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Synthesis and Characterization of Rhenium(V) Oxo Complexes with *N*-[*N*-(3-Diphenylphosphinopropionyl)glycyl]cysteine Methyl Ester. X-ray Crystal Structure of {ReO[$Ph_2P(CH_2)_2C(O)$-Gly-Cys-OMe(*P*,*N*,*N*,*S*)]}

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The PN_2S chelate *N*-[*N*-(3-diphenylphosphinopropionyl)glycyl]-*S*-tritylcysteine methyl ester [PN_2S(Trt)-OMe] was synthesized and reacted with $ReOCl_3(PPh_3)_2$ and $Ph_4P[ReOCl_4]$. The reactions of both tritylated and detritylated ligands with Re^VO precursors gave two diastereomers, **9a** and **9b**, of the ReO(PN_2S-OMe) complex. The two isomers, produced in a 1:1 molar ratio, are stable and do not interconvert. They were separated by reverse-phase HPLC and characterized by NMR, FT-IR, and UV–visible spectroscopy and electrospray mass spectrometry. X-ray analysis established for **9a** the presence in the solid of the syn isomer. Compound **9a**, $C_{21}H_{23}N_2O_5PSRe$, crystallized from warm acetonitrile in the triclinic space group $P\bar{1}$, $a = 9.828(2)$ Å, $b = 11.163(2)$ Å, $c = 11.641(2)$ Å, $\alpha = 106.48(3)°$, $\beta = 109.06(3)°$, $\gamma = 102.81(3)°$, $V = 1085.7(4)$ Å^3, $Z = 2$. The PN_2S coordination set is in the equatorial plane, and the complex shows a distorted square pyramidal coordination. The anti configuration assigned to **9b** is consistent with all the available physicochemical data. Follow-up of the reaction of the detritylated ligand with $Ph_4P[ReOCl_4]$ in ethanol or acetonitrile indicated that the phosphorus atom of the chelate binds first to the metal and that this bond acts as the driving force for coordination.

Introduction

The development of site-specific radioimaging/radiotherapeutic agents involves the labeling of biologically active molecules with ^{99m}technetium and $^{186/188}$rhenium. ^{99m}Tc is widely used for diagnostic imaging due to its ideal nuclear properties,[1] while $^{186/188}$Re are two of the most promising isotopes for radiotherapy.[2] In addition, because of the analogy in the chemical properties of the two metals, rhenium complexes are excellent structural models for Tc complexes with identical ligands. Labeling of a bioactive molecule with ^{99m}Tc and $^{186/188}$Re can be achieved by the direct method, or alternatively by means of a bifunctional chelating agent that coordinates the radioisotope to form an in vivo stable complex and that contains a functional group for the conjugation to the target molecule. In the past few years, tetradentate bifunctional chelators containing one or more phosphine moieties have been investigated. Due to the σ-donor and π-acceptor properties of phosphine phosphorus,[3] the ligands were found able to produce highly stable rhenium(V) oxo and ^{99m}technetium(V) oxo complexes.[4] In a previous work,[5] we investigated the PN_2S ligand *N*-(*N*-(3-diphenylphosphinopropionyl)glycyl)-*S*-benzylcysteine methyl ester and we found that it shows high affinity toward the Tc^VO

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and Re^{V}O moieties. The ReOCl[PN$_2$S(Bzl)-OMe] complex shows the ReO^{3+} unit in an octahedral configuration with the thioether in the equatorial plane together with the deprotonated amides and phosphorus; a chlorine atom trans to an oxo group completes the hexacoordination, and the methyl ester group leans out of the coordination sphere.

In this paper, we report the synthesis and characterization of the analogous *N*-(*N*-(3-diphenylphosphinopropionyl)-glycyl)-*S*-tritylcysteine methyl ester ligand and of the related Re^{V}O complexes. The ligand, where the benzyl group was replaced by the triphenylmethyl group easily removable under acidic conditions, was chosen in order to have the cysteine sulfur available for coordination as thiolate instead of thioether.

Since upon coordination the methyl ester can assume the syn or anti configuration with respect to the ReO^{3+} core, two diastereomeric complexes are expected. The two isomers were isolated, and the X-ray crystal structure of the syn Re^{V}O isomer was determined.

Experimental Section

Materials and Methods. All chemicals and solvents were reagent grade and were used without further purification. Amino acids and coupling reagents (*N*,*N*′-dicyclohexylcarbodiimide, DCC; 1-hydroxybenzotriazole, HOBT; benzotriazolyloxy-tris[pyrrolidino]-phosphonium hexafluorophosphate, PyBOP) were purchased from Novabiochem (Laufelfingen, Switzerland) and Fluka (Sigma-Aldrich, Milano, Italy); 3-diphenylphosphinopropionic acid succinimidic ester was provided from Argus Spechem S.a.s. (Prato, Italy). Rhenium was purchased from Sigma-Aldrich (Milano, Italy) as $KReO_4$, whereas solvents employed in the syntheses were obtained from Carlo Erba Reagenti (Div. Antibioticos, Milano, Italy). The syntheses of the ligands and of the Re complexes were carried out under an argon atmosphere using solvents degassed and tested for peroxides before use. The precursors $Ph_4P[ReOCl_4]$ and $ReOCl_3(PPh_3)_2$ were synthesized according to the literature.[6] Reagents and solvents for HPLC eluents were purchased from Fluka (Sigma-Aldrich, Milano, Italy) and Carlo Erba Reagenti (Div. Antibioticos, Milano, Italy); MilliQ water was obtained from a Millipore system (Millipore, Vimodrone, Milano, Italy). Solvents for ^{1}H NMR, ^{13}C NMR, and ^{31}P NMR analysis ($CDCl_3$ and DMSO-d_6) were purchased from Sigma-Aldrich (Milano, Italy). Analytical thin-layer chromatography (TLC) was performed on Merck (Darmstadt, Germany) silica gel 60-F_{254} plates using the following elution systems: (a) $CHCl_3$:CH_3OH:AcOH (9:0.8:0.2), (b) $CHCl_3$:CH_3OH (9:1). Visualization was accomplished by UV detection at 254 nm and/or staining with ninhydrin, Ellman's,[7] or iodine reagents. Flash chromatography was carried out using silica gel 230−400 mesh (Merck, Darmstadt, Germany) according to the method of Still.[8] Reverse-phase high-performance liquid chromatography (RP-HPLC) was performed at 35°C using a Waters chromatography system (Waters, Milford, MA) comprising a binary Waters 510 programmable gradient pump controlled by a gradient controller 680 automated. For analytical injections a Hamilton PRP-1 column (4.1 × 250 mm, Alltech Italia, Sedriano, Milano) was used with a 20 μL injection loop. For semipreparative injections a Hamilton PRP-1 column (7.1 × 350 mm, Alltech Italia, Sedriano, Milano) was used with a 100 μL injection loop. Both columns were equipped with a Hamilton PRP-1 precolumn (Alltech Italia, Sedriano, Milano). The analyses were monitored with a Waters 486 tunable absorbance detector set at 215 nm. Two different elution methods were used. Method 1 consisted of a linear gradient (0−6 min, 0−100% solvent B; 6−14 min, 100% B; 14−15 min, 100−0% B; 15−21 min, 0% B) using a mobile phase of 0.05% TFA in water (solvent A) and 0.05% TFA in 95% acetonitrile (solvent B), at a flow rate of 1 mL min^{-1}. Method 2 consisted of a linear gradient (0−20 min, 40−60% B; 20−25 min, 60−100% B; 25−30 min, 100% B; 30−32 min, 100−40% B) using a mobile phase of 0.05% TFA in water (solvent A) and 0.05% TFA in acetonitrile (solvent B) at a flow rate of 1 mL min^{-1}. Elemental analyses were performed on a Perkin-Elmer 2400/II automated analyzer. ^{1}H NMR and ^{13}C NMR were obtained on Bruker 300 MHz spectrometer, and chemical shifts (ppm) were referenced to tetramethylsilane (TMS). ^{31}P NMR were recorded on a Bruker AC-200 spectrometer using 85% aqueous H_3PO_4 as external reference. Multiplicities were reported as s (singlet), d (doublet), dd (doublet of doublet), t (triplet), q (quadruplet), m (multiplet), and b (broad signal). Infrared spectra were recorded in the range 4000−500 cm^{-1} on a NICOLET FT-IR model 5SXC spectrophotometer as KBr pellets. UV−visible spectra were performed on a Perkin-Elmer Lambda 25 UV/vis spectrometer in $CHCl_3$ and DMF at room temperature. Electrospray ionization mass spectra (ES-MS) were obtained on an Applied Biosystems Mariner System 5220 mass spectrometer (PerSeptive Biosystems Inc, Framingham, MA) in the positive-ion mode dissolving the samples in MeCN (10^{-3} M). Optical rotations were measured in a Perkin-Elmer 141 polarimeter. The X-ray crystal structure determination was performed by means of a graphite-monochromated Nicolet R3m/V four-circle diffractometer.

Synthesis of the Ligand. *N*-Tritylglycyl-*S*-trityl-L-cysteine (1), [Trt-Gly-L-Cys(Trt)-OH]. To a chilled solution of *N*-tritylglycine (1.00 g, 3.15 mmol) and HOBt (468 mg, 3.46 mmol) in anhydrous CH_2Cl_2 (100 mL) was added DCC (651 mg, 3.15 mmol). The mixture was kept at 0 °C for 15 min, left to attain room temperature, and stirred for 3 h. The precipitate *N*,*N*′-dicyclohexylurea was filtered off, and to the filtrate were added *S*-trityl-L-cysteine (1.374 g, 3.78 mmol) and TEA (526 μL, 3.78 mmol). After 5 h the solvent was removed under vacuum and the remaining residue was taken up with EtOAc (100 mL), washed with 2% $KHSO_4$ solution (2 × 50 mL), 5% Na_2CO_3 solution (2 × 50 mL), and brine (1 × 50 mL), and then dried over Na_2SO_4. The solvent was evaporated under vacuum, and the oily residue was crystallized as a white powder upon addition of Et_2O/petroleum ether. Yield: 91% (1.90 g, 2.87 mmol). TLC (a): R_f = 0.68. Anal. Calcd for $C_{43}H_{38}N_2SO_3$: C, 77.91; H, 5.78; N, 4.22; S, 4.83. Found: C, 77.51; H, 5.42; N, 4.13; S, 4.66. ^{1}H NMR ($CDCl_3$): 2.69 (dd, 1H, ^2J = 12.5, ^3J = 4.8, Cys-βCH), 2.77 (dd, 1H, ^2J = 12.5, ^3J = 6.5, Cys-βCH), 2.89 (d, 2H, J_{CH-NH} = 3.7, Gly-CH_2), 4.32 (m, 1H, ^3J = 4.8, ^3J = 6.5, J_{CH-NH} = 7.1, Cys-αCH), 7.08−7.40 (m, 31H, ArH, Gly-NH), 7.91 (d, 1H, J_{NH-CH} = 7.1, Cys-NH). ^{13}C NMR ($CDCl_3$): 35.1 (Cys-C_β), 49.0 (Gly-C_α), 61.5 (Cys-C_α), 67.4 (*C*Ph$_3$), 72.0 (*C*Ph$_3$), 127.7, 127.8, 129.0, 129.2 129.8, 130.6, 145.6, 146.2 (C_{Arom}), 172.6 (CONH).

Glycyl-*S*-trityl-L-cysteine Acetate (2), [H-Gly-L-Cys(Trt)-OH AcOH]. To a solution of compound **1** (1.00 g, 1.51 mmol) in CH_2Cl_2 (2 mL) were added AcOH (5 mL) and water (1 mL). and the mixture was heated for 30 min on a steam bath. On cooling, triphenylcarbinol separated out. Upon addition of Et_2O, the carbinol was dissolved and **2** precipitated as a white powder. Yield: 99%

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(715 mg, 1.48 mmol). TLC (a): R_f = 0.11. Anal. Calcd for $C_{26}H_{28}N_2SO_5$: C, 64.98; H, 5.87; N, 5.83; S, 6.67. Found: C, 64.82; H, 5.63; N, 5.46; S, 6.60. ^{1}H NMR (DMSO-d_6): 1.80 (s, 3H, $COCH_3$), 2.26 (dd, 1H, ^2J = 11.0, ^3J = 6.2, Cys-βCH), 2.30 (dd, 1H, ^2J = 11.0, ^3J = 4.8, Cys-βCH), 3.02 (d, 1H, ^2J = 16.0, Gly-CH), 3.08 (d, 1H,^2J = 16.0, Gly-CH), 3.84 (dd, 1H, ^3J = 6.2, ^3J = 4.8, Cys-αCH), 7.09−7.24 (m, 15H, ArH), 7.91 (b, 1H, Cys-NH).

***N*-[*N*-(3-Diphenylphosphinopropionyl)glycyl]-*S*-trityl-L-cysteine (3), [PN$_2$S(Trt)-OH].** To a suspension of compound **2** (700 mg, 1.45 mmol) in CH_2Cl_2 (100 mL) was added TEA (403 μL, 2.90 mmol). To the resulting clear solution was added 3-diphenylphosphinepropionylsuccinimide ester (468 mg, 1.32 mmol), and the pH was adjusted to 9 with TEA. The mixture was stirred under an argon atmosphere for 5 h and the solvent removed under vacuum. The oily residue was taken up with EtOAc (100 mL), washed with 2% $KHSO_4$ solution (2 × 50 mL), 5% Na_2CO_3 solution (2 × 50 mL), and brine (1 × 50 mL), and then dried over Na_2SO_4. The solvent was evaporated under vacuum, and the oily residue was crystallized as a white powder upon addition of Et_2O/petroleum ether. Yield: 92% (882 mg, 1.33 mmol). TLC (a): R_f = 0.65. HPLC (method 1): R_t = 12.0 min. Anal. Calcd for $C_{39}H_{37}N_2PSO_4$: C, 70.89; H, 5.64; N, 4.24. Found: C, 69.98; H, 5.67; N, 4.22. FT-IR (KBr, cm^{-1}): 1734 (C═O carboxylic group); 1654, 1642 (C═O amides); 742, 699 (phenyls). ES-MS: *m*/*z* 661 [M$^+$]. $[\alpha]^{25}_D = -7°$ (*c* 1, CH_3OH). ^{31}P NMR ($CDCl_3$): −14.87. ^{1}H NMR ($CDCl_3$): 2.25−2.36 (m, 4H, $P(CH_2)_2$), 2.67 (dd, 1H, ^2J = 12.7, ^3J = 4.7, Cys-βCH), 2.74 (dd, 1H, ^2J = 12.7, ^3J = 6.3, Cys-βCH), 3.78 (dd, 1H, ^2J = 17.0, J_{CH-NH} = 4.7, Gly-CH), 3.91 (dd, 1H, ^2J = 17.0, J_{CH-NH} = 5.0, Gly-CH), 4.37 (m, 1H, ^3J = 4.7, ^3J = 6.3, J_{CH-NH} = 7.3, Cys-αCH), 6.45 (m, 1H, J_{NH-CH} = 4.7, J_{NH-CH} = 5.0, Gly-NH), 6.85 (d, 1H, J_{NH-CH} = 7.3, Cys-NH), 7.17−7.39 (m, 25H, ArH). ^{13}C NMR ($CDCl_3$): 23.3 (P*CH*$_2$), 32.2 (PCH$_2$*CH*$_2$), 32.4 (Cys-C$_\beta$), 42.7 (Gly-C$_\alpha$), 52.1 (Cys-C$_\alpha$), 66.3 (*C*Ph$_3$), 126.6, 127.8, 128.4, 128.5, 128.6, 129.5, 132.6, 132.7, 137.8, 144.6 (C$_{Arom}$), 172.5 (CONH), 174.6 (CONH).

***N*-[*N*-(3-Diphenylphosphinopropionyl)glycyl]-*S*-tritylcysteine Methyl Ester (4), [PN$_2$S(Trt)-OMe].** To a solution of **3** (660 mg, 1.00 mmol) and PyBOP (728 mg, 1.40 mmol) in MeOH (60 mL) was added diisopropylethylamine (349 μL, 2.00 mmol). The mixture was cooled to 0 °C and stirred for 6 h. The solvent was removed under vacuum, and the oily residue was taken up with EtOAc (100 mL), washed with 2% $KHSO_4$ solution (2 × 50 mL), 5% Na_2CO_3 solution (2 × 50 mL), and brine (1 × 50 mL), and then dried over Na_2SO_4. The product was separated by silica gel flash column chromatography (EtOAc) and crystallized as a yellow powder upon addition of Et_2O/petroleum ether. Yield: 90% (590 mg, 0.87 mmol). TLC (a): R_f = 0.77. HPLC (method 1): R_t = 10.5 min. Anal. Calcd for $C_{40}H_{39}N_2PSO_4$: C, 71.20; H, 5.83; N, 4.15. Found: C, 71.45; H, 5.98; N, 4.03. FT-IR (KBr, cm^{-1}): 1748 (C═O methyl ester); 1650, 1642 (C═O amides); 742, 698 (phenyls). ES-MS: *m*/*z* 674 [M$^+$]. $[\alpha]^{25}_D = 0°$ (*c* 1, CH_3OH). ^{31}P NMR ($CDCl_3$): −15.46 ppm. ^{1}H NMR and ^{13}C NMR spectral data are reported in Tables 1 and 2, respectively.

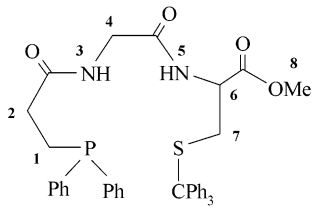
Deprotection of PN$_2$S(Trt)-OMe (5), (PN$_2$S-OMe). Compound **4** (65 mg, 0.15 mmol) was dissolved in TFA (5 mL). When triethylsilane was added (23 μL, 0.3 mmol), the solution became colorless, and it was stirred for 45 min. TFA was then removed under vacuum, and the resulting residue was used immediately in the reactions with rhenium(V) oxo starting materials without further purifications. TLC (a): R_f = 0.60. HPLC (1): R_t = 9.7 min.

Synthesis of the Re Complexes. Ph$_4$P[ReOCl$_3$(PN$_2$S-OMe)] (7). The residue obtained by deprotection of **4** (100 mg, 0.15 mmol) was dissolved in absolute ethanol (or MeCN, 10 mL) and then added to a solution of $Ph_4P[ReOCl_4]$ (101 mg, 0.15 mmol) in the same solvent (10 mL) at room temperature. After a few minutes a green precipitate (**6**) separated out. The suspension was stirred under argon for 4 h, and compound **6** was converted in a second matt green precipitate (**7**). Compound **7** was recovered by filtration and washed with absolute ethanol. Yield: 73% (118 mg, 0.11 mmol).

Anal. Calcd for $C_{45}H_{44}N_2P_2SCl_3O_5Re$: C, 50.07; H, 4.11; N, 2.60; P, 5.74; S, 2.97; Cl, 9.85. Found: C, 49.27; H, 3.95; N, 2.24; P, 5.31; S, 2.58; Cl, 9.33. FT-IR (KBr, cm^{-1}): 1737 (C═O methyl ester); 1669, 1654 (C═O amides); 1558, 1543, 1535, 1522 (N−H, C−N); 906 (Re═O); 1440, 1108, 993, 751, 723, 689 (PPh_4^+ and phenyls).

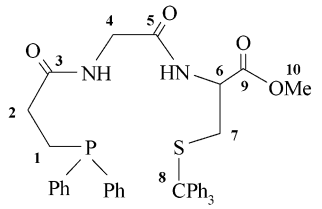
[ReO(PN$_2$S-OMe)] (9a, 9b). Method 1. To a suspension of **7** (100 mg, 0.09 mmol) in absolute ethanol was added TEA till pH 8.5, and after 10 min a reddish-brown solution was obtained. The solution was treated with $NaBPh_4$ (30 mg, 0.09 mmol), and a precipitate was formed and removed by filtration. The filtrate was then evaporated under vacuum, and the residue was taken up with CH_2Cl_2 and washed with 2% $KHSO_4$ solution (2 × 20 mL) and brine (1 × 20 mL). The organic layer was dried over Na_2SO_4 and concentrated to 2 mL. Upon addition of Et_2O/petroleum ether an

Table 1. ^{1}H NMR Data for Compound **4**



	δ (ppm)	J_{H-H} (Hz)
H(1), (2)	2.41−2.25 (m, 4H)	
H(3)	6.10 (t, 1H)	J_{H3-H4} = 4.8
H(4)	3.84 (d, 2H)	J_{H4-H3} = 4.8
H(5)	6.27 (d, 1H)	J_{H5-H6} = 7.7
H(6)	4.48 (m, 1H)	J_{H6-H7a} = 4.6, J_{H6-H7b} = 6.2, J_{H6-H5} = 7.7
H(7a)	2.70 (dd, 1H)	$J_{H7a-H7b}$ = 12.6, J_{H7a-H6} = 4.6
H(7b)	2.63 (dd, 1H)	$J_{H7b-H7a}$ = 12.6, $^3J_{H7b-H6}$ = 6.2
H(8)	3.76 (s, 3H)	
H(Aromatic)	7.43−7.18 (m, 25H)	

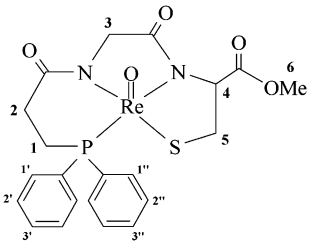
Table 2. ^{13}C NMR Data for Compound **4**



	δ (ppm)	J_{C-P} (Hz)
C(1)	23.7	46.8
C(2)	32.8	78.0
C(3)	172.8	54.0
C(4)	43.2	
C(5)	170.8	
C(6)	67.5	
C(7)	33.8	
C(8)	67.5	
C(9)	168.7	
C(10)	51.7	
C(Ph-Trt)	*a*	
C(Ph$_2$P)	*b*	

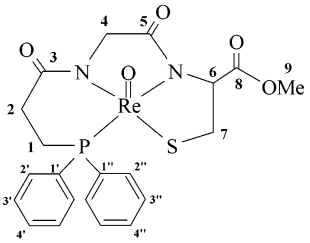
[a] (J_{C-P}, Hz): 129.0 (26.7), 129.2, 133.1 (74.7), 138.0 (47.1). [b] 127.3, 128.4, 129.8, 144.6.

Table 3. ^{1}H NMR Data for Compounds **9a** and **9b**



	compound **9a**		compound **9b**	
	δ (ppm)	J_{H-H} (Hz)	δ (ppm)	J_{H-H} (Hz)
H(1)	2.60−2.51 (m, 1H)		2.67−2.55 (m, 1H)	
H(1), H(2)	3.18−3.02 (m, 3H)		3.25−3.08 (m, 3H)	
H(3a)	4.64 (d, 1H)	$J_{H3a-H3b} = 19.5$	4.75 (d, 1H)	$J_{H3a-H3b} = 19.8$
H(3b)	4.88 (d, 1H)	$J_{H3b-H3a} = 19.5$	4.90 (d, 1H)	$J_{H3b-H3a} = 19.8$
H(4)	5.74 (dd, 1H)	$J_{H4-H5a} = 7.9$, $J_{H4-H5b} = 2.1$	5.43 (d, 1H)	$J_{H4-H5a} = 7.5$
H(5a) exo	3.24 (dd, 1H)	$J_{H5a-H5b} = 12.4$, $J_{H5a-H4} = 7.9$	3.68 (dd, 1H)	$J_{H5a-H5b} = 12.6$, $J_{H5a-H4} = 7.5$
H(5b) endo	3.83 (dd, 1H)	$J_{H5b-H5a} = 12.4$, $J_{H5b-H4} = 2.1$	4.13 (d, 1H)	$J_{H5a-H5b} = 12.6$
H(6)	3.69 (s, 3H)		3.61 (s, 3H)	
H(1′)	7.28−7.22 (m, 2H)		7.13−7.09 (m, 2H)	
H(1″)	7.91−7.84 (m, 2H)		8.04−7.98 (m, 2H)	
H(2′, 2″, 3′, 3″)	7.68−7.44 (m, 6H)		7.67−7.26 (m, 6H)	

Table 4. ^{13}C NMR Data for Compounds **9a** and **9b**



	compound **9a**		compound **9b**	
	δ (ppm)	J_{C-P} (Hz)	δ (ppm)	J_{C-P} (Hz)
C(1)	23.3	139.1	23.6	137.1
C(2)	33.1		33.8	
C(3)	185.6		188.3	
C(4)	61.4		61.2	
C(5)	180.9		180.1	
C(6)	65.7		69.5	
C(7)	45.8	22.2	48.6	23.4
C(8)	171.9		172.4	
C(9)	52.9		52.8	
C(1′, 1″)	125.1	227.0	129.2	213.0
C(2′) or (2″)	132.7	32.7	133.0	40.2
C(2′) or (2″)	136.6	45.9	134.4	44.1
C(3′) (3″)	130.2	45.0	130.2	45.0
C(4′) or (4″)	132.5	9.0	132.5	9.0
C(4′) or (4″)	133.7	7.8	133.7	7.8

equimolar mixture of two compounds was recovered. Yield: 89% (50 mg, 0.08 mmol). The two compounds were separated by RP-HPLC (method 2).

Fraction 1 (**9a**) was obtained as a brownish-green powder and crystallized from warm acetonitrile producing purple-red crystals suitable for X-ray crystallographic work. TLC (b): $R_f = 0.74$. HPLC (2): $R_t = 9.15$ min. Anal. Calcd for $C_{21}H_{22}N_2PSO_5Re$: C, 39.93; H, 3.51; N, 4.43. Found: C, 40.12; H, 3.64; N, 4.80. FT-IR (KBr, cm^{-1}): 1743 (C═O methyl ester); 1656 (C═O amides) 989 (Re═O); 751, 724, 695 (phenyls). ES-MS: *m*/*z* 631 [M$^+$]. UV−vis (nm): ($CHCl_3$) 509; (DMF) 450. ^{31}P NMR ($CDCl_3$): 28.36. ^{1}H NMR and ^{13}C NMR spectral data are reported in Tables 3 and 4, respectively.

Fraction 2 (**9b**) was obtained as a brownish-green powder and crystallized from MeOH/Et_2O by slow evaporation affording green thin sheet crystals unsuitable for X-ray crystallographic analysis. TLC (b): $R_f = 0.69$. HPLC (2): $R_t = 11.45$ min. Anal. Calcd for $C_{21}H_{22}N_2PSO_5Re$: C, 39.93; H, 3.51; N, 4.43. Found: C, 39.50; H, 3.78; N, 4.60. FT-IR (KBr, cm^{-1}): 1744 (C═O methyl ester); 1654 (C═O amides); 987 (Re═O); 750, 735, 695 (phenyls). ES-MS: *m*/*z* 631 [M$^+$]. UV−vis (nm): ($CHCl_3$) 502; (DMF) 447. ^{31}P NMR ($CDCl_3$): 28.69. ^{1}H NMR and ^{13}C NMR spectral data are reported in Tables 3 and 4, respectively.

Method 2. The residue obtained by deprotection of **4** (100 mg, 0.15 mmol) was dissolved in absolute ethanol (or MeCN, 10 mL) and then added to a solution of $Ph_4P[ReOCl_4]$ (101 mg, 0.15 mmol) in the same solvent (10 mL) at room temperature. After a few minutes a green precipitate (**6**) separated out. Compound **6** was recovered by filtration, washed with absolute ethanol, and dissolved in CH_2Cl_2, resulting in a green-orange solution. By addition of TEA till pH 8.5 the color changed to reddish-brown. The latter solution was treated with $NaBPh_4$ (51 mg, 0.15 mmol), and the precipitate was removed by filtration. The filtrate was then evaporated under vacuum and the residue taken up with CH_2Cl_2 and washed and treated as in method 1, resulting in an equimolar mixture of two compounds. Yield: 67% (63 mg, 0.10 mmol). The two compounds were separated by RP-HPLC (method 2) and showed TLC, HPLC, and NMR behavior identical to that of compounds prepared by method 1.

Method 3. A solution of compound **4** (100 mg, 0.15 mmol) in $CHCl_3$ (10 mL) was added dropwise to a solution of $Ph_4P[ReOCl_4]$ (101 mg, 0.15 mmol) in the same solvent (10 mL). After 4 h at room temperature TEA was added till pH to 8.5 and in a few minutes the solution became reddish-brown. After 15 minutes of stirring, $NaBPh_4$ (51 mg, 0.15 mmol) was added and the precipitate was removed by filtration. The filtrate was then evaporated under vacuum and the residue taken up with CH_2Cl_2 and then washed and treated as in method 1, resulting in an equimolar mixture of two compounds. Yield: 65% (61 mg, 0.09 mmol). The two compounds were separated by RP-HPLC (method 2) and showed TLC, HPLC, and NMR behavior identical to that of **9a** and **9b** prepared by method 1.

Method 4. The residue obtained by deprotection of **4** (100 mg, 0.15 mmol) was dissolved in absolute ethanol (10 mL) and added dropwise to a suspension of $ReOCl_3(PPh_3)_2$ (123 mg, 0.15 mmol)

Table 5. Summary of Crystal and Refinement Data for Compound **9a**[a]

empirical formula	$C_{21}H_{23}N_2O_5PSRe$
fw	632.64
cryst syst; space group	triclinic; $P\bar{1}$ (No. 2)
a, Å	9.828 (2)
b, Å	11.163 (2)
c, Å	11.641 (2)
α, deg	106.48 (3)
β, deg	109.06 (3)
γ, deg	102.81 (3)
V, Å^3	1085.7 (4)
Z; d_{calc}, Mg m^{-3}	2; 1.935
λ , Å; μ, cm^{-1}	0.71073; 58.0
θ range, deg	3.3−27.5
reflns collected	4966
reflns obsd ($I > 2\sigma(I)$)	4142
data/params ratio	4966/281
R1[b] (obsd reflns)	0.044
wR2[c] (obsd reflns)	0.111
GOF[d] on F^2	0.986
largest peak, e Å^{-3}	1.56

[a] Other details of data collection/refinement: Nicolet/Siemens R3m/V diffractometer; $T = 294(2)$ K; highly oriented graphite monochromator; $\omega-2\theta$ scans; two standard reflections every 150; refinement by full-matrix least-squares method on F_o^2; riding model for H atoms. [b] $R1 = \sum||F_o| - |F_c||/\sum|F_o|$. [c] $wR2 = [\sum w(F_o^2 - F_c^2)^2/\sum w(F_o^2)^2]^{1/2}$. [d] $GOF = [\sum w(|F_o^2| - |F_c^2|)^2/(N_{obs} - N_{parameters})]^{1/2}$.

in the same solvent (10 mL). The mixture was heated under reflux for 1 h till a reddish-brown color appeared. After cooling at room temperature, the solvent was evaporated under vacuum. The residue was taken up with CH_2Cl_2 and then washed and treated as in method 1, resulting in an equimolar mixture of two compounds. Yield: 58% (54 mg, 0.08 mmol). The two compounds were separated by RP-HPLC (method 2) and showed TLC, HPLC, and NMR behavior identical to that of compounds prepared by method 1.

Method 5. The residue obtained by deprotection of **4** (100 mg, 0.15 mmol) was dissolved in CH_2Cl_2 (10 mL) and added dropwise to a solution of $ReOCl_3(PPh_3)_2$ (123 mg, 0.15 mmol) in the same solvent (10 mL) at room temperature. By addition of TEA the pH was increased to 8.5, and after 10 min the color of the solution changed to reddish-brown. The mixture was then washed and treated as in method 1, resulting in an equimolar mixture of two compounds. Yield: 73% (69 mg, 0.11 mmol). The two compounds were separated by RP-HPLC (method 2) and showed TLC, HPLC, and NMR behavior identical to that of **9a** and **9b** prepared by method 1.

X-ray Crystallography. A single purple-red crystal of compound **9a**, measuring ca. 0.12 × 0.10 × 0.08 mm, was mounted on a glass fiber, coated with epoxy resin, and transferred to the diffractometer. The structure was solved by heavy-atom methods, completed by subsequent difference Fourier syntheses, and refined with full-matrix least-squares procedures based on F^2. In the final least-squares cycles the non-hydrogen atoms have been refined anisotropically. The hydrogen atoms have been included in calculated positions and refined with the riding model. Of the two possible space groups, *P*1 and $P\bar{1}$, a satisfactory solution has been reached in the centrosymmetric alternative, $P\bar{1}$. The structure has been solved and refined using the SHELXTL NT[9] and SHELXL-97[10] software. The most significant details of the crystallography study are reported in Table 5. A collection of selected bond distances and angles are shown in Table 6.

Table 6. Selected Bond Distances (Å) and Angles (deg) for Compound **9a**

Re−S	2.268(2)	S−C(7)	1.823(9)
Re−P	2.446(2)	P−C(1)	1.819(9)
Re−N(1)	2.027(7)	N(1)−C(3)	1.36(1)
Re−N(2)	1.976(6)	N(2)−C(6)	1.47(1)
Re−O(1)	1.682(5)		
P−Re−N(1)	90.5(2)	Re−P−C(1)	107.7(3)
N(1)−Re−N(2)	79.3(3)	Re−S−C(7)	103.1(3)
N(2)−Re−S	81.6(2)	Re−N(1)−C(3)	132.1(6)
O(1)−Re−P	102.8(2)	Re−N(1)−C(4)	113.8(5)
O(1)−Re−N(1)	112.7(3)	Re−N(2)−C(5)	120.8(5)
O(1)−Re−N(2)	107.8(3)	Re−N(2)−C(6)	120.2(5)
O(1)−Re−S	113.5(2)		

Results and Discussion

Synthesis of the Ligands. The synthesis of the PN_2S-(Trt)-OMe ligand (**4**) was achieved following the procedure in Scheme 1. The N_2S part of the chelate was produced by coupling *N*-tritylglycine with *S*-trityl-L-cysteine to obtain the *N*-tritylglycyl-*S*-trityl-L-cysteine dipeptide (**1**). On the basis of the general method described by Theodoropoulos et al.,[11] *N*-tritylglycine was activated as the 1-hydroxybenzotriazole ester using DCC/HOBt. The reaction was accomplished in situ without isolation of the pure active ester. Compound **1** was obtained in excellent yields (>90%) employing a slight excess of HOBt (10%) instead of the recommended 50% excess. The choice of the same protecting group for both amino acids was based on the different sensitivity of *N*-trityl and *S*-trityl groups to the cleavage under acidic conditions.[12] The selective N-detritylation was achieved using acetic acid/water (5:1) on a steam bath and afforded recovery of the dipeptide **2** in almost quantitative yields by crystallization from diethyl ether. Compound **2** was conjugated to the succinimidyl ester of diphenylphosphinopropionic acid in degassed dichloromethane to complete the PN_2S coordination set obtaining PN_2S(Trt)-OH (**3**). The esterification of the cysteine carboxylic group was accomplished under mild conditions in a one-step procedure by using methanol, acting at the same time as solvent and reagent, in the presence of PyBOP and diisopropylethylamine as reported by Coste et al.[13] Although the esterification proceeded at room temperature, it was necessary to operate at low temperature (0−4 °C) to avoid side reactions which involved the formation of some aromatic products not characterized. Therefore, compound **4** was obtained in high yield (>90%) from purification by flash column chromatography (ethyl acetate) and crystallization from diethyl ether/petroleum ether. On the other side, the use of PyBOP and diisopropylethylamine enhanced the sensitivity of cysteine to racemization and optical rotation analysis of the final product confirmed the presence of the racemic mixture. Attempts to use different esterification conditions, as DCC/4-dimethylaminopyridine in methanol,[14] were unsuccessful. Since the specific chirality of cysteine is

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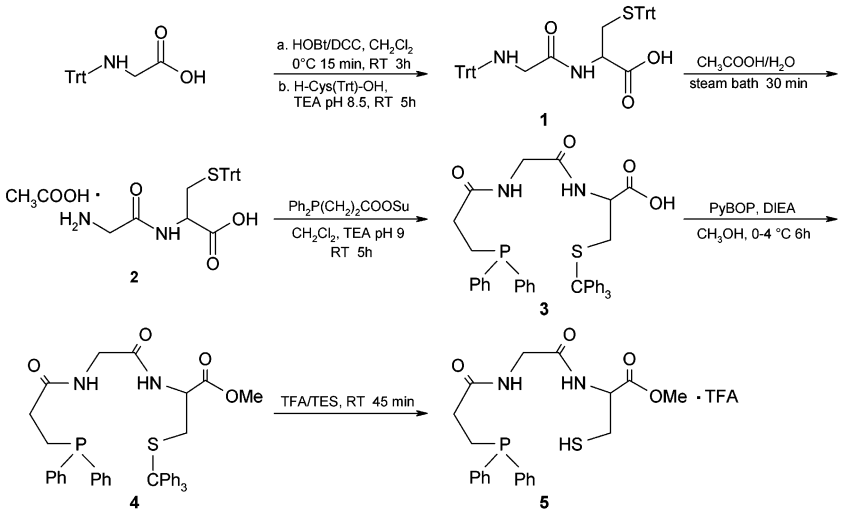
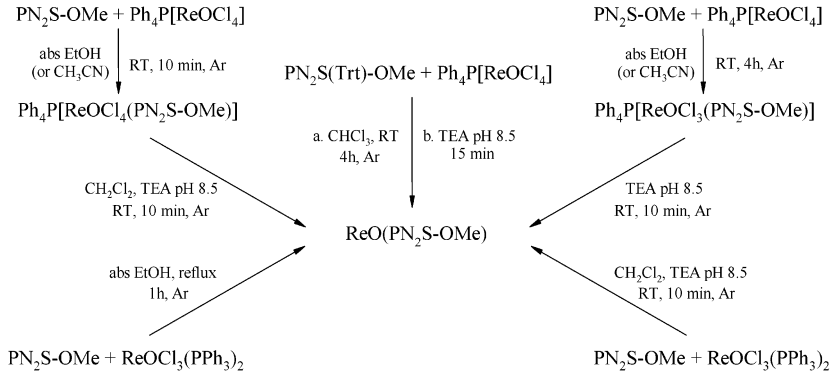
(10) Sheldrick, G. M. *SHELXL-97−Program for the Refinement of Crystal Structures*; University of Göttingen: Göttingen, Germany, 1997.
(11) (a) Matsoukas, J.; Tsegenidis, Th.; Cordopatis, P.; Theodoropoulos, D. *Tetrahedron* **1984**, *40*, 1869−1872. (b) Barlos, K.; Papaioannou, D.; Theodoropoulos, D. *Int. J. Pept. Protein Res.* **1984**, *23*, 300−305.
(12) Zervas, L.; Photaki, I. *J. Am. Chem. Soc.* **1962**, *84*, 3887−3897.
(13) Coste, J.; Campagne, J. M. *Tetrahedron Lett.* **1995**, *36*, 4235−4256.
(14) Hassner, A.; Alexanian, V. *Tetrahedron Lett.* **1978**, *19*, 4475−4478.

Scheme 1. Syntheses of Compounds **4** and **5**



Scheme 2. Syntheses of Compounds **9a** and **9b**



of no chemical consequences, the PyBOP-mediated method was preferred and the racemic mixture was employed for the following coordination reactions without separation of the two enantiomers.

The PN_2S(Trt)-OMe ligand, water insoluble, shows high solubility in methanol, ethanol, dichloromethane, chloroform, and acetonitrile. It was characterized by elemental analysis, NMR, FT-IR, and UV−visible spectroscopy, and electrospray mass spectrometry. ^{1}H NMR and ^{13}C NMR data are listed in Tables 1 and 2, respectively.

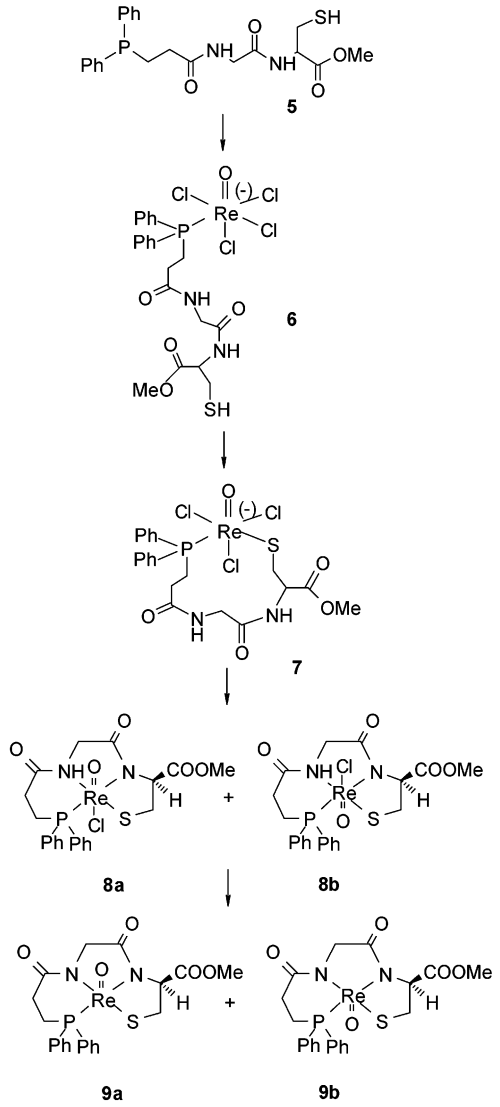
The cleavage of the trityl group before coordination reactions was achieved with trifluoroacetic acid and triethylsilane.[15] The reaction was complete in 45−60 min, and the residue obtained by removal of trifluoroacetic acid was immediately employed in the reactions with rhenium(V) oxo starting materials. The detritylated PN_2S-OMe (**5**) is soluble in most organic solvents and fairly soluble in basic water solution, as demonstrated by TLC analysis.

Synthesis of Rhenium Complexes. The coordination reactions were performed using both PN_2S(Trt)-OMe and PN_2S-OMe with either $ReOCl_3(PPh_3)_2$ or $Ph_4P[ReOCl_4]$ as Re^VO starting materials (Scheme 2). The ligands and the Re^VO precursors were always used in equimolar quantities.

The reaction of PN_2S-OMe with $Ph_4P[ReOCl_4]$, carried out in absolute ethanol or acetonitrile at room temperature, afforded an explanation of the coordination mechanism of the PN_2S-OMe ligand around the ReO^{3+} core (Scheme 3). In both solvents a bright green precipitate (**6**) separated after a few minutes. Compound **6** undergoes complete hydrolysis in the presence of moisture, and it was not possible to add triethylamine at this point without obtaining a dark solution due to the decomposition of the intermediate. The powder was supposed to have the formulation $Ph_4P[ReOCl_4(\mathbf{P}N_2S\text{-}OMe)]$, the phosphine phosphorus being the only atom of the chelating system bound to the metal center. When the suspension was stirred and degassed for some hours, compound **6** was affected by a heterogeneous phase conversion to a matt green precipitate (**7**) insensitive to hydrolysis. In this case the coordination of PN_2S-OMe went on involving the cysteine sulfur as thiolate with the formation of $Ph_4P[ReOCl_3(\mathbf{P},N_2,\mathbf{S}\text{-}OMe)]$. The formulation is supported by chlorine elemental analysis (calcd 9.85, found 9.33). By addition of triethylamine till pH 8.5 the suspension turned to a reddish-brown solution supposed to be due to the deprotonation of both amido groups holding the final ReO-(**PN_2S**-OMe) complex. Alternatively, compound **6** was recovered by filtration and dissolved in dichloromethane, resulting in a green-orange solution. In this case no intermediate compounds were isolated. The dissolution caused

(15) Pearson, D. A.; Blanchette, M.; Baker M. L.; Guindon C. A. *Tetrahedron Lett.* **1989**, *30*, 2739−2742.

Scheme 3. Coordination Mechanism in the Synthesis of Compounds **9a** and **9b**



the proceeding of the coordination which was completed increasing pH to 8.5.

The reaction of PN_2S(Trt)-OMe with $Ph_4P[ReOCl_4]$ was performed in chloroform at room temperature. The TLC analysis of the reaction mixture suggested that the cleavage of trityl group (9:1 $CHCl_3/CH_3OH$: $R_f = 0.98$) was accomplished during the coordination of the ligand to the $Re{=}O^{3+}$, core and this is in agreement with the acidic contribution of the metal in the mechanism of sulfur detritylation. The complete deprotection occurred in 4 h without isolation of assailable intermediates. Finally the pH was increased to 8.5 by addition of triethylamine and the solution became reddish-brown in a few minutes.

The detritylated ligand was also employed in the coordination reactions with $ReOCl_3(PPh_3)_2$ either in absolute ethanol or in dichloromethane. The reaction in absolute ethanol was achieved under reflux for increasing the solubility of the rhenium(V) oxo precursor. The initially green mixture turned to a reddish-brown color in 1 h. These reaction conditions avoided the necessity to add a base since the amide deprotonation was favored by heating. On the other hand, the heating induced a partial decomposition of the ligand, thus lowering the final yield (58%).

The reaction of **5** with $ReOCl_3(PPh_3)_2$ in dichloromethane was accomplished at room temperature due to quite good solubility of $ReOCl_3(PPh_3)_2$ in this solvent, and the proceeding of coordination favored its complete dissolution. The addition of triethylamine (pH 8.5) determined a rapid change of the solution color from a greenish-yellow to the final reddish-brown. The coordination took place in a short time (almost 10 min) and with the highest yield (73%).

In both reactions of **4** and **5** with $ReOCl_3(PPh_3)_2$ or $Ph_4P[ReOCl_4]$ at room temperature, if the final product was recovered in a short time after triethylamine addition, it was accompanied by a variable amount of a brown powder (**8**) insoluble in absolute ethanol. The brown compound dissolved in chloroform or dichloromethane at pH 8.5−9, leading slowly to the final ReO(**PN_2S**-OMe) complex. The conversion was sped up by increasing the pH to 10−11. Compound **8** was supposed to have the formulation ReOCl(**P**,**N**,**N**H,**S**-OMe)] in which one of the two amide nitrogens is still protonated and a chlorine trans to the oxo oxygen compensates for the residual positive charge on the metal. The formula is supported by chlorine elemental analysis (calcd 5.21, found 4.99) and by its transformation to the final complexes by increasing pH.

All of the above-described methods led to the same final reddish-brown solution containing two lipophilic and neutral compounds (**9a**, **9b**) detected by TLC (9:1 $CHCl_3/CH_3OH$: $R_f = 0.74$, **9a**; 0.69, **9b**). The compounds were recovered as a mixture by precipitation with diethyl ether/petroleum ether. The NMR and HPLC analysis of the mixture indicated that they were always produced in a nearly 1:1 molar ratio, independently from the synthetic method confirming that **9a** and **9b** retain the same formation probability. Attempts to separate them by fractional crystallization failed due to the almost equal solubility properties. Semipreparative RP-HPLC was employed successfully: method 2 HPLC conditions (see Experimental Sections) allowed their separation with a retention time difference greater than 2 min (fraction 1:, $R_t = 9.15$ min, **9a**; fraction 2, 11.45 min, **9b**). Both fractions were recovered as green powders. Fraction 1 crystallized from warm acetonitrile gave purple crystals suitable for X-ray analysis. Fraction 2 crystallized from methanol/diethyl ether by slow evaporation afforded green thin sheet crystals unsuitable for X-ray work. The compounds are soluble in acetonitrile, chloroform, dichloromethane, ethanol, and methanol, slightly soluble in diethyl ether (**9a** more than **9b**), and insoluble in petroleum ether and aqueous solutions. They are indefinitely stable in the solid state at room temperature and stable for a period of weeks in organic or organic/aqueous solutions, and they do not convert, as shown by NMR and HPLC analysis. In addition, **9a** and **9b** showed high stability in a wide range of pH (3−12). They were characterized using elemental analysis, NMR, FT-IR, and UV−visible spectroscopy, and electrospray mass spectrometry.

Spectrometry and Spectroscopy. In the positive ion electrospray mass spectra of **9a** and **9b** the same peak at

m/*z* 631 ([M$^+$]) was detected. The latter exhibited the characteristic rhenium(V) oxo isotopic pattern and confirmed the presence of a mononuclear monoligand complex.

The infrared spectra showed an absorption band at 989 and 987 cm^{-1} for **9a** and **9b**, respectively, which correspond to the characteristic Re═O stretching vibration within the accepted range for five-coordinated rhenium(V) oxo complexes (945−1067 cm^{-1}).[16] These frequencies are lower than that observed for the ReOCl[PN$_2$S(Bzl)-OMe] analogue (968 cm^{-1}), due to the higher strength of the Re═O bond when the trans position is unoccupied. The bands at 1743 cm^{-1} (**9a**) and at 1744 cm^{-1} (**9b**) were assigned to the methyl ester $\nu_{C=O}$, and the strong broad bands at 1656 cm^{-1} (**9a**) and 1654 cm^{-1} (**9b**) were assigned to the $\nu_{C=O}$ of both amides lumped together. The latter values were higher than the corresponding bands observed in the free ligand (1650 and 1642 cm^{-1}) and in the benzyl derivative (1651 and 1619 cm^{-1}) spectra.

FT-IR spectroscopy was useful also to characterize compounds **7** and **8**. The IR spectrum of **7** exhibited three bands at 1737, 1669, and 1654 cm^{-1} assigned to the methyl ester and to the amides $\nu_{C=O}$, respectively. Multiple strong bands were detected in the range 1560−1500 cm^{-1}, confirming that the amides were still protonated. In fact this range is known as the region of secondary amide II bands due to the coupling of NH bending and C−N stretching vibrations.[17] A strong band at 906 cm^{-1}, related to $\nu_{Re=O}$, indicated the presence of the hexacoordinated rhenium(V) oxo complex. The bands at 1440, 1108, 993, 751, 723, and 689 cm^{-1} corresponded to the PPh$_4^+$ moiety. These data were in accordance with the Ph$_4$P[ReOCl$_3$(**P**,N$_2$,**S**-OMe)] formulation.

The IR spectrum of compound **8** showed the methyl ester $\nu_{C=O}$ at 1735 cm^{-1} and the $\nu_{Re=O}$ at 975 cm^{-1}. Two very strong broad bands were detected in the range of carbonyl amide frequencies (1653, 1638 cm^{-1}) and four strong bands at 1561, 1541, 1523, and 1508 cm^{-1}. Differently from compound **7**, no frequencies related to the PPh$_4^+$ were shown. These data established the presence of a mixture of two compounds (**8a**, **8b**), both in accordance with the ReOCl(**P**,**N**,**N**H,**S**-OMe)] formulation.

The ^{31}P NMR spectra of **9a** and **9b** showed a singlet at +28.36 and +28.69 ppm, respectively. These values are significantly downfield referred to the chemical shift of the phosphine phosphorus in the PN$_2$S(Trt)-OMe (−15.46 ppm), confirming its coordination to the Re═O^{3+} core.

^{1}H NMR and ^{13}C NMR spectra of compounds **9a** and **9b** were recorded in CDCl$_3$ at room temperature. Complete assignment of resonances, reported in Tables 3 and 4, was based on data obtained from single-resonance proton−proton decoupling, NOESY and HETCOR experiments. The two ^{1}H NMR spectra showed analogous spin−spin systems. The β protons of the Cys residue were detected at 3.24 and 3.83 ppm for **9a** and to 3.68 and 4.13 ppm for **9b**, strongly downfield shifted compared to the same protons of PN$_2$S(Trt)-OMe (2.63 and 2.70 ppm). In the same way the Cys α proton was deshielded from 4.48 ppm in the uncoordinated ligand spectrum to 5.74 and 5.43 ppm for **9a** and **9b**, respectively. A reasonable explanation of these changes in ^{1}H NMR resonances is that the Cys protons of **9a** and **9b** were included in a chelate ring due to the coordination of the cysteine N$_{amide}$ and S$_{thiolate}$ atoms and were affected by the magnetic anisotropy induced by rhenium. The same suggestion resulted from the ^{13}C NMR assignments. In particular the Cys β carbons, at 45.8 and 48.6 ppm for **9a** and **9b**, respectively, showed a relevant deshielding compared to PN$_2$S(Trt)-OMe Cys β carbons (33.8 ppm) being more influenced by the coordination of the cysteine sulfur. In addition, the splitting pattern of the **9b** Cys α proton appeared as a doublet (^2J = 7.5 Hz) indicating that it was at a nearly right angle to one of the β protons, thereby giving no coupling constant. In the **9a** spectrum it appeared as a doublet of doublets (^2J = 7.9, ^3J = 2.1 Hz) indicating that the torsion angles between the Cys α and β protons were different from those in **9b**. The Gly α protons signals, showing in the PN$_2$S(Trt)-OMe spectrum the same chemical shift (3.84 ppm) and the same coupling constant with the amido proton (4.8 Hz), were detected in **9a** and **9b** spectra as two deshielded doublets (4.64 and 4.88 ppm, **9a**; 4.75 and 4.90, **9b**), with a geminal coupling of 19.5 and 19.8 Hz for **9a** and **9b**, respectively. A possible explanation of these changes in ^{1}H NMR resonances is the coordination of the deprotonated glycine and cysteine N$_{amide}$, leading to a second chelate ring with the two Gly protons differently oriented with respect to the oxygen of the Re═O group. Moreover, the deprotonation of the amido groups was proved by the lack of the signals related to their protons found at 6.10 and 6.27 ppm in the free ligand spectrum and by the absence of the couplings with Cys and Gly α protons. The amido carbons (185.6 and 180.9 ppm, **9a**; 188.3 and 180.1 ppm, **9b**) appeared significantly downfield, more than 10 ppm, related to the free ligand. Upon coordination the deprotonated nitrogens leave their electronic density less available to participate in resonance delocalization with the carbonyl groups. Their coordination to the central Re═O^{3+} core seriously influenced the glycine α carbon chemical shift deshielding the singlet from 43.2 ppm in PN$_2$S(Trt)-OMe spectrum to 61.4 and 61.2 in **9a** and **9b**, respectively.

The coordination of phosphorus and glycine N$_{amide}$ atoms was also established by the analysis of the diphenylphosphinopropionic proton and carbon resonances. The aromatic proton signals in the PN$_2$S(Trt)-OMe spectrum appeared as a single multiplet (7.43−7.18 ppm) which was split in three different multiplets in either the **9a** or **9b** spectra in the range 7.22−7.91 and 7.09−8.04 ppm, respectively. The presence of six doublets (due to the C−P couplings) related to the aromatic carbons in the spectra of both compounds suggested that the two phenyl rings were freely rotating around the C$_{ipso}$−P bound. As a consequence the two C$_{ortho}$ in the same ring were magnetically equivalent, as well as the two C$_{meta}$. Moreover, the C$_{meta}$ of both rings were overlapped. Finally,

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C_{meta} signals of compounds **9a** and **9b** were detected at the same chemical shift, as well as those of C_{para}, since these carbons were less influenced by the environment changes induced by coordination. On the basis of these considerations and the observed C−P coupling constants, the signals were assigned as reported in Table 4. The carbon assignments were in accordance with the correlations shown in the HETCOR spectra. The middle multiplet in the ^{1}H NMR, integrating for six protons, was assigned to the contributions of H_{meta} and H_{para} of both rings, the upfield multiplet (integrating for two protons) to H_{ortho} of one of phenyl rings, and the downfield multiplet (integrating for two protons) to the H_{ortho} of the other ring. From the comparison between **9a** and **9b** spectra it arose that the main difference was in the C_{ortho} and H_{ortho} of both rings.

The methylene protons in the free ligand spectrum exhibited similar chemical shifts, resulting in a single array of overlapping resonances in the range 2.41−2.25 ppm, complicated by the couplings with vicinal protons and phosphorus. In the **9a** and **9b** spectra, these signals appeared as two different deshielded multiplets owing to the formation of a chelate ring which imposed a rigidity onto the P−CH_2−CH_2−C(O)N backbone. In the **9a** spectrum, the multiplet at 2.60−2.51 (integrating for one proton) showed NOE correlation with the multiplet at 3.18−3.02 ppm (integrating for three protons) and with the upfield ortho aromatic protons, and a HETCOR correlation with the carbon at 23.3 ppm; in addition, the multiplet at 3.18−3.02 ppm showed a NOE interaction with the downfield ortho aromatic protons and with both carbons at 23.3 and 33.1 ppm. On the basis of these considerations the signals at 2.60−2.51 ppm were assigned to one of the protons α to the phosphorus atom and the multiplet at 3.18−3.02 to the remaining protons. According to this, the carbon resonances at 23.3 and 33.1 ppm were associated with the carbon α and β to the phosphorus atom, respectively. Similar arguments were employed in the assignment of the signals at 2.67−2.55 ppm and 3.25−3.08 ppm shown in the **9b** spectrum, and of the related carbons. The collected ^{1}H NMR and ^{13}C NMR data suggested that the most significant structural difference in the two compounds was related to the cysteine residue.

The UV−vis spectra recorded in chloroform showed a weak band at 509 nm for **9a** and 502 nm for **9b**; these values exhibited a bathochromic shift in *N*,*N*-dimethylformamide to 450 and 447 nm, respectively, and they are probably due to a d → d transition resulting in a brownish-red color.

The data from NMR, FT-IR, UV−vis, HPLC, and ES-MS indicated that **9a** and **9b** were two pentacoordinated rhenium(V) oxo diastereomers and that the main difference concerned the cysteine moiety. On the basis of the two possible orientations of the methyl ester group with respect to the oxygen of the Re═O moiety they were identified as ReO(PN_2S-OMe) syn and anti isomers. These isomers are often observed in Tc(V) and Re(V) complexes with tetradentate amino/amido-thiolato ligands.[18] Moreover, the unobserved syn/anti interconversion is consistent with the literature data on N_2S_2- and N_3S- type complexes containing only amido N donors, and it gives a reason for the origin of the two isomers from the nonreversible coordination reaction mechanism.

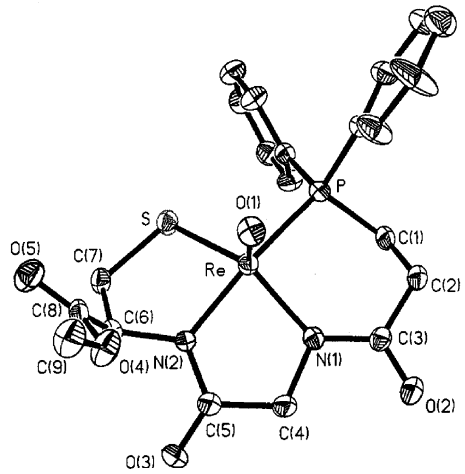

Figure 1. A perspective view of the asymmetric unit of complex **9a** with its numbering scheme. Thermal ellipsoids are drawn at the 50% probability level.

Compounds **7** and **8** were not completely characterized since during the analysis in solution they converted to **9a** and **9b** complexes. On the other hand, on the basis of the results obtained for **9**, compound **8** was identified as a mixture of syn and anti isomers of the ReOCl(**P**,**N**,**N**H,**S**-OMe)] complex.

X-ray Crystallography. Figure 1 shows an ORTEP[20] view of the monomeric neutral complex **9a** and the numbering scheme used. In this molecule the coordination at the rhenium is distorted square pyramidal. The apical position is occupied by the oxo group, whereas the vertices of the basal square plane are occupied by the deprotonated nitrogen atoms, by the phosphorus of the linked phosphine, and by the deprotonated sulfur atom of the cysteine. The atoms defining the basal plane lie alternatively above and below (±0.15 Å) the PN_2S mean plane, while the rhenium atom deviates from the same plane by 0.71 Å toward the apical O(1). This value is in agreement with the displacement normally observed for square pyramidal rhenium complexes,[18a] and it is greater than that observed in the analogous hexacoordinated ReOCl[PN_2S(Bzl)-OMe] (0.34 Å). These deviations lead to O(1)−Re donor angles well above 90°, the expected value for a regular square pyramidal geometry, as reported in Table 6. The methyl ester moiety adopts a syn arrangement with respect to the Re═O bond, and the

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mean plane passing through the same methyl ester group defines an angle of 69.3° with the PN_2S mean plane. In ReOCl[PN_2S(Bzl)-OMe] the methyl ester is also syn-arranged with respect to the Re═O bond, and the angle with the mean basal plane is 78.3°. The two phenyl rings are roughly perpendicular to the PN_2S plane; in fact the mean planes passing through them make angles of 68.3° and 84.2° with the pyramidal basal plane, respectively. The same planes define an angle to each other of 120.7°. The bond distances are within the expected limits and do not present any special features, with the exception of the Re−S distance. The latter is 2.268(5) Å, and it is far shorter than that reported for the benzyl compound, 2.485(6) Å. However, in ReOCl[PN_2S-(Bzl)-OMe] the tetradentate ligand is dianionic and the sulfur atom acts as a neutral ligand, while in the present study the ligand is trianionic and the sulfur acts as thiolate. The ligand forms a three (6,5,5)-membered chelate ring system around the rhenium atom, a situation found also in the benzyl derivative. The six-membered ring shows a *twist-boat* (D_2) arrangement, the Re−N(1)−C(4)−C(5)−N(2) ring tends to be planar, with a small *envelope* (C_s) deviation of the ring from planarity (pertinent greatest torsion angle of 13.7°), and the Re−N(2)−C(6)−C(7)−S assumes clearly the *envelope* (C_s) arrangement. The torsion angles (τ) of the six-membered ring vary between the −49.3° of C(1)−C(2)−C(3)−N(1) and +80.0° of P−C(1)−C(2)−C(3). In the middle ring τ ranges between −13.1° and +13.7°; finally, in the ring including sulfur τ is comprised within −33.2° and +47.4°. In the analogous ReOCl[PN_2S(Bzl)-OMe] the three rings assume a *boat* (C_{2v}), a roughly planar, and a *twist-envelope* (C_2) conformation, respectively. With respect to cell packing, no significant intermolecular interactions were detected. The centrosymmetric space group $P\bar{1}$ indicates the presence in the cell of the two enantiomers of **9a** as a consequence of the PN_2S(Trt)-OMe ligand racemization.

Theoretical vicinal coupling constants, obtained by entering torsion angles (Φ) measured from the X-ray crystal coordinates into a Karplus-type equation ($^3J = 11.0 \cos^2 \Phi - 1.4 \cos \Phi + 1.6 \sin^2 \Phi$),[21] were compared to experimental NMR data to differentiate cysteine β protons as endo (facing toward the oxygen of the Re═O core) and exo (remote from the oxygen of the Re═O core). The torsion angles for $HC_\alpha C_\beta H_{endo}$ and $HC_\alpha C_\beta H_{exo}$ were 90.52° and −27.80°, respectively, and the related calculated coupling constants 1.6 and 7.7 Hz. These values were consistent with the coupling constants observed in the ^{1}H NMR, and on the basis of this consideration the signal at 3.83 ppm ($^3J = 2.1$) was assigned to the endo and the signal at 3.24 ppm ($^3J = 7.9$) to the exo proton. The assignments were in accordance with the literature-reported data on oxo−rhenium complexes,[22] indicating that protons spatially close to the oxygen of the Re═O are deshielded relative to those remote from the oxygen.

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Conclusions

We have shown that PN_2S *N*-[*N*-(3-diphenylphosphinopropionyl)glycyl]-*S*-tritylcysteine methyl ester ligand exhibits high chelating affinity for rhenium(V) oxo. Two lipophilic and neutral complexes were produced in a 1:1 molar ratio by reacting tritylated or detritylated ligand with Re^VO precursors. They displayed high stability both in the solid state and in solution, and no interconversion was detected. Furthermore the reaction of PN_2S-OMe with $Ph_4P[ReOCl_4]$ in absolute ethanol or acetonitrile at room temperature afforded the possibility of investigating the coordination mechanism. The isolation of some intermediates suggested that phosphorus is the first atom which binds the metal center followed by the sulfur and the two protonated nitrogens of the amido groups. After deprotonation, the two ReO(PN_2S)-OMe isomers are the final and most stable complexes. The supposed mechanism enhances the utility of the PN_2S bifunctional chelating ligand in labeling bioactive molecules with $^{186/188}$Re by means of the post conjugation labeling strategy. In fact, the conjugation of the PN_2S ligand through the cysteine carboxylic group to a terminal amino group of a biomolecule orients Re radiotracers in the labeling reaction to the phosphine phosphorus, avoiding competitive coordination of other sets present in the biomolecule. On the other hand, the high stability of ReO(PN_2S)-OMe allows one to use the complex in the conjugation reaction to the biomolecule after labeling, required in the case of biomolecule instability under labeling conditions.

Supporting Information Available: Tables of X-ray crystallographic data in crystallographic information file (CIF) format, including full X-ray data, fractional atomic coordinates and thermal parameters, and bond lengths and angles. This material is available free of charge via the Internet at http://pubs.acs.org.

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