Journal of Coordination Chemistry

ISSN: 0095-8972 (Print) 1029-0389 (Online) Journal homepage: http://www.tandfonline.com/loi/gcoo20

Synthesis and stability of 2+1 complexes of N,N-diethylbenzoylthiourea with $[M^{I}(CO)_3]^+$ (M = Re, ^{99m}Tc)

Thomas R. Hayes, Ashton S. Powell, Charles L. Barnes & Paul D. Benny

To cite this article: Thomas R. Hayes, Ashton S. Powell, Charles L. Barnes & Paul D. Benny (2015) Synthesis and stability of 2+1 complexes of N,N-diethylbenzoylthiourea with $[M^{I}(CO)_3]^+$ (M = Re, ^{99m}Tc), Journal of Coordination Chemistry, 68:19, 3432-3448, DOI: 10.1080/00958972.2015.1071801

To link to this article: http://dx.doi.org/10.1080/00958972.2015.1071801

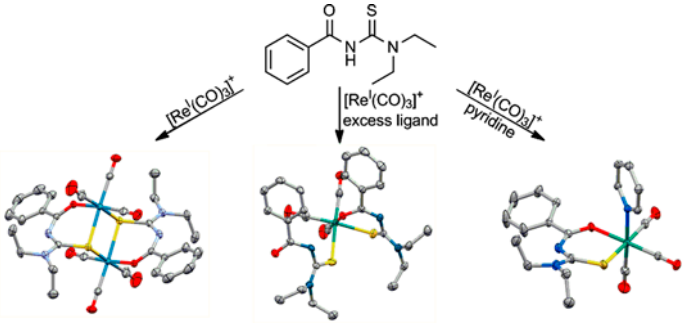
Journal of Coordination Chemistry, 2015
Vol. 68, No. 19, 3432–3448, http://dx.doi.org/10.1080/00958972.2015.1071801

Synthesis and stability of 2+1 complexes of N, N-diethylbenzoylthiourea with $[M^{I}(CO)_3]^+$ (M = Re, ^{99m}Tc)

THOMAS R. HAYES†, ASHTON S. POWELL†, CHARLES L. BARNES‡ and PAUL D. BENNY*†

†Department of Chemistry, Washington State University, Pullman, WA, USA
‡Department of Chemistry, University of Missouri, Columbia, MO, USA

(*Received 16 March 2015; accepted 22 June 2015*)



In the last two decades, the *fac*-$[M^{I}(OH_2)_3(CO)_3]^+$ (M = Re, ^{99m}Tc) core has been an important synthon in the development of new radiopharmaceuticals and methodologies. A bidentate N,N-diethylbenzoylthiourea (HEt_2btu, **H1**) ligand was reacted with *fac*-$[M^{I}(OH_2)_3(CO)_3]^+$ as part of a 2+1 strategy consisting of a bidentate and a monodentate ligands to saturate the coordination sphere. When excess **H1** was used in the reaction, an unusual 2+1 complex of *fac*-$[Re^{I}(CO)_3(\mathbf{1})(\mathbf{H1})]$, **2**, was isolated that contained two **H1** ligands, OS bidentate and S monodentate in different protonation states. Equimolar concentrations of HEt_2btu and *fac*-$[Re^{I}(CO)_3]^+$ generated the single addition of the ligand; however, isolation of the solid revealed a μ-sulfur-bridged dimer of *fac*-$[Re^{I}(CO)_3\mathbf{1})]_2$, **3**. Reaction of **H1** with *fac*-$[M^{I}(OH_2)_3(CO)_3]^+$ followed by the addition of a monodentate ligand (i.e. pyridine, methyl imidazole, cyclohexyl isocyanide, triphenylphosphine) yielded the corresponding 2+1 *fac*-$[M^{I}(CO)_3(\mathbf{1})(L)]$ complexes. Single-crystal X-ray structural analysis of the Re complexes revealed conformational differences of the ligand on the metal depending on the protonation state of the central amine in the protonated **H1** *versus* **1** ligand. Comparison of these complexes with previous complexes and free **H1** revealed significant shifts in the bond angles, bond lengths, and ligand planarity that are discussed further. The corresponding 2+1 *fac*-$[^{99m}Tc^{I}(CO)_3(\mathbf{1})(L)]$ complexes were also prepared in moderate to good yields and their stability examined by amino acid challenge studies.

Keywords: Rhenium; Technetium; Carbonyl; 2+1

*Corresponding author. Email: bennyp@wsu.edu

Introduction

Group VII congeners, technetium and rhenium, have contributed significantly to the development of new radioactive compounds in nuclear medicine. The nuclear properties of this pair permits use of ^{99m}Tc [$t_{1/2}$ = 6.0 h, g = 140 keV (89%)] in clinical diagnostic imaging via single photon emission computed tomography and $^{186/188}$Re (186: $t_{1/2}$ = 3.7 d, β^-_{max} = 1.0 MeV, 188: $t_{1/2}$ = 17 h, β^-_{max} = 2.1 MeV) in imaging and therapy (theranostic) from medium-high-energy beta particles for tissue damage and gamma emissions for scintigraphy [1–6]. While a number of Tc and Re complexes have been prepared over the years, *fac*-$[M^I(OH_2)_3(CO)_3]^+$ (M = Re, ^{99m}Tc) has become an important precursor in the design of new radiopharmaceutical agents. The low-spin nature of the cationic d^6 of the M^I center and stabilization of the carbonyl ligands permits the ready substitution of the aquo ligands in the complex with a variety of ligands. The flexibility of the $M^I(CO)_3$ to accommodate different ligands has led to the development of several strategies to saturate the coordination sphere with ligands from a single ligand (tridentate) to a combination of two or more ligands [7].

In particular, the 2+1 approach provides a combination strategy of a bidentate and a monodentate ligand to yield a unique asymmetric tailoring of the complex. Either ligand or both can be utilized for incorporating a targeting vector for directed delivery of the radionuclide *in vivo*. The flexibility to exchange mono or bidentate ligands in the complex also permits facile modification of the overall chemical properties (e.g. charge, lipophilicity, and stability) to impact pharmacological properties (e.g. clearance, permeability, and half-life) of the complex. Using this 2+1 strategy, a variety of combination of bidentate ligand systems (e.g. picolinic acid, bipyridines, acetylacetone, dithionites, dithiocarbamates, and imidazole carboxylic acid) and monodentate ligands (e.g. pyridine, imidazole, phosphines, and isonitriles) have been reported. While many of these systems can be readily prepared radiochemically with ^{99m}Tc and characterized with Re in X-ray structures, the 2+1 strategy has generally suffered from poor labeling yields and *in vitro*/*in vivo* stability. Dissociation or displacement kinetics of the monodentate ligand has typically been considered responsible for the limited effectiveness. However, recent reports by our group and Valliant's group have indicated the overall stability with 2+1 complexes with bidentate ligands (e.g. bipyridine and picolinate) can be improved by changing the basicity of the monodentate pyridine ligands with functional groups [8–10].

These recent reports led us to re-evaluate another bidentate ligand, acetylacetone (acac), that is reacted with *fac*-$[M^I(CO)_3]^+$ to yield a deprotonated OO-bound aromatic neutral complex, *fac*-$[M^I(CO)_3(acac)(OH_2)]$. This acac complex has also been extensively studied in a 2+1 approach with monodentate ligands (e.g. imidazole, pyridine, isocyanides, cyanide and phosphines) [11–15], but still retains the limitations of 2+1 complexes. In addition, modification of acac to incorporate a biological targeting vector for directed *in vivo* delivery at the C1 and C3 positions can be synthetically challenging. Outside of reports using curcumin, a natural homolog of acac, for targeting β-amyloid plaques in Alzheimer's patients, limited functionalized acac systems have gained significant clinical attention with *fac*-$[M^I(OH_2)_3(CO)_3]^+$ [14, 16].

An alternative to acac, N,N-dialkylbenzoylthiourea ligands offer a similar coordination behavior to acac, but have modified characteristics. Both systems can be readily deprotonated upon complexation to yield a planar aromatic six-membered coordination ring with the metal center. A key difference from the hard oxygen donors in acac, N,N-dialkylbenzoylthioureas utilizes both a hard, O, and a soft, S, donor to increase electron donation to

the metal center. Benzoylthioureas have been utilized with Re(III) and Re(V) oxo and imido complexes [17–23]; however, only limited studies with the N,N-dialkylbenzoylthioureas with the *fac*-$[Re^{I}(CO)_3]^+$ core have been performed [24, 25]. Benzoylthioureas can be prepared easily and in high yields through reaction of a primary or secondary amine with an isothiocyanate containing compound [26, 27]. The synthetic flexibility presents a facile method for functionalization of the ligand to adjust the chemical properties of the complex or incorporate a targeting vector for directed delivery in radiopharmaceutical applications [28].

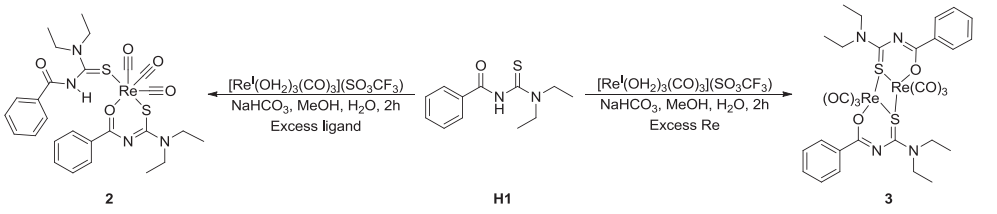
In this report, we examine the behavior of a series of 2+1 complexes with *fac*-$[M^{I}(CO)_3]^+$ (M = Re, ^{99m}Tc) and N,N-diethylbenzoylthiourea (HEt_2btu), **H1**. Neutral monodentate ligands (i.e. methyl imidazole (Me-imid), pyridine (py), cyclohexyl isocyanide (CyNC), or triphenylphosphine (PPh_3)) were added to saturate the *fac*-$[M^{I}(CO)_3]^+$ coordination sphere and generate an overall neutral complex. Complexes were characterized by NMR, high-resolution MS, IR, and single-crystal X-ray crystallography. The corresponding radioactive ^{99m}Tc complexes were also prepared in reasonable yields and further evaluated for stability using challenge assays (i.e. histidine and cysteine) to model *in vivo* behavior.

Results and discussion

Thiourea-based ligands have been widely investigated with a variety of metals, including rhenium and technetium [17–23, 29–39]. Organometallic complexes of Re have typically been prepared utilizing the starting precursors, $(NEt_3)_2[ReX_3(CO)_3]$ or $ReX(CO)_5$ (X = Cl, Br), for complexation with thiourea ligands in organic solvents. The halide often remains coordinated to the metal center balancing the charge of the complex with neutral mono or bidentate ligands [24, 25]. However, the behavior of rhenium does not necessarily mimic the behavior of the radiotracer ^{99m}Tc counterpart. Prepared under aqueous conditions from $^{99m}TcO_4^-$ using an Isolink® Kit in normal saline (0.9%) solution, the aquo species *fac*-$[Tc^{I}(OH_2)_3(CO)_3]$ is observed. In order to mitigate the effect of the halide coordinating in the Re species, *fac*-$[Re^{I}(OH_2)_3(CO)_3](SO_3CF_3)$ was utilized as the starting precursor for complexation and to more closely mimic the chemistry with ^{99m}Tc.

Synthesis of Et_2btu 2+1 fac-$[Re^{I}(CO)_3]^+$ complexes

The preparation of *fac*-$[Re^{I}(CO)_3]$ complexes with N,N-diethylbenzoylthiourea (HEt_2btu, **H1**) was examined as a function of ligand concentration with respect to the metal to replicate the high ligand conditions observed with tracer-level concentrations of ^{99m}Tc. Excess **H1** (two or more equivalents) was reacted with *fac*-$[Re^{I}(OH_2)_3(CO)_3](SO_3CF_3)$ to generate an unusual 2+1 complex *fac*-$[Re^{I}(CO)_3(Et_2btu)(HEt_2btu)]$, **2** (scheme 1). Single crystals of **2** obtained by slow cooling of a saturated methanolic solution were of sufficient quality for X-ray diffraction analysis. In the complex, the ligands exist in two different coordination modes, a bidentate oxygen and sulfur coordinated **1** as well as a monodentate **S** coordinated **H1**. Complex **2** differs from previously reported structures obtained from $Re(CO)_5Br$, which contained two S-bound monodentate ligands with the bromide [24]. A similar product, however, was observed with thiocarbamoylbenzamidines [40]. MS analysis of **2** showed the parent ion at 743.0 *m/z* as well as dissociation of the complex into HEt_2btu at 259.1 *m/z* (**H1** + Na) and *fac*-$[Re^{I}(OH_2)(CO)_3(Et_2btu)]^+$ at 524.5 *m/z* or *fac*-$[Re^{I}(CO)_3(Et_2btu)]$ at



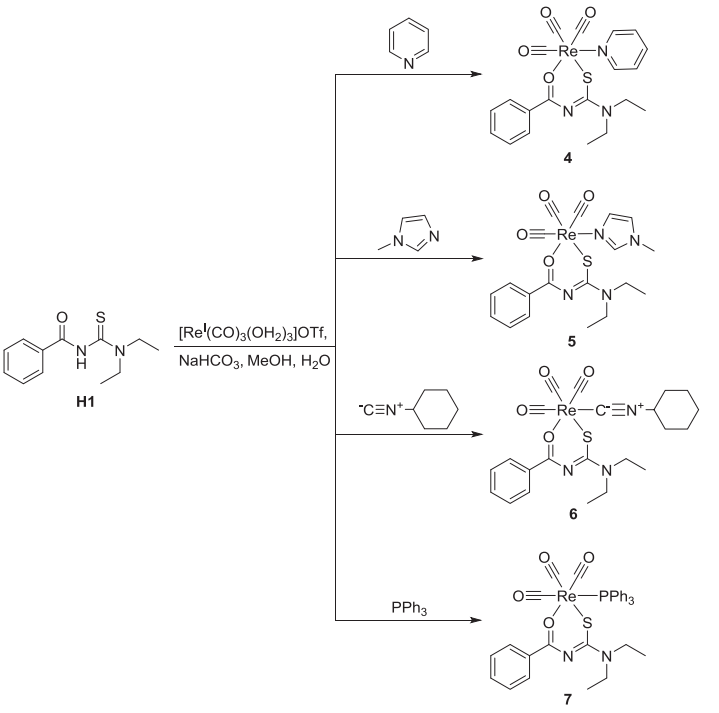
Scheme 1. Synthesis of *fac*-[Re(CO)$_3$(**1**)(**H1**)], **2**, and of *fac*-[Re(CO)$_3$(μ-**1**)]$_2$, **3**.

507 *m*/*z* with no associated water. ^{1}H NMR analysis of **2** also confirmed the presence of two distinct coordination modes of HEt$_2$btu through the slight shifts in splitting patterns observed in the spectrum. Both the **H1** and **1** ligands exhibit similar chemical shifts with some distinct differences. The aromatic shifts are different between the two ligands due to the lack of coordination to O of **H1**. Another indicator of the coordination of two benzoylthioureas was the presence of a singlet at 10.18 ppm, corresponding to the highly acidic central amide proton in the protonated **H1**. Sufficient base had been added to the reaction mixture to completely deprotonate the nitrogen of both **H1** ligands; however, only deprotonation of one of the two ligands was observed. This suggests formation of a neutral complex is preferred over the formation of a charged complex regardless to the presence of excess base.

Utilizing excess *fac*-[Re^I(OH$_2$)$_3$(CO)$_3$](SO$_3$CF$_3$) with **H1** in a basic water/methanol solution gave a poorly soluble yellow solid. NMR analysis of the crude solid prior to purification indicated the existence of multiple species (e.g. *fac*-[Re^I(CO)$_3$(Et$_2$btu)(X)] (X = OH$_2$, OH, or **H1**). Purification of the solid by basic alumina column chromatography yielded a rather unusual μ-disulfur dimer *fac*-[Re^I(CO)$_3$(Et$_2$btu)]$_2$, **3**, in good yield (scheme 2). The product was recrystallized by slow evaporation from a chloroform/heptane solution yielding pale yellow crystals suitable for X-ray diffraction analysis. MS analysis of **3** showed the presence of the dimeric complex at 1034.9 *m*/*z* (M + Na) or the ionized monomeric species similar to those observed with **2**. ^{1}H NMR showed a single product deprotonated at the central nitrogen on both ligands correlating with the splitting pattern observed for the bidentate ligand in **2** but slightly shifted.

To improve the overall reaction yields, a one-pot approach was developed to directly prepare the 2+1 complexes from *fac*-[Re^I(OH$_2$)$_3$(CO)$_3$](SO$_3$CF$_3$) with **H1** and a series of neutral monodentate ligands. The general synthesis involved stepwise addition of a single equivalent of **H1** with a slight excess of *fac*-[Re^I(OH$_2$)$_3$(CO)$_3$](SO$_3$CF$_3$) in a water/methanol solution in the presence of NaHCO$_3$ at room temperature followed by the addition of the monodentate ligand (py, Me-imid, CyNC, or PPh$_3$) to the reaction mixture (scheme 2). The corresponding 2+1 complexes, *fac*-[Re^I(CO)$_3$(Et$_2$btu)(py)], **4**, *fac*-[Re^I(CO)$_3$(Et$_2$btu)(Me-imid)], **5**, *fac*-[Re^I(CO)$_3$(Et$_2$btu)(CyNC)], **6**, and *fac*-[Re^I(CO)$_3$(Et$_2$btu)(PPh$_3$)], **7**, were prepared in poor to moderate yields after purification by column chromatography. While the precursor had extremely poor solubility, the isolated 2+1 complexes were highly soluble in a wide range of polar and non-polar organic solvents (e.g. methanol, diethyl ether, benzene, chloroform, and ethyl acetate), which was not previously observed with similar [Re^I(CO)$_3$(acac)(L)] 2+1 complexes [12, 15].

The ^{1}H NMR of each of the 2+1 complexes, **4–7**, showed restricted rotation of the two ethyl groups. Restricted rotation of the N substitution of these classes of complexes has been studied repeatedly and is evident in both the free ligand and metal complexes of



Scheme 2. Synthesis of *fac*-$[Re^{I}(CO)_3]^+$ 2+1 complexes with **H1**.

benzoylthioureas. Barriers to rotation for **H1** have been shown to be 43.3 J mol^{-1} giving a shift of 43.3 Hz (~0.14 ppm) [41, 42]. Larger chemical shifts have also been observed due to coordination of the metal to the free ligand. The restricted rotation resulted in two triplets with separation of 0.1–0.2 ppm corresponding to terminal methyl groups. Peaks had baseline separation except in the case of cyclohexyl isocyanide derivative, **6**, due to overlap with the cyclohexyl ring. Variance in this splitting pattern may be due to the planarity of the complex or formation of an aromatic ring involving the metal center. The methylene groups adjacent to the nitrogen were separate due to restricted rotation and were split into multiplets rather than the expected quartets. No peak was observed corresponding to N protonation as was observed in **2**, indicating coordination of the ligand in the deprotonated Et_2btu form.

MS analysis of the 2+1 complexes showed parent ion peaks for **4–7**. Complex **4** showed a parent ion peak (M + Na) of 607.9 as well as peaks corresponding to dissociation of the py ligand and gain of water at *m*/*z* 524.8, corresponding to *fac*-$[Re^{I}(OH_2)(CO)_3$(**1**)]. Me-imid coordinated **5** showed only the parent ion peak at 611.0 *m*/*z* (M + Na) and no dissociation of the monodentate ligand. MS of **6** showed a parent ion peak at 638.0 *m*/*z* (M + Na) as well as a significant peak corresponding to loss of CO from the complex at 587.9 *m*/*z*. Complex **7** primarily gave the parent ion peak at 790.9 *m*/*z* (M + Na); however, a peak corresponding to the loss of CO was present at 763.1 *m*/*z*. In lieu of elemental analysis, high-resolution MS was also used to confirm the identity of complexes and gave results within <5 ppm.

X-ray crystallographic characterization

Single crystals of **2**, **3,** and **4** were obtained and analyzed by X-ray diffraction analysis. Complete experimental parameters and tables of bond angles and distances for each of these compounds can be found in the Supplementary material. An abridged version of the relevant bond angles and distances describing the interactions of Re and **1** can be found in tables 1 and 2. In the structures of **2** and **4**, the $[Re^I(CO)_3]^+$ core maintained the facial geometry with slightly distorted bond angles and distances of the Re carbonyl bond similar to previously observed Re analogs [25]. The most significant structural differences were observed in the interaction of **1** with the metal center as independently discussed below with each complex.

Crystals of **2** were obtained by slow cooling of a saturated methanolic solution that crystallized in a monoclinic P 2_1/n space group with one molecule in the asymmetric unit (figure 1). The structure of **2** indicated a composite 2+1 complex occupying the facial positions opposite to the carbonyl ligands through incorporation of two asymmetrically bound molecules of HEt_2btu. One ligand was observed in a bidentate OS coordination mode and the second molecule through monodentate S. The structural solution indicated the absence of additional counterions in the unit cell suggesting the overall charge of the complex was neutral. Correlating with NMR data, deprotonation of the central nitrogen of the bidentate ligand exhibits a more planar coordination due to the aromatization gained upon metal coordination. This behavior correlates with the analogous *fac*-$[Re^I(CO)_3]^+$ acac complexes that undergo a similar deprotonation upon complexation [12, 15, 43]. While both the bidentate acac and OS coordinated benzoylthiourea in **2** have six-membered coordination rings, the O–Re–O (84.5°) in the acac complexes is more constrained than the O1–Re1–S1 bite angle (88.5°) in **2**. While it would be predicted the deprotonated bidentate **1** would be relatively planar, steric interactions of the bulky aromatic rings appear to impact the torsional twist of the bidentate ligand (figure 1, inset). The second **H1** ligand in **2** is found as an S-bound monodentate ligand with a Re1–S2 bond length of 2.5236(8) Å, which is typical

Table 1. Bond lengths obtained from crystal structures of **H1** [45], *fac*-$[Re^I(CO)_3$(**H1**)Br] [25], **2**, **3,** and **4**.

	Bond lengths (Å)						
Complex	C2–O1	C2–N2	N1–N2	C1–S1	C1–N1	Re1–O1	Re1–S1
H1	1.2188(15)	1.3869(15)	1.4183(15)	1.6767(12)	1.3258(15)	–	–
fac-$[Re^I(CO)_3$(**H1**)Br]	1.285(10)	1.410(10)	1.349(12)	1.709(9)	1.326(11)	2.142(8)	2.466(2)
2	1.301(3)	1.290(3)	1.349(3)	1.739(3)	1.325(3)	2.1694(18)	2.5236(8)
3	1.268(3)	1.323(3)	1.326(3)	1.787(2)	1.326(3)	2.1487(17)	2.5017(8)
4	1.273(3)	1.315(4)	1.353(4)	1.722(3)	1.372(5)	2.1189(19)	2.4475(8)

Table 2. Bond angles obtained from crystal structures of **H1** [45], *fac*-$[Re^I(CO)_3$(**H1**)Br] [25], **2**, **3,** and **4**.

	Bond angles (°)					
Complex	O1–C2–N2	C1–N2–C2	N2–C1–S1	Re–S1–C1	O1–Re1–S1	Re1–O1–C2
H1	122.07	120.52	119.76	–	–	–
fac-$[Re^I(CO)_3$(**H1**)Br]	121.66	127.93	123.07	106.75	82.04	131.41
2	127.53	126.51	122.94	103.59	88.47	122.18
3	128.3	127.6	125.49	100.36	85.30	132.42
4	130.87	126.42	129.18	109.48	87.05	132.57

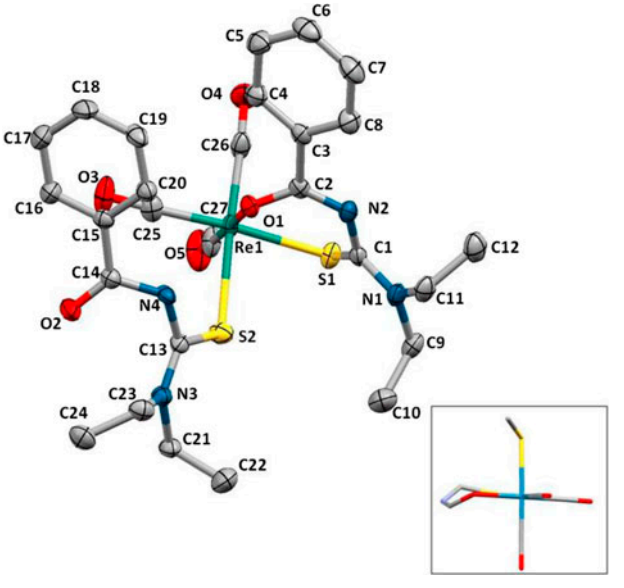


Figure 1. Labeled molecular structure of **2** with ellipsoids drawn at the 50% probability level.
Note: Hydrogens have been omitted for clarity.

of a Re–S bond [44]. Coordination of Re to sulfur in **H1** had a bond angle for Re1–S2–C13 (112.09°) that was consistent with the bonding mode of a thiourea. A minor lengthening of the S2–C13 double bond (1.701(3) Å) compared to the free ligand (1.6767 Å) was also observed [45]. The corresponding thiourea bond angles (S2–C13–N3 122.36°, S2–C13–N4 118.49°) of **H1** were also slightly shifted from the bond angles of the free ligand (124.46° and 119.76°, respectively). The orientation of the coordinated **H1** in **2** had a similar rotational twist and bond angles of the central nitrogen as the protonated-free ligand, which also correlates with the protonation of the monodentate ligand to generate the neutral charge of **2**. Despite the steric interactions of the alkyl and aryl substituents between **H1** and **1**, the complex appears to be stabilized by the presence of a hydrogen bond at O1–N4 (2.900 Å) between the ligands.

Crystals of **3** suitable for X-ray diffraction analysis were obtained by slow evaporation of a solution of chloroform/heptane and pack in the monoclinic P 2_1/n space group (figure 2). The dimer structure is composed of two distorted octahedral *fac*-[$Re(CO)_3$(**1**)] subunits with two μ-S thioureas bridging the metal centers similar to reported complexes obtained from N,N-diphenylbenzoylthiourea with Re and thiocarbamoylbenzamidines with ^{99}Tc [24, 40]. The dimer core is an asymmetric rectangular parallelogram, where the edges consist of Re–S bonds of differing lengths from the bidentate Re1–S1 bond (2.5017(8) Å) and the monodentate Re1–S1' (2.5406(8) Å) bond of the second subunit. The asymmetry of the distorted octahedron and Re–S bond lengths produce a more constricted bond angle on the S1–Re1–S1' (83.15(2)°) compared to the bridging thiourea in Re1–S1–Re1' (96.85°). While similar Re_2S_2 dimers have been observed with thiols, SH^- and S^{2-} [46–48], few examples have been reported incorporating Re or ^{99m}Tc with μ-sulfur similar to **3** with thioureas [25, 40]. While the bridging of the thiourea appears to impact the planarity of the

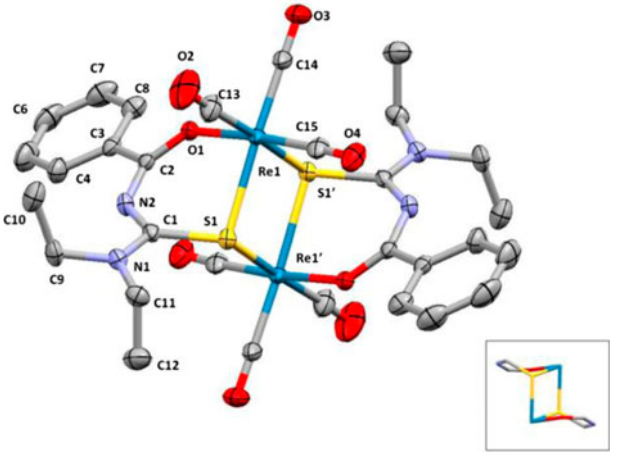


Figure 2. Labeled molecular structure of **3** with ellipsoids drawn at the 50% probability level.
Notes: Hydrogens have been omitted for clarity. Bond distances (Å): Re1–S1' 2.5406(8), S1–Re1' 2.5407(8). Bond angles (°): O1–Re1–S1' 78.01(5), S1–Re1–S1' 83.15(2), Re1–S1–Re1' 96.85(2).

bidentate **1**, N2 remains deprotonated in an sp^2 hybridization due to the reorientation of the thiourea donor to accommodate the μ-bridge (figure 2, inset).

Crystals of **4** suitable for X-ray diffraction analysis were obtained by slow diffusion of hexanes into a solution of ethyl acetate. The crystals packed in the monoclinic C 2/c space group with one molecule per asymmetric unit. The structure yielded the expected 2+1 complex consisting of the deprotonated bidentate **1** ligand and the monodentate py (figure 3). The Re1–N3 bond length (2.213(2) Å) of **4** was consistent with other Re–py bond distances observed in 2+1 complexes with *fac*-$[Re^{I}(CO)_3]^+$ [15]. Consistent with **2**, the absence of a

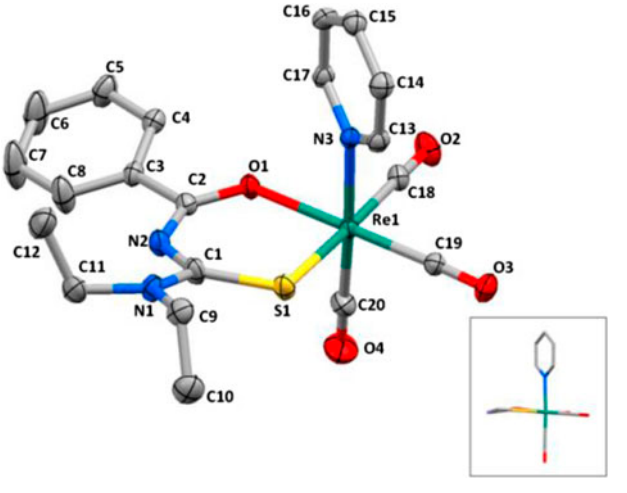


Figure 3. Labeled molecular structure of **4** with ellipsoids drawn at the 50% probability level.
Note: Hydrogens have been omitted for clarity.

counterion in the unit cell was also observed indicating deprotonation of the central nitrogen in coordinated **1**. Slight differences are observed in **4** as the O1–Re1–S1 bond angle (87.0°) is slightly more pinched than in **2**. A substantial structural variation is the increased planarity of **1** with py as the monodentate ligand in **4** (figure 3, inset). The difference in planarity of **1** observed in **2** is postulated to be related to the steric interactions of the benzyl groups of the two **H1** ligands contributing to the out of plane bending of the coordination ring. In contrast, the py has minimal steric interactions with **1**, suggesting the bulkiness of the monodentate ligand may contribute to the formation and stability of the overall complex.

The protonation state of bidentate **1** revealed the most striking structural differences in the bond lengths, angles, and planarity in the complexes compared to the free ligand (tables 1 and 2). Several distinctions can be highlighted between these two types of complexes based on the protonation state of the nitrogen. In the deprotonated complexes, **2**, **3,** and **4**, the aromaticity of the ligand alters the overall bond angles and geometry of the coordination ring. The negative charge on the amide nitrogen significantly shortens the C2–N2 bond in **2–4** (1.290(3)–1.323(3) Å) to increase double bond character closer to an imine. This also correlates with an elongation of the C2–O1 carbonyl in **2–4** (1.268(3)–1.301(3) Å) due to the electron withdrawing effect of the metal compared to the free ligand (1.2188 Å). Similar bond shortening and lengthening can also be observed in the thiourea donor. The C1–N2 are also shortened in **2–4** (1.326(3)–1.349(3) Å) as compared to the free ligand (1.4183 Å); however, this bond does not shorten to the degree observed with C2–N2. The C1–S1 thiourea bond is similarly elongated in **2–4** (1.722(3)–1.787(2) Å). The degree of bond lengthening of the thiourea is significantly smaller than the carbonyl suggesting that while the system appears aromatic, the coordinated ligand has a degree of asymmetry as denoted in table 1. There is no significant change observed in the C1–N1 bond length between **2** and **3** from **H1** (~1.325 Å); however, a significant elongation of the C1–N1 bond length (1.372(5) Å) was observed in **4**, indicating increased aromaticity mimicking an sp^2 hybridized aromatic tertiary amine compared to a typical thiourea [49]. Overall, **1** in **4** shows a greater degree of aromatic character in comparison to **2** and **3** due to the relative planarity of the ligand to the *fac*-$[Re^{I}(CO)_3]^+$ core.

In comparison, a structural representation of the protonated *fac*-$[Re^{I}(CO)_3$(**H1**)Br] complex illustrates the differences in coordination orientation and planarity of the protonated bidentate **H1** reported by Abram (figure 4) [25]. Contrary to the deprotonated complexes, the C2–N2 bond (1.410 Å) was observed at a slightly longer distance than even the free ligand (1.3869 Å). The corresponding O1–C2–N2 bond angle (121.66°) was very similar to free ligand (122.07°) but was more constrained in comparison to **2–4** (127.53–130.87°). Coordination of **H1** in *fac*-$[Re^{I}(CO)_3$(**H1**)Br] had a similar increase in distances for the adjacent bonds. Re1–S1 (2.466 Å) and Re1–O1 (2.142 Å) were comparable to Re1–S1 (2.4475(8)–2.5236(8) Å) and Re1–O1 (2.1189(19)–2.1694(18) Å) of **2–4**. The protonated ligand had a similar elongation of C2–O1 (1.285 Å) as for **2–4**, while the C1–S1 bond (1.709 Å) was intermediate between the free ligand and the deprotonated complex. Overall, **H1** in *fac*-$[Re^{I}(CO)_3$(**H1**)Br] exhibited significant out of plane bending relative to the octahedral geometry of the metal (figure 4, inset). The O1-Re1–S1 bite angle in *fac*-$[Re^{I}(CO)_3$(**H1**)Br] was significantly more constricted (82.04°) compared to deprotonated complexes (85.3–88.5°). The constriction and out-of-plane orientation of **H1** may be directly related to the protonated central nitrogen and bent bonding modes of the carbonyl and thiourea.

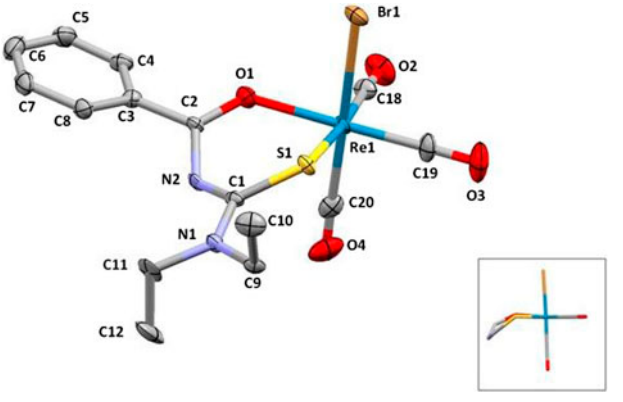
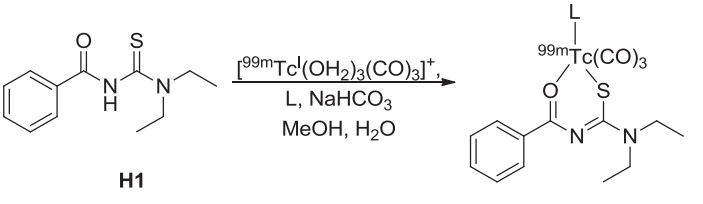


Figure 4. Labeled molecular structure of *fac*-[$Re^I(CO)_3$(**H1**)Br] with ellipsoids drawn at the 30% probability level.
Note: Hydrogens have been omitted for clarity [25].

fac-[$^{99m}Tc^I(CO)_3]^+$ Synthesis and stability

The corresponding radioactive 2+1 ^{99m}Tc complexes (**4a**–**7a**) were prepared in a two-step one-pot reaction mixture using the *fac*-[$^{99m}Tc^I(OH_2)_3(CO)_3]^+$ precursor. In the first step, the complexation of the bidentate **H1** was accomplished by heating the bidentate ligand solution (10^{-4} M) with the *fac*-[$^{99m}Tc^I(CO)_3]^+$ species in water/methanol with sodium bicarbonate for 30 min to generate a ^{99m}Tc intermediate complex containing **1** (scheme 3). While the exact speciation of the intermediate is uncertain, it is proposed to be either, *fac*-[$^{99m}Tc^I(CO)_3$(**1**)(X)] (X = OH_2 or H1), with either a water or a second benzoylthiourea ligand coordinated, based on the behavior of Re complexes with excess ligand. The addition of the monodentate ligand in methanol to the intermediate solution yielded the corresponding ^{99m}Tc 2+1 complexes **4a**–**7a** in good to excellent yields (table 3). Peak retention times observed for the ^{99m}Tc complexes correlated with the analogous Re complexes. Aromatic amine 2+1 complexes, *fac*-[$^{99m}Tc^I(CO)_3$(**1**)(py)], **4a**, and *fac*-[$^{99m}Tc^I(CO)_3$(**1**)(Me-imid)], **5a**, were obtained in reasonable yields of 70 and 94%, utilizing 10^{-2} M solutions of py and Me-imid, respectively. Utilizing CyNC and PPh_3 monodentate ligands, the corresponding *fac*-[$^{99m}Tc^I(CO)_3$(**1**)(CyNC)], **6a**, and *fac*-[$^{99m}Tc^I(CO)_3$(**1**)(PPh_3)], **7a**, complexes were prepared



H1

4a L = py
5a L = Me-imid
6a L = CyNC
7a L = PPh_3

Scheme 3. Synthesis of *fac*-[$^{99m}Tc^I(CO)_3]^+$ complexes with **1**.

Table 3. Yields for 2+1 complexes of **H1** with *fac*-[^{99m}Tc^{I}(CO)$_3$]$^+$. Reactions were complexed at 10^{-4} M total concentration of **H1** for 30 min followed by reaction with monodentate ligand at room temperature.

Ligand	Conc. ligand (M)	Time (min)	Complex	Yield (%)
Py	10^{-2}	30	**4a**	70
Me-imid	10^{-2}	30	**5a**	94
CyNC	10^{-4}	30	**6a**	90
PPh_3	10^{-4}	30	**7a**	81

in good yields of 90 and 81%, respectively. These yields were obtained at significantly lower monodentate ligand concentrations (10^{-4} M) than the aromatic amine ligands.

Stability studies were conducted on purified 2+1 products **4a**–**7a** isolated from the radio-HPLC to examine the effect of dissociation of the complex. The effect of the monodentate and overall complex stability was tested by a competitive amino acid challenge assay. Aliquots of purified **4a**–**7a** were incubated in 1-mM solutions of histidine or cysteine in pH 7.4 phosphate buffer at 37 °C and analyzed by radio-HPLC at 1 and 4 h (table 4). In general, the 2+1 complexes showed limited to poor stability under the conditions of the challenge assays. At 1 h, significant degradation was observed in most of the samples. While the aromatic amines had poor stability, the pyridine complex **4a** was slightly more stable than the methyl imidazole complex **5a** under both histidine and cysteine. However, at 4 h only the pyridine complex had limited stability in histidine (27%), while **5a** was with either amino acid. The isonitrile (CyNC) complex **6a** exhibited the highest stability of the monodentate ligands examined in histidine (88%) and cysteine (53%) at 1 h, with a modest decomposition to histidine (73%) and cysteine (31%) at 4 h. Complexes **4a**-**6a** appeared slightly more stable under challenge with histidine than cysteine. Surprisingly, 2+1 complex **7a** with PPh_3 showed lack of stability in both challenge assays. At 1 h, less than 10% of the complex could be observed with both cysteine and histidine. This behavior with PPh_3 was unexpected as phosphine ligands typically exhibit strong binding to the *fac*-[^{99m}Tc^{I}(CO)$_3$]$^+$ core as observed with other ^{99m}Tc 2+1 [14, 50]. In **7a**, steric interactions of the large cone angle of PPh_3 coupled with phenyl rings of **1** may have contributed to destabilization of the complex that might be avoided using phosphine ligands with a smaller cone angle.

Table 4. Stability of *fac*-[^{99m}Tc^{I}(CO)$_3$]$^+$ complexes **4a**–**7a** under amino acid challenge (1-mM cysteine or histidine, 10-mM pH 7.4 phosphate buffer, 37 °C) conditions.

Complex		1 h	4 h
4a	His	44%	27%
	Cys	16%	16%
5a	His	15%	5%
	Cys	15%	2%
6a	His	88%	73%
	Cys	53%	31%
7a	His	9%	–
	Cys	ND	–

Conclusion

N,N-diethylbenzoylthiourea (**H1**) was utilized as a model system to explore coordination with both *fac*-$[M^I(OH_2)_3(CO)_3]^+$ (M = Re, ^{99m}Tc) and assess the potential for a 2+1 complex. The concentration of **H1** relative to the metal impacts the formation of the complex observed. Excess **H1** yielded a complex with two coordinated ligands, while equimolar equivalents of ligand relative to the metal yielded a mixture of insoluble complexes including an unusual μ-sulfur dimer. In situ, addition of a monodentate ligand (i.e. py, Me-imid, CyNC and PPh_3) modestly improved the overall solubility and behavior of the Re analogs. The corresponding ^{99m}Tc complexes were readily prepared in moderate to high yields; however, stability issues were also observed with these complexes. The 2+1 cyclohexyl isocyanide complex exhibited the highest stability over the aromatic amines or phosphine in both Re and ^{99m}Tc. While benzoylthioureas represent a versatile system for ligand design, low yields and stability issues presented significant limitations for utilization of this ligand platform in a 2+1 strategy with *fac*-$[M^I(CO)_3]$ core in radiopharmaceutical applications.

Experimental

Materials and methods

All reagents and organic solvents of reagent grade or better were used as purchased from Aldrich, Acros or Fluka without purification. Rhenium starting material *fac*-$[Re^I(OH_2)_3(CO)_3](SO_3CF_3)$ was prepared by literature methods from $Re_2(CO)_{10}$ purchased from Strem [51, 52]. ^{99m}Tc was obtained in the form of Na[$^{99m}TcO_4$] from Cardinal Health (Spokane, WA) and the *fac*-$[^{99m}Tc^I(OH_2)_3(CO)_3]^+$ complex was prepared using a commercially available Isolink® kit from Covidien. **H1** was synthesized as previously described [26]. ^{1}H and ^{13}C NMR spectra were recorded on a Varian 300 MHz instrument at 25 °C in $CDCl_3$. FT-IR spectra were obtained on a Thermo Nicolet 6700 FT-IR with an ATR cell and analyzed with OMNIC 7.1 software. Mass spectra were obtained on a Thermo Finnigan LCQ Advantage ESI-MS. High-resolution mass spectra were taken on a Waters Q-Tof Premier quadrupole–time-of-flight mass spectrometer. Separation and identification of compounds were conducted on a Perkin Elmer Series 200 high pressure liquid chromatography (HPLC) equipped with a UV/vis Series 200 detector and a Radiomatic 610TR detector. Utilizing a Varian Pursuit XRs 5 μm particle and 250 × 4.6 mm C-18 column, the compounds were separated with a reverse phase gradient system beginning with a 2-mM pH 7.4 phosphate buffer aqueous phase and shifting gradually to methanol. HPLC analysis was performed using 0–3.0 min (100% phosphate), 3.0–9.0 min (75% phosphate, 25% MeOH), 9.0–20.0 min (25–100% MeOH linear gradient) and 20.0–30.0 min (100% MeOH) at a flow rate of 1.0 mL min^{-1}.

Synthesis of fac-[$Re^I(CO)_3(Et_2btu)(HEt_2btu)$], 2

H1 (50 mg, 0.210 mmol) was dissolved in MeOH (10 mL). A 0.1-M solution of *fac*-$[Re^I(OH_2)_3(CO)_3](SO_3CF_3)$ (2 mL, 0.200 mmol) and 1-M $NaHCO_3$ (300 μL, 0.300 mmol) were added and the reaction was stirred for 1 h. Additional MeOH (10 mL) was added until all precipitate redissolved to give a yellow solution. After 1 h, H_2O was added until a white

precipitate began to form and the solution was left at 5 °C overnight. The resulting precipitate was collected by filtration, washed with cold 1 : 1 MeOH/H_2O, and dried in vacuo. The resulting solid was recrystallized to give **2** as a yellow crystal (21.8 mg, 29%). Single crystals of **2** were obtained from slow cooling of a saturated solution in MeOH. ^{1}H NMR ($CDCl_3$, 300 MHz) δ 1.20 (t, 6H, J = 7.1 Hz), 1.37 (q, 6H, J = 7.4 Hz), 3.38–3.64 (m, 3H), 3.80–4.20 (m, 5H), 7.36 (q, 4H, J = 7.7 Hz), 7.50 (t, 1H, J = 6.3 Hz), 7.58 (t, 1H, J = 6.6 Hz), 7.75 (d, 2H, J = 8.2 Hz), 7.90 (d, 2H, J = 8.0 Hz), 10.18 (s, 1H); ^{13}C NMR ($CDCl_3$, 75 MHz) δ 12.0, 13.0, 13.1, 13.9, 45.7, 47.3, 48.3, 48.9, 128.3, 128.9, 129.0, 129.8, 131.8, 132.2, 133.4, 136.9, 164.7, 170.0, 176.8, 181.0, 192.4, 193.5, 194.3; IR: 2014, 1893, 1700 cm^{-1}; MS (m/z): $[M + H]^+$ 743.0. HRMS (ESI+) Calcd for $C_{26}H_{31}N_4O_4ReS_2$ requires m/z 741.1344 [M + H], found m/z 741.1355 (1.4 ppm).

Synthesis of fac-[Re^I(CO)$_3$(Et$_2$btu)]$_2$, 3

To a solution of **H1** (60 mg, 0.254 mmol) in 10 mL of MeOH, a 0.1-M solution of *fac*-[Re^I(OH$_2$)$_3$(CO)$_3$](SO$_3$CF$_3$) (2.8 mL, 0.280 mmol) was added. 1-M $NaHCO_3$ (381 µL, 0.381 mmol) was added and the solution was stirred at room temperature for 3 h. H_2O was added to the reaction mixture and the solution was cooled at 5 °C overnight. The resulting precipitate was collected by filtration, washed with 1 : 1 cold MeOH/H_2O, and dried in vacuo. The resulting solid was purified by column chromatography on basic alumina with hexanes/DCM (1 : 1) to give **3** as a yellow solid (26 mg, 51%). Complex **3** was then recrystallized by slow evaporation of a chloroform/heptane solution to give the product as pale yellow plates. ^{1}H NMR ($CDCl_3$, 300 MHz) δ 1.35 (t, 6H, J = 6.96 Hz), 1.39 (t, 6 H, J = 7.1 Hz), 3.68–3.62 (m, 2H), 4.01–3.88 (m, 4H), 4.23–4.18 (m, 2H), 7.57–7.45 (m, 6H), 8.30 (d, 4H, J = 7.1 Hz); ^{13}C NMR ($CDCl_3$, 75 MHz) δ 13.3, 14.0, 47.1, 47.5, 128.2, 130.4, 132.6, 136.9, 169.4, 173.5, 191.5, 192.6, 194.9; IR: 2012, 1921, 1891 cm^{-1}; MS (m/z): $[M + Na]^+$ 1034.9. HRMS (ESI+) Calcd for $C_{30}H_{30}N_4O_8Re_2S_2$ requires m/z 1009.0643 [M + H], found m/z 1009.0686 (4.2 ppm).

Synthesis of fac-[Re^I(CO)$_3$(Et$_2$btu)(py)], 4

To a solution of **H1** (40 mg, 0.169 mmol) in 4 mL of MeOH, a 0.1-M solution of *fac*-[Re^I(OH$_2$)$_3$(CO)$_3$](SO$_3$CF$_3$) (2 mL, 0.200 mmol) was added. 1-M $NaHCO_3$ (406 µL, 0.406 mmol) was added and the solution was stirred at room temperature for 6 h. Pyridine (21.9 µL, 0.271 mmol) and MeOH (15 mL) were added and the solution was stirred for an additional 2 h. H_2O was added to the reaction to induce precipitation of the solid and the reaction flask was placed at 5 °C overnight. The resulting yellow solid was collected by filtration, washed with cold 1 : 1 MeOH/H_2O, dried in vacuo, and purified by column chromatography on basic alumina with hexanes/DCM (3 : 2) to yield **4** (41 mg, 41%) as a yellow solid. Complex **4** was recrystallized by slow diffusion of hexanes into an ethyl acetate solution. ^{1}H NMR ($CDCl_3$, 300 MHz) δ 1.10 (t, 3H, J = 7.1 Hz), 1.29 (t, 3H, J = 7.1), 3.73–3.81 (m, 2H), 3.91 (q, 2H, J = 7.1 Hz), 7.27–7.49 (m, 5H), 7.78 (t, 1H, J = 6.3 Hz), 8.08 (d, 2 H, J = 6.9 Hz), 8.82 (d, 2H, J = 6.3 Hz); ^{13}C NMR ($CDCl_3$, 75 MHz) δ 12.9, 13.4, 45.2, 46.6, 125.3, 128.1, 129.4, 131.5, 137.4, 138.1, 153.1, 170.1, 176.1; IR: 2009, 1890, 1861 cm^{-1}; MS (m/z): $[M + Na]^+$ 607.9. HRMS (ESI+) Calcd for $C_{20}H_{20}N_3O_4ReS$ requires m/z 584.0783 [M + H], found m/z 584.0808 (4.2 ppm).

Synthesis of fac-[$Re^I(CO)_3$(Et_2btu)(Me-imid)], 5

To a solution of **H1** (40 mg, 0.169 mmol) in 4 mL of MeOH a 0.1-M solution of *fac*-[$Re^I(OH_2)_3(CO)_3$](SO_3CF_3) (2 mL, 0.200 mmol) was added. 1-M $NaHCO_3$ (406 μL, 0.406 mmol) was added and the solution was stirred at room temperature for 6 h. 1-Methylimidazole (0.020 mL, 0.271 mmol) and MeOH (15 mL) were added and the solution was stirred for an additional 18 h. Water was added to the reaction to induce precipitation of the solid and the reaction flask was placed at 5 °C overnight. The reaction was filtered, washed with cold 1 : 1 MeOH/H_2O, and the precipitate was dried in vacuo. The resulting solid was purified by basic alumina column chromatography with hexanes/DCM (3 : 2) and concentrated to give **5** as a yellow solid (23 mg, 23%). ^{1}H NMR ($CDCl_3$, 300 MHz) δ 1.21 (t, 3H, J = 7.0 Hz), 1.32 (t, 3H, J = 7.0 Hz), 3.62 (s, 3H), 3.65–3.76 (m, 1H), 3.83–4.06 (m, 3H), 6.77 (t, 1 H, J = 1.5 Hz), 7.23 (t, 1 H, J = 1.5 Hz), 7.33–7.38 (m, 2 H), 7.42–7.47 (m, 1 H), 7.71 (s, 1 H), 8.05 (d, 2 H, J = 7.0 Hz); ^{13}C NMR ($CDCl_3$, 75 MHz) δ 13.0, 13.5, 34.4, 45.2, 46.6, 120.9, 128.0, 129.4, 131.3, 137.7, 140.0, 170.2, 176.7; IR: 2009, 1890, 1870 cm^{-1}; MS (*m*/*z*): $[M + Na]^+$ 611.0. HRMS (ESI+) Calcd for $C_{19}H_{21}N_4O_4ReS$ requires *m*/*z* 587.0892 [M + H], found *m*/*z* 587.0895 (0.5 ppm).

Synthesis of fac-[$Re^I(CO)_3$(Et_2btu)(CyNC)], 6

To a solution of **H1** (52 mg, 0.220 mmol) in 4 mL of MeOH was added a 0.1-M solution of *fac*-[$Re^I(OH_2)_3(CO)_3$](SO_3CF_3) (2 mL, 0.200 mmol). 1-M $NaHCO_3$ (0.240 mL, 0.240 mmol) was then added and the solution was stirred at room temperature for 6 h. Cyclohexyl isocyanide (33 mg, 0.300 mmol) and MeOH (20 mL) were added and the solution was stirred for an additional 18 h. Water was added to the reaction mixture to further precipitate the product and the mixture was allowed to sit overnight at −20 °C. The mixture was filtered and washed with cold 1 : 1 MeOH/H_2O. The sticky solid was redissolved in Et_2O and washed with 1 M $NaHCO_3$. The organic layer was then dried with $MgSO_4$ and concentrated in vacuo. The resulting solid was purified by basic alumina column chromatography with hexanes/diethyl ether (9 : 1) and concentrated to give **6** as a brown oil (17 mg, 16%). ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.24–1.37 (m, 11H), 1.64–1.76 (m, 6H), 3.71–3.78 (m, 1H), 3.80–3.85 (m, 1H), 3.90–4.04 (m, 3H), 7.38 (q, 2H, J = 6.6 Hz), 7.45 (t, 1H, J = 7.7 Hz), 8.06 (d, 2H, J = 7.2 Hz);^{13}C NMR (75 MHz, $CDCl_3$) δ 13.2, 13.7, 22.4, 25.0, 32.3, 45.5, 46.7, 54.2, 128.1, 129.5, 137.6, 164.7, 172.0, 176.7, 190.4, 192.3, 193.1; IR: 2193, 2017, 1928, 1894 cm^{-1}; MS (*m*/*z*): $[M + Na]^+$ 638.0. HRMS (ESI+) Calcd for $C_{22}H_{26}N_3O_4ReS$ requires *m*/*z* 614.1252 [M + H], found *m*/*z* 614.1268 (2.6 ppm).

Synthesis of fac-[$Re^I(CO)_3$(Et_2btu)(PPh_3)], 7

To a solution of **H1** (52 mg, 0.220 mmol) in 10 mL of MeOH was added a 0.1-M solution of *fac*-[$Re^I(OH_2)_3(CO)_3$](SO_3CF_3) (2 mL, 0.200 mmol). 1-M $NaHCO_3$ (0.240 mL, 0.240 mmol) was added and the solution was stirred at room temperature for 6 h. Triphenylphosphine (63 mg, 0.240 mmol) and MeOH (20 mL) were added and the solution was stirred for an additional 18 h. Water was added to the reaction mixture to further precipitate the product and the mixture was allowed to sit overnight at 5 °C. The mixture was filtered, washed with cold 1 : 1 MeOH/H_2O, and dried in vacuo. The resulting solid was purified by basic alumina column chromatography with hexanes/DCM (3 : 2) and concentrated to give **7** as a yellow solid (36 mg, 27%). ^{1}H NMR (300 MHz, $CDCl_3$) 1.20 (t, 3H, J = 6.9 Hz),

1.28 (t, 3H, $J = 6.9$ Hz), 3.55–3.62 (m, 1H), 3.83–3.94 (m, 3H), 7.21–7.40 (m, 12H), 7.49–7.65 (m, 6H), 7.66 (d, 2H, $J = 7.1$ Hz); ^{13}C NMR (75 MHz, $CDCl_3$) δ 13.2, 13.6, 45.3, 46.6, 127.7, 128.5, 125.6, 128.7, 128.8, 129.5, 130.3, 130.4, 131.2, 131.6, 132.2, 132.3, 132.4, 134.2, 134.4, 137.4, 171.6, 177.2; IR: 2017, 1914, 1890 cm^{-1}; MS (*m*/*z*): $[M + Na]^+$ 790.9. HRMS (ESI+) Calcd for $C_{33}H_{30}N_2O_4PReS$ requires *m*/*z* 767.1272 [M + H], found *m*/*z* 767.1266 (0.8 ppm).

Synthesis of fac-[^{99m}Tc^{I}(CO)$_3$]$^+$ complexes

A 10^{-3}-M solution of **H1** in MeOH (100 µL) was added to 10-mM $NaHCO_3$ (400 µL) and MeOH (300 µL) in a sealable vial. The vial was sealed and sparged with N_2 for 5 min. A solution of *fac*-[^{99m}Tc^{I}$(OH_2)_3(CO)_3]^+$ (100 µL) prepared from an Isolink® kit was injected and the solution was heated to 70 °C for 30 min. The solution was cooled on ice and a solution of the monodentate ligand in MeOH (100 µL, 10^{-1} M for pyridine and methyl imidazole and 10^{-3} M for cyclohexyl isocyanide and triphenylphosphine) in MeOH was added. The vial was stirred for 30 min at room temperature. The sample was analyzed and purified by radio-HPLC.

Stability of fac-[^{99m}Tc^{I}(CO)$_3$]$^+$ complexes 4a–7a

Purified solutions of **4a**, **5a**, **6a,** and **7a** were sealed in vials under N_2. The solutions were diluted 1 : 1 with 2-mM solutions of cysteine or histidine in 10-mM pH 7.4 phosphate buffer to give a final amino acid concentration of 1 mM. The resulting solutions were incubated at 37 °C and samples were analyzed at 1 and 4 h by radio-HPLC. Decomposition of complexes was determined by analysis of the peak areas in the radiochromatogram.

X-ray structure determination and refinement for 2, 3 and 4

Intensity data were obtained at −100 °C (**2**, **3**) or 23 °C (**4**) on a Bruker SMART CCD area detector system using the ω-scan technique with Mo Kα radiation from a graphite monochromator. Intensities were corrected for Lorentz and polarization effects. Equivalent reflections were merged and absorption corrections were made using the multi-scan method [53]. Space group, lattice parameters and other relevant information are given in Supplementary material tables S1, S7, and S13 (see online supplemental material at http://dx.doi.org/10.1080/00958972.2015.1071801). The structures were solved by direct methods with full-matrix least-squares refinement using the SHELX package [54, 55] with the aid of X-SEED [56]. All non-hydrogen atoms were refined with anisotropic thermal parameters. The hydrogens were placed at calculated positions and included in the refinement using a riding model, with fixed isotropic *U*. The diethylamino group in **4** showed minor translational disorder which was included in the model with refinement of the occupancy. The final difference maps for both structures contained no features of chemical significance.

Supplementary material

Complete X-ray structural information for **2**, **3,** and **4** (CCDC 997273-997275) are available as a CIF file and additional characterization data of selected complexes as a PDF.

Acknowledgement

Isolink® kits were graciously provided by Dr Mary Dyszlewski at Covidien.

Disclosure statement

No potential conflict of interest was reported by the authors.

Funding

This work was funded in part by the DOE, Radiochemistry and Radiochemistry Instrumentation Program [#DE-FG02-08-ER64672].

References

[1] R. Alberto. *Eur. J. Nucl Med. Mol. Imaging*, **30**, 1299 (2003).
[2] R. Alberto, R. Schibli, A.P. Schubiger, U. Abram, H.J. Pietzsch, B. Johannsen. *J. Am. Chem. Soc.*, **121**, 6076 (1999).
[3] R. Alberto, R. Schibli, R. Waibel, U. Abram, A.P. Schubiger. *Coord. Chem. Rev.*, **190–192**, 901 (1999).
[4] S.S. Jurisson, J.D. Lydon. *Chem. Rev.*, **99**, 2205 (1999).
[5] G. Liu, D.J. Hnatowich. *Anti-Cancer Agents Med. Chem.*, **7**, 367 (2007).
[6] D. Papagiannopoulou. *Curr. Inorg. Chem.*, **2**, 228 (2012).
[7] R. Alberto, J.K. Pak, D. van Staveren, S. Mundwiler, P. Benny. *Biopolymers*, **76**, 324 (2004).
[8] T.R. Hayes, P.A. Lyon, C.L. Barnes, S. Trabue, P.D. Benny. *Inorg. Chem.*, **54**, 1528 (2015).
[9] A. Yazdani, N. Janzen, L. Banevicius, S. Czorny, J.F. Valliant. *Inorg. Chem.*, **54**, 1728 (2015).
[10] T.S. Pitchumony, L. Banevicius, N. Janzen, J. Zubieta, J.F. Valliant. *Inorg. Chem.*, **52**, 13521 (2013).
[11] A.L. Moore, B. Twamley, C.L. Barnes, P.D. Benny. *Inorg. Chem.*, **50**, 4686 (2011).
[12] M. Sagnou, C. Tsoukalas, C. Triantis, C.P. Raptopoulou, A. Terzis, I. Pirmettis, M. Pelecanou, M. Papadopoulos. *Inorg. Chim. Acta*, **363**, 1649 (2010).
[13] P.D. Benny, G.A. Fugate, T. Ganguly, B. Twamley, D.-K. Bučar, L.R. MacGillivray. *Inorg. Chim. Acta*, **365**, 356 (2011).
[14] C. Triantis, T. Tsotakos, C. Tsoukalas, M. Sagnou, C. Raptopoulou, A. Terzis, V. Psycharis, M. Pelecanou, I. Pirmettis, M. Papadopoulos. *Inorg. Chem.*, **52**, 12995 (2013).
[15] P.D. Benny, G.A. Fugate, A.O. Barden, J.E. Morley, E. Silva-Lopez, B. Twamley. *Inorg. Chem.*, **47**, 2240 (2008).
[16] M. Sagnou, D. Benaki, C. Triantis, T. Tsotakos, V. Psycharis, C.P. Raptopoulou, I. Pirmettis, M. Papadopoulos, M. Pelecanou. *Inorg. Chem.*, **50**, 1295 (2011).
[17] P.I. da S. Maia, H.H. Nguyen, A. Hagenbach, S. Bergemann, R. Gust, V.M. Deflon, U. Abram. *Dalton Trans.*, **42**, 5111 (2013).
[18] J.R. Dilworth, J.S. Lewis, J.R. Miller, Y. Zheng. *Polyhedron*, **12**, 221 (1993).
[19] H.H. Nguyen, U. Abram. *Inorg. Chem.*, **46**, 5310 (2007).
[20] H.H. Nguyen, U. Abram. *Z. Anorg. Allg. Chem.*, **634**, 1560 (2008).
[21] H.H. Nguyen, V.M. Deflon, U. Abram. *Eur. J. Inorg. Chem.*, **2009**, 3179 (2009).
[22] H.H. Nguyen, U. Abram. *Polyhedron*, **28**, 3945 (2009).
[23] H.H. Nguyen, P.C. Thang, A. Rodenstein, R. Kirmse, U. Abram. *Inorg. Chem.*, **50**, 590 (2010).
[24] B. Schmitt, T.I.A. Gerber, E. Hosten, R. Betz. *Inorg. Chem. Commun.*, **24**, 136 (2012).
[25] U. Abram, S. Abram, R. Alberto, R. Schibli. *Inorg. Chim. Acta*, **248**, 193 (1996).
[26] I.B. Douglass, F.B. Dains. *J. Am. Chem. Soc.*, **56**, 719 (1934).
[27] J. Imrich, T. Bušová, P. Kristian, J. Džara. *Chem. Pap.*, **48**, 42 (1994).
[28] F. Dietze, S. Schmidt, E. Hoyer. *Z. Anorg. Allg. Chem.*, **595**, 35 (1991).
[29] W. Hernández, E. Spodine, R. Richter, K.-H. Hallmeier, U. Schröder, L. Beyer. *Z. Anorg. Allg. Chem.*, **629**, 2559 (2003).
[30] H.H. Nguyen, U. Abram. *Inorg. Chem. Commun.*, **11**, 1478 (2008).
[31] K.R. Koch. *Coord. Chem. Rev.*, **216–217**, 473 (2001).
[32] D.A. Edwards, J. Marshalsea. *J. Organomet. Chem.*, **131**, 73 (1977).
[33] A. Irving, K.R. Koch, M. Matoetoe. *Inorg. Chim. Acta*, **206**, 193 (1993).
[34] G. Kemp, A. Roodt, W. Purcell, K.R. Koch. *Rhodium Express*, 16 (1996).

[35] K.R. Koch, T. Grimmbacher, C. Sacht. *Polyhedron*, **17**, 267 (1998).
[36] V.L. Beyer, E. Hoyer, J. Libscher, H. Hartmann. *Z. Chem.*, **21**, 81 (1981).
[37] P. Muhl, K. Gloe, F. Dietze, E. Hoyer, L. Beyer. *Z. Chem.*, **26**, 81 (1986).
[38] L. Beyer, E. Hoyer, H. Hennig, R. Kirmse, H. Hartmann, J. Liebscher. *J. Prakt. Chem.*, **317**, 829 (1975).
[39] S. Schmidt, F. Dietze, E. Hoyer. *Z. Anorg. Allg. Chem.*, **603**, 33 (1991).
[40] H. Braband, U. Abram. *J. Organomet. Chem.*, **689**, 2066 (2004).
[41] S. Behrendt, L. Beyer, F. Dietze, E. Kleinpeter, E. Hoyer, E. Ludwig, E. Uhlemann. *Inorg. Chim. Acta*, **43**, 141 (1980).
[42] E. Kleinpeter, S. Behrendt, L. Beyer, W. Dietzsch, R. Borsdorf. *J. Prakt. Chem.*, **324**, 29 (1982).
[43] P.D. Benny, T. Ganguly, L. Raiford, G.A. Fugate, B. Twamley. *Inorg. Chem. Commun.*, **14**, 392 (2011).
[44] H. He, J.E. Morley, B. Twamley, R.H. Groeneman, D.-K. Bučar, L.R. MacGillivray, P.D. Benny. *Inorg. Chem.*, **48**, 10625 (2009).
[45] L.R. Gomes, L.M.N.B.F. Santos, J.A.P. Coutinho, B. Schröder, J.N. Low. *Acta Crystallogr., Sect. E: Struct. Rep. Online*, **66**, o870 (2010).
[46] K.W. Lee, T.L. Brown. *Inorg. Chem.*, **26**, 1852 (1987).
[47] R. Schaefer, W. Kaim, J. Fiedler. *Inorg. Chem.*, **32**, 3199 (1993).
[48] V. Küllmer, H. Vahrenkamp. *Chem. Ber.*, **110**, 3799 (1977).
[49] F.H. Allen, O. Kennard, D.G. Watson, L. Brammer, A.G. Orpen, R. Taylor. *J. Chem. Soc., Perkin Trans. 2*, **12**, S1 (1987).
[50] T.R. Hayes, B.B. Kasten, C.L. Barnes, P.D. Benny. *Dalton Trans.*, **43**, 6998 (2014).
[51] S.P. Schmidt, J. Nitschke, W.C. Trogler, S.I. Huckett, R.J. Angelici. *Inorg. Synth.*, **26**, 113 (1989).
[52] H. He, M. Lipowska, X. Xu, A.T. Taylor, M. Carlone, L.G. Marzilli. *Inorg. Chem.*, **44**, 5437 (2005).
[53] G.M. Sheldrick, *SADABS (Version 2.10)*, University of Göttingen, Göttingen, Germany (2003).
[54] G.M. Sheldrick. *SHELXS-97, Crystal Structure Solution*, University of Göttingen, Göttingen, Germany (1997).
[55] G.G. Sheldrick. *SHELXL-97, Crystal Structure Refinement*, University of Göttingen, Göttingen, Germany (1997).
[56] X-SEED: L.J. Barbour. *J. Supramol. Chem.*, **1**, 189 (2001).