



	<b>Experiment title:</b> EXAFS studies of [ <sup>99</sup> Tc] technetium carbonyl complexes	<b>Experiment number:</b> LS - 1395
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<b>Names and affiliations of applicants</b> (* indicates experimentalists): S. Seifert* <sup>1)</sup> , J.-U. Kuenstler* <sup>1)</sup> , T. Reich* <sup>2)</sup> , H. Funke* <sup>2)</sup> , C. Hennig* <sup>2)</sup> , A. Rossberg* <sup>2)</sup> , B. Johannsen <sup>1)</sup> <sup>1)</sup> Institute of Bioinorganic and Radiopharmaceutical Chemistry <sup>2)</sup> Institute of Radiochemistry Research Center Rossendorf P.O. Box 510119, 01314 Dresden, Germany		

## Report:

Nowadays, most of all radiopharmaceuticals used in routine nuclear medicine imaging procedures are <sup>99m</sup>Tc labelled compounds, and intensive research is going on to design new <sup>99m</sup>Tc radiopharmaceuticals. Complexes with N-donor or S-donor chelating ligands coordinated to the Tc(I) or Re(I) tricarbonyl moiety with high stability in aqueous solution may serve for the design of radiopharmaceuticals. Coordination of peptides with the [Tc(CO)<sub>3</sub>]<sup>+</sup> moiety is of great interest for the design of <sup>99m</sup>Tc labelled peptides [1, 2].

The aim of this work was to investigate the stability of model Tc(I) thioether tricarbonyl complexes in aqueous solution, their reactivity against histidine and to estimate structural parameters of the dissolved compounds. The reaction of the precursor [Tc(CO)<sub>3</sub>Cl<sub>3</sub>]<sup>2-</sup> with histidine, glycine and simple model peptides was studied. The possibility of detecting other than the ligating atoms of the amino acids and peptides was to be checked by Tc K-edge EXAFS analysis.

Due to the extremely small concentration of the radionuclide present in the radiopharmaceutical preparation (10<sup>-6</sup> – 10<sup>-8</sup> M), XAS studies as well as most of chemical investigations employ the long-lived isotope <sup>99</sup>Tc. We prepared the complexes [Tc(CO)<sub>3</sub>ClL<sup>1</sup>], **Tc1**, [Tc(CO)<sub>3</sub>(H<sub>2</sub>O)L<sup>1</sup>]<sup>+</sup>, **Tc1a**, [Tc(CO)<sub>3</sub>L<sup>2</sup>], **Tc2**, and as reference [Tc(CO)<sub>3</sub>His], **Tc3**, both at <sup>99</sup>Tc and <sup>99m</sup>Tc levels using 3,6-dithiaoctane, L<sup>1</sup>, and 1-carboxy-3,6-dithiaheptane, L<sup>2</sup>, as prototypical bidentate thioethers without and with an additional donor atom in the chelating unit. <sup>99</sup>Tc solutions of **Tc1** and **Tc1a** were prepared by dissolving solid **Tc1** in water/methanol and adding of Cl<sup>-</sup> or Ag<sup>+</sup>, respectively. With an excess of chloride the equilibrium between **Tc1** and **Tc1a** is on the side of the **Tc1** complex. If chlorine is removed from the equilibrium by precipitation as AgCl, **Tc1a** is the solution species. For studying a possible substitution of the thioether ligands in the complexes **Tc1**

and **Tc2** by histidine, an excess of the challenge ligand was given to the complex solutions; the resulting complexes were named **Tc1b** and **Tc2b**. The samples **TcGlyHis**, **TcHisGly**, **TcGlyGlyGly**, **TcHisGlyGly**, **TcGlyGlyHis**, **TcGlyHisLys**, **TcGly** and **TcGlyGly** were prepared by dissolving the precursor complex  $(\text{NET}_4)_2[\text{Tc}(\text{CO})_3\text{Cl}_3]$  in water/methanol (1/1) and adding Gly-His, His-Gly, Gly-Gly-Gly, His-Gly-Gly, Gly-Gly-His, Gly-His-Lys (in an approximately 1.2-fold excess) and Gly, Gly-Gly (approximately 2-fold excess).

We applied Tc K-edge EXAFS measurements (Tc K-edge at 21044 eV), chromatography (UV and  $\gamma$ -ray detection), electrophoresis and mass spectroscopy to investigate the behaviour of the complexes. EXAFS analyses (transmission mode, room temperature, Tc concentration  $\sim$  0.01 to 0.02 M) were carried out at the Rossendorf beamline (ROBL). To obtain a satisfactory evaluation of the EXAFS spectra and the Fourier transforms, single-scattering paths including the C, O, N/O, and S/Cl coordination shells and multiple-scattering paths to model the oxygen atom of the carbon monoxide ligand were taken into account.

We found that the complex **Tc1** is reversibly transformed into the complex **Tc1a** in aqueous solution. Tc K-edge EXAFS measurements confirmed the composition of the compounds. The estimated atomic distances are 1.92 Å (Tc-C), 2.49 Å (Tc-S or Tc-Cl), 2.19 Å (Tc-O<sub>aq</sub>) and 3.06 Å (Tc-O<sub>CO</sub>). EXAFS analysis of **Tc1a** shows the same structural parameters for the C, S/Cl and CO coordination shells as **Tc1** except that the coordination number for the S/Cl coordination shell is lowered by approximately one. In addition, the detection of  $1.2 \pm 0.3$  oxygen atoms at 2.19 Å proves the substitution of chlorine by water.

We found that the complexes **Tc1** and **Tc2** react with histidine to the products **Tc1b** and **Tc2b** in aqueous solution which are identically with the reference **Tc3**. The estimated atomic distances in **Tc1** (see above) and **Tc2** (1.92 Å (Tc-C), 2.21 Å (Tc-O), 2.50 Å (Tc-S) and 3.07 Å (Tc-O<sub>CO</sub>)) are almost the same as those found by single crystal X-ray diffraction analyses of similar Re carbonyl complexes containing dithioether ligands [2]. The estimated coordination numbers agree with the expected values. The results of the EXAFS measurements of **Tc1b** and **Tc2b** are almost identical to the results of EXAFS analysis of reference **Tc3** (atomic distances: 1.91 Å (Tc-C), 2.20 Å (Tc-N and Tc-O) and 3.06 Å (Tc-O<sub>CO</sub>)). This fact proves the substitution of bidentate dithioether and tridentate carboxylato dithioether ligands by histidine, which coordinates in a tridentate manner. The obtained atomic distances are in agreement with those found for the Re congener by single crystal X-ray diffraction analysis [3]. Detection of any other than the ligating atoms of the thioether ligands or histidine was not possible mainly because of overlapping with the very strong backscattering off the three oxygen atoms of carbon monoxide, which is enhanced by a strong multiple-scattering effect.

The products of the reaction of the precursor  $[\text{Tc}(\text{CO})_3\text{Cl}_3]^{2-}$  with histidine, glycine and small peptides containing these amino acids were studied in solution. Tc K-edge EXAFS measurements confirmed the exchange of chloride, the mononuclear structure and typical structural parameters expected for such type of complexes (atomic distances: 1.91 Å (Tc-C), 2.20 Å (Tc-N and Tc-O) and 3.06 Å (Tc-O<sub>CO</sub>)). For all complexes, the same local structure was observed. Detection of any other than the ligating atoms of the amino acids or peptides was not possible.

#### References:

- [1] R. Alberto et al., *Coordination Chem. Rev.* **190-192** (1999) 901.
- [2] H.-J. Pietzsch et al., *Bioconj. Chem.* **11** 414 (2000) 414.
- [3] R. Schibli et al., *Bioconj. Chem.* **11** (2000) 345.