

PRELIMINARY NOTE

A Survey of Reducing Agents for the Synthesis of Tetraphenylarsonium Oxotechnetiumbis(ethanedithiolate) from [^{99}Tc] Pertechnetate in Aqueous Solution

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We have studied the effectiveness of various reducing agents in the production of the well-characterized complex [$^{99}\text{TcO}(\text{SCH}_2\text{CH}_2\text{S})_2$] $^-$ from pertechnetate and ethanedithiol in aqueous solution. The reductants tested included sodium dithionite, hypophosphorous acid, formamidine sulfinic acid, dithiothreitol, hydrazine, and hydroxylamine. Of these, only sodium dithionite in the pH range 11–13 was found to give quantitative yields of the required technetium complex.

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The reduction of TcO_4^- in aqueous solution in the presence of a suitable ligand is common to the preparation of most Tc-99m-labeled radiopharmaceuticals. Despite their extensive use in clinical medicine, however, the nature of the technetium species formed in these preparations is often unknown. In order to understand better the chemical behavior of the element, we are in the process of investigating ligand affinities and preferred stereochemistry in a variety of oxidation states. The recent synthesis and characterization of oxotechnetium (+5) complexes containing mercaptothioacetate (I) (1), ethanedithiolate (II) (2), and toluene-3,4-dithiolate (III) (3) show that technetium in the +5 oxidation state has a marked affinity for sulfur donor sets. X-ray crystallographic data in the case of both I and II indicate that an oxotechnetium center (TcO^{3+}) is bound in a virtually square pyramidal arrangement to four sulfur atoms in the basal plane. These results are significant because several radiopharmaceuticals (4–8) may contain this type of structural unit.

During the early phases of our work (1), the process of characterizing the complexes produced was simplified by using the thiolate ligand as both a reducing agent and

as a chelate. In order to offset the disadvantages of this approach as the investigation was extended to other sulfur-donor ligands, it was decided to compare the relative effectiveness of a variety of reductants in producing a known product (complex II) in aqueous solution beginning with [^{99}Tc] ammonium pertechnetate.

METHODS

All the materials used in this study were obtained commercially. The starting materials were aqueous ammonium pertechnetate ($\text{NH}_4^{99}\text{TcO}_4$, 0.33–0.35M)* and ethanedithiol.† The reducing agents tested included dithiothreitol, formamidine sulfinic acid, sodium dithionite, hydrazine monohydrate, hydroxylamine hydrochloride, 50% hypophosphorous acid, and stannous chloride.

For the reducing agent studies, standard aliquots of $^{99}\text{TcO}_4^-$ (0.15 mmol) and ethanedithiol (0.84 mmol) were added to 25 ml of distilled water. The reducing agent was then added, and the reaction allowed to take place. At completion, the tetraphenylarsonium salt of the required complex was precipitated by the addition of 0.34 mmol of tetraphenylarsonium chloride, the solution filtered, and the product recrystallized from acetone-water. The final yield was determined by weighing. The nature of the purified material was confirmed by

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infrared and optical spectroscopy by comparison with the data of Smith (2), using a grating spectrometer[†] and a recording spectrophotometer.[‡]

The primary goal was to find a reducing agent that would rapidly give quantitative yields of the desired complex at room temperature, in aqueous solution, and as close to physiologic pH as possible. Thus, many preparations were carried out with each of the chosen reducing agents under a wide variety of conditions. These included molar ratios of reductant to Tc-99 ranging from a maximum of 100:1 to 1:1 at pH values between 5 and 13, temperatures between ambient and 100°C, and reaction times of 10 min to 24 hr. In those cases where stannous chloride was used as the reducing agent, the molar ratio of Sn to ligand varied from 10:1 to 1:10, and the Sn:Tc ratio thus from 56:1 to 0.56:1.

RESULTS

Table 1 lists the optimum results obtained with each of the reducing agents used. The yields quoted are the highest observed in each case under the conditions specified. The last column indicates the other components present where the reaction proved to be less than quantitative.

Using sodium dithionite [(a) in Table 1] the reaction was found to be rapid at room temperature over the pH range 5.5 to 13, with a molar concentration of reductant ranging from that equivalent to a stoichiometric two-electron reduction up to a fivefold excess. Technetium dioxide was formed as a by-product up to pH 10, and the formation of the required complex was consistently quantitative only between pH 11 and 13.

With hypophosphorous acid (b), the highest yields of the desired product at room temperature were found with a molar excess of 16:1 over technetium and with a reaction time of 6 hr. Traces of TcO₂ were still formed at these optimum conditions. At lower molar concentrations, reduction was incomplete, as evidenced by the presence of unchanged TcO₄⁻ in the reaction mixture.

Formamidine sulfinic acid (FSA) (c) showed little tendency to reduce pertechnetate at room temperature, with an unidentified green complex being observed in solution both in the presence and absence of ethanedithiol. At 50°C complete reduction was achieved, but the highest yield of the desired product was 88%, with TcO₂ forming the remainder.

Dithiothreitol (d) produced significant yields of Complex II only with extended periods under reflux; at room temperature no detectable reduction occurred. The highest yield obtained was 87% at a pH of 5.5, with a fourfold excess of reductant and 6 hr under reflux. Even under these conditions, traces of unreduced pertechnetate could be detected in the product mixture.

With hydrazine (e), used in excess (35:1) at pH 7 and

TABLE 1. YIELDS OF TETRAPHENYLARSONIUM OXOTECHNETIUMBIS(ETHANEDITHIOLATE) OBTAINED WITH A VARIETY OF REDUCING AGENTS IN AQUEOUS SOLUTION

Reducing agent	Conditions	Yield (%)	Comments
a. Sodium dithionite			
(i) 0.46 mmol	pH 5.5 10 min, 22°C	13	TcO ₂ *
(ii) 0.46 mmol	pH 11.0 10 min, 22°C	100	—
b. Hypophosphorous acid			
(i) 0.23 mmol	pH 5.5 12 hr, 22°C	10	TcO ₂ , TcO ₄ ⁻ *
(ii) 0.92 mmol	pH 5.5 12 hr, 22°C	48	TcO ₂ , TcO ₄ ⁻
(iii) 2.30 mmol	pH 5.5 6 hr, 22°C	98	trace TcO ₂
c. Formamidine sulfinic acid (FSA)			
(i) 1.5 mmol	pH 7.4 30 min, 50°C	88	trace TcO ₂
d. Dithiothreitol (DTTE)			
(i) 0.47 mmol	pH 5.5 6 hr, reflux	87	trace TcO ₂ , TcO ₄ ⁻
e. Hydrazine			
(i) 5.0 mmol	pH 7 5 min, 22°C	70	TcO ₂ , TcO ₄ ⁻
f. Hydroxylamine			
(i) 2.0 mmol	pH 7 1 hr, 22°C	10	TcO ₄ ⁻

* TcO₄⁻ identified by ultraviolet spectroscopy; TcO₂ identified by reoxidation to TcO₄⁻.

with a reaction time of 5 min at room temperature, a maximum yield of 70% was achieved. Increasing the reaction time beyond 5 min led to increasingly higher levels of TcO₂ among the products as a result of the breakdown of the rust-colored Complex II.

Hydroxylamine (f) showed no great tendency to reduce pertechnetate, even at large molar excesses. Thus a 1-hr reaction at room temperature and pH 7 gave a maximum yield of 10%; increasing the temperature produced no change in the amount of coordination complex isolated.

The details of the reactions at various pH values as described are contained in Table 1.

Attempts to use stannous chloride as a reductant were not satisfactory in this particular system, even at ligand concentrations representing a 10:1 excess over stannous tin. Both in the presence and absence of pertechnetate, stannous ion and ethanedithiol formed an insoluble solid

that gave the following elemental composition by microanalysis⁸: Sn, 55.92%; C, 11.39%; H, 1.93%; S, 30.60%. Such an analytical formulation, indicating the presence of one ligand per metal atom, requires the species to be a polymer. In those cases where the molar ratio of Sn:Tc was approximately 2:1 or greater, complete reduction occurred, with the formation of a gray mass consisting of the insoluble polymer and about 80% of the technetium as the dioxide. The remaining 20% was incorporated into an as yet unidentified green complex, which could be recrystallized from methylene chloride-hexane mixtures. No pertechnetate could be detected among the products under these conditions.

DISCUSSION

The objective of these experiments was to find conditions under which the chemically characterized complex $[(C_6H_5)_4As][TcO(SCH_2CH_2S)_2]$ could be quantitatively prepared from $[^{99}Tc]$ pertechnetate in aqueous solution. In order to assess fully the relative effectiveness of the reducing agents chosen, it was essential to study a known end product. This contrasts with a previous study (9) in which complexes were prepared using Tc-99m. Note that in the present work the molar ratio of reductant to TcO_4^- was not as excessive as is currently found in radiopharmaceutical preparations.

The results show that several reducing agents give high yields of Complex II under a variety of conditions. Other than Complex II, the only other complexes observed were those in the FSA and stannous ion reductions. The unidentified green complex seen in mixtures of FSA and TcO_4^- in the absence of ethanedithiol lends support to the previous contention (9) that FSA may weakly bind technetium before the production of a more stable complex with an appropriate ligand. Stannous chloride was found to form an insoluble polymeric species with the ligand, and thus did not function adequately as a reducing agent in this system under the particular conditions investigated.

The fact that several of the reductants gave reasonable amounts of product in the pH range 5–8 is significant in that it demonstrates that a stable complex of technetium in the +5 oxidation state can be prepared under conditions close to physiologic pH. In terms of the original objective, however, quantitative yields of Complex II were obtained only with the free radical reductant dithionite at a pH greater than 11. At pH levels greater

than 10, no evidence of $TcO_2 \cdot xH_2O$ was observed with this agent. The most likely explanation for this observation is that the kinetics of reduction and chelation to yield Complex II are very rapid and thus suppress the competing hydrolysis reaction that is found at lower pH values. In other experiments conducted in the pH range 5–8, on the other hand, lower yields of the complex were obtained, together with significant amounts of the hydrated dioxide, clearly limiting the potential of this reductant at close to physiological pH.

FOOTNOTES

- * New England Nuclear Corp., Billerica, MA.
- † Eastman Kodak,
- ‡ Perkin-Elmer, PE 180,
- § Cary 17,
- ¶ Galbraith Laboratories Inc., Knoxville, TN.

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