distribution of $^{185,187}Re$. Similarly for **Re-1a**, the peak at *m*/*z* 988.69 corresponds to $[Re_2(CO)_6(L1a)Br_2H]^+$.

Crystallographic Studies. X-ray structural results of **Re-2** confirm that this model neutral complex exhibit the chemically robust *fac*-$\{Re(CO)_3\}^+$ core and distorted octahedral geometry (Figure 3). The octahedral environment of the Re(I) site is defined by the three facially bound CO groups, the aliphatic amine, one of the aromatic amines, and the sulfur donor of the ligand. One pyridyl group is present as a pendant arm. The Re−carbonyl bond distances are unexceptional and consistent with those found in similar complexes. The Re−N2 distance of 2.1814(15) Å is somewhat longer than that of 2.2704(16) Å for Re−N1, consistent with sp^3- and sp^2-hybridized nitrogen donors, respectively. The Re−S distance of 2.4666(5) Å is unexceptional.[28−30] The trans angles fall in the 171.03(7)−177.09(6)° exhibiting only minor deviations from the octahedral limit. The most significant angular distortions arise from the formation of strained five-membered chelate rings and are associated with the N1−Re−N2 and N1−Re−S angles of 76.45(6) and 83.66(4)°, respectively.

(28) Pietzsch, H.-J.; Gupta, A.; Reisgys, M.; Drews, A.; Seifert, S.; Syhre, R.; Spies, H.; Alberto, R.; Abram, U.; Schubiger, P. A.; Johannsen, B. *Bioconjugate Chem.* **2000**, *11*, 414.

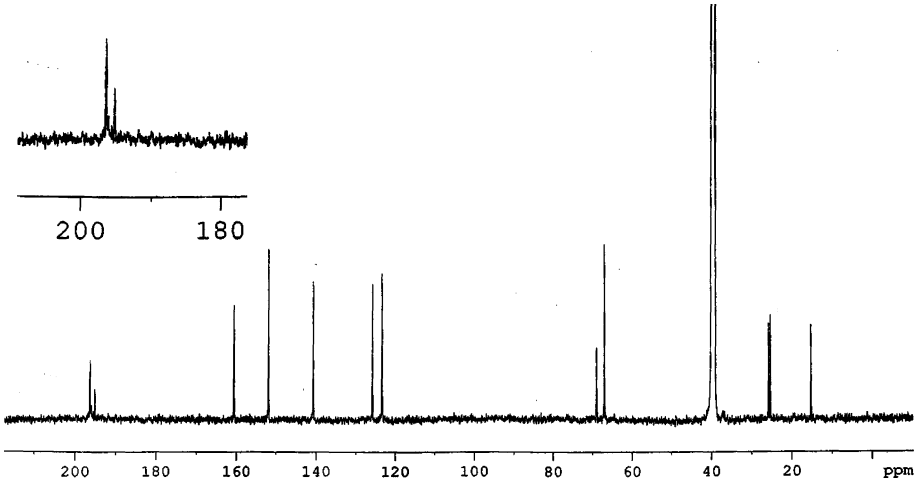


Figure 1. ^{13}C NMR spectrum of $[Re(CO)_3(L1)]Br$, **Re-1**, in $CDCl_3$ solution. The inset shows the carbonyl carbon region, 170−210 ppm.

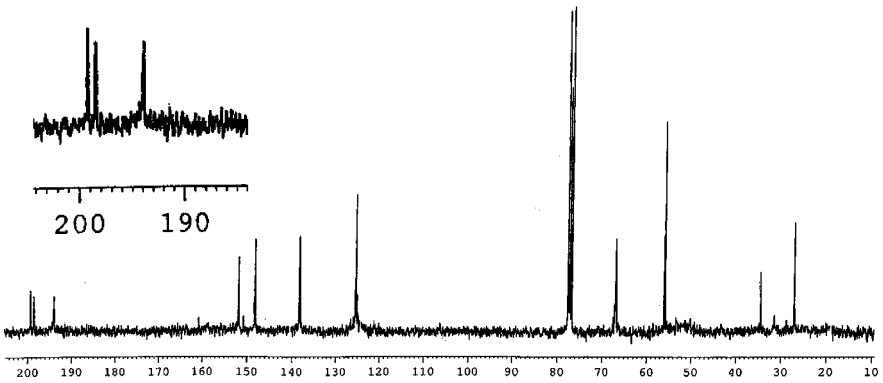


Figure 2. ^{13}C NMR spectrum of $[Re(CO)_3(L3)]$, **Re-3**, in $CDCl_3$ solution. The inset shows the carbonyl carbon region, 180−210 ppm.

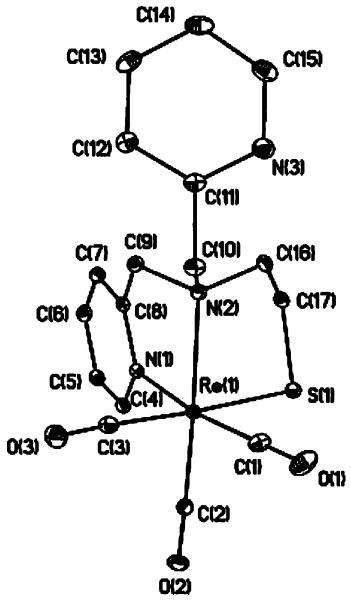


Figure 3. ORTEP view of the structure of $[Re(CO)_3(L2)]$, **Re-2**, showing the atom-labeling scheme and 50% thermal ellipsoids.

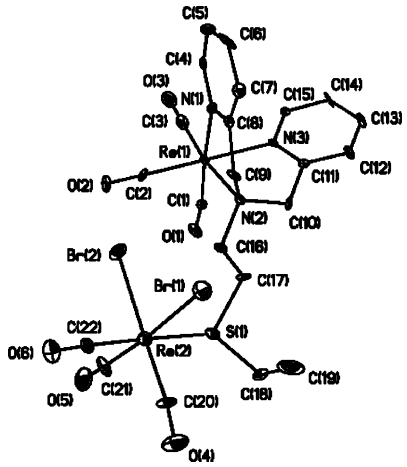
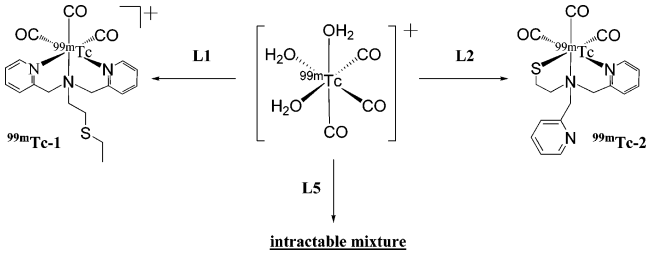


Figure 4. ORTEP view of the structure of $[Re(CO)_3(L1)Re(CO)_3Br_2]]$, **Re-1a**, showing the atom-labeling scheme and 50% thermal ellipsoids.

As shown in Figure 4, the structure of **Re-1a** consists of discrete binuclear units. The structure may be described as an $[Re(CO)_3)(L1)]^+$ subunit linked to an $[Re(CO)_3Br_2]^-$ group through the sulfur of the pendant thioether arm. Consequently, there are two distinct coordination geometries *fac*-$\{ReC_3N_3\}$ and *fac*-$\{ReC_3Br_2S\}$. The coordination geometry at the Re1 site is similar to that previously observed for $[Re(CO)_3\{(2\text{-pyridylmethyl})_2NCH_2CO_2H\}]Br$[10] with unexceptional metrical parameters. The Re−carbonyl bond

(29) Meltzer, P. C.; Blundell, P.; Jones, A. G.; Mahmood, A.; Garada, B.; Zimmerman, R. E.; Davison, A.; Holman, B. L.; Madras, B. K. *J. Med. Chem.* **1997**, *40*, 1835.
(30) Friebe, M.; Mahmood, A.; Bolzati, C.; Drews, A.; Johannsen, B.; Eisenhut, M.; Kraemer, D.; Davison, A.; Jones, A. G. *J. Med. Chem.* **2001**, *44*, 3132.

Scheme 4



distances at the Re2 site are in the usual range, and the Re2−Br and Re−S distances are 2.621(3) and 2.561(6) Å, respectively.

The X-ray structures and the spectroscopic data would suggest that thiolate sulfur is an effective ligand for the $\{M(CO)_3\}^+$ core and may, in fact, compete with the pyridyl group as a donor. In contrast, thioether sulfur is not as effective a donor group, suggesting an apparent coordination preference of the $\{Re(CO)_3\}^+$ unit of pyridyl nitrogen ≈ thiolate > carboxylate > thioether sulfur ≫ thiophene sulfur. Previous studies suggested that while thioether sulfurs are somewhat effective donors in general, thiophene is an especially weak ligand.[10] As a relatively soft acid, the $\{Re(CO)_3\}^+$ core would be expected to prefer ligands with aromatic nitrogen and sulfur donors.[10,19,31−33]

Labeling of the Ligands with ^{99m}Tc. The organometallic starting material $[^{99m}Tc(CO)_3(H_2O)_3]^+$ was prepared by a modified literature procedure. Reactions of the ligands L1 and L2 (0.0035−0.0044 mmol) with $[^{99m}Tc(CO)_3(H_2O)_3]^+$ yielded $[^{99m}Tc(CO)_3(L1)]^+$, **^{99m}Tc-1**, and $[^{99m}Tc(CO)_3(L2)]$, **^{99m}Tc-2**, in 93% and 88% yield, respectively, after purification using a C-18 Sep-Pak to remove unreacted ligand (Scheme 4).

Ligand challenge experiments were carried out to determine whether **^{99m}Tc-1** and **^{99m}Tc-2** remain intact in the presence of large excesses of competing thiol and nitrogen donor ligands common to physiological systems. To demonstrate the stability of the complexes, **^{99m}Tc-1** and **^{99m}Tc-2** were incubated in separate experiments with 1000-fold excesses of cysteine and histidine at 37 °C in phosphate buffered saline (pH = 7.2) for 24 h. The radiochromatograms of the resultant solutions showed greater than 95% retention of the complex (Supporting Information Figures 5 and 6), suggesting that complexes such as **^{99m}Tc-1** and **^{99m}Tc-2** are sufficiently robust to be used as synthons for preparing radiopharmaceuticals, as are the ^{99m}Tc complexes of the parent *N*,*N*-bis(2-pyridylmethyl)amine analogues.[34] In contrast to the rhenium analogue **Re-5**, the complex **^{99m}Tc-5** could not be obtained as a single product. This observation most likely relfects the relative thermal instability of L5 with respect to the other ligands of this study. Solutions of L5 subjected to heating were observed to produce disulfides and other unidentified products within the time periods required for labeling with ^{99m}Tc.

Conclusions

Encouraged by the effectiveness of bifunctional chelators of the single amino acid (SAAC) design in forming robust complexes with the $\{M(CO)_3\}^+$ core (M = ^{99m}Tc, Re), we have prepared a series of analogous tridentate chelates incorporating thiolate and thioether sulfur donors, in addition to the more conventional nitrogen and carboxylate donors. The ligands react cleanly and in high yield with $[NEt_4]_2[Re(CO)_3Br_3]$ or $[Re(CO)_3(H_2O)_3]Br$ to provide cationic complexes, such as $[Re(CO)_3)(L1)]^+Br^-$, **Re-1**, and neutral species, such as $[Re(CO)_3(Ln)]$ (n = 2−5; **Re-2**−**Re-5**, respectively). The series of complexes establish that thiolate sulfur is an effective ligand for the $\{M(CO)_3\}^+$ core, a not unexpected observation in view of the relatively soft receptor properties of the $\{M(CO)_3\}^+$ core. A qualitative ordering of the coordination preferences for the core may be established: pyridyl nitrogen ≈ thiolate > carboxylate > thioether sulfur ≫ thiophene sulfur. The greater affinity of the $\{M(CO)_3\}^+$ core for the thiolate group relative to carboxylate donors is established by the isolation in high yield of **Re-5** in which the ligand binds through the thiolate donor in preference to the carboxylate group and by trends previously discussed by others.[16,19]

The comparative synthetic studies using $(NEt_4)_2[Re(CO)_3Br_3]$ and $[Re(CO)_3(H_2O)_3]Br$ establish that the latter facilitates product purification by avoiding the formation of the undesired byproduct NEt_4Br. Not only does the similarity in the polarities of NEt_4Br and $\{M(CO)_3L\}^+$ complexes render purification by silica or alumina chromatography difficult but NEt_4Br also preferentially cocrystallizes with such complexes.

The ^{99m}Tc analogues $[^{99m}Tc(CO)_3(L1)]^+$, **^{99m}Tc-1**, and $[^{99m}Tc(CO)_3(L2)]$, **^{99m}Tc-2**, were readily prepared using standard unit formulations in >90% radiochemical purity and high yield. The compounds are resistant to cysteine and histidine challenges.

The ligands of this study provide models for the design of SAAC-based bifunctional chelates incorporating thiolate ligands. Replacement of one or more pyridyl arms of the chelating terminus of the SAAC by thiolate or thioether groups should reduce the lipophilicity of the complex and, consequently, improve hepatobiliary concerns. The ligands are not only effective reagents for the design of ^{99m}Tc-based nuclear imaging agents but may also play a role in the development of therapeutics based on ^{186,188}Re.

Acknowledgment. This work was supported by a grant from the National Institutes of Health, National Institute of Allergy and Infectious Diseases, STTR 1R41 A1044080-01.

Supporting Information Available: Tables of complete crystal data, final coordinates, temperature factors, distances, and angles for the crystallographic study of **Re-L1a** and **Re-2** in CIF format, a summary of the elution fractions and activities for the Sep-Paks purifications, and radiochromatograms for **^{99m}Tc-1** and **^{99m}Tc-2**. This material is available free of charge via the Internet at http://pubs.asc.org.

IC050735A

(31) Santos, I. G.; Abram, U.; Alberto, R.; Lopez, E. V.; Sanchez, A. *Inorg. Chem.* **2004**, *43*, 1834.
(32) Alves, S.; Paulo, A.; Correia, J. D. G.; Domingos, A.; Santos, I. *J. Chem. Soc., Dalton Trans.* **2002**, 4714.
(33) Wüst, F.; Skaddan, M. B.; Leibnitz, P.; Spies, H.; Katzenellenbogen, J. A.; Johannsen, B. *Bioorg. Med. Chem.* **1999**, *7*, 1827.
(34) Liu, G.; Dou, S.; He, J.; Vanderheyden, J.-L.; Rusckowski, M.; Hnatorwich, D. J. *Bioconjugate Chem.* **2004**, *15*, 1441.