

Physical Sciences Section

Pakistan J.Sci. Ind. Res., Vol. 27, No. 1, February 1984

REDUCTION OF ^{99}Tc IN THE PRESENCE OF STANNOUS CHLORIDE – CORRELATION OF CHROMATOGRAPHIC BEHAVIOR WITH THE U.V. / VISIBLE SPECTRA FOR THE IDENTIFICATION OF REDUCED SPECIES

S.A. Adelusi*

*Department of Pharmaceutical Chemistry, University of Strathclyde,
Glasgow G1 1XW, U.K.*

(Received May 30, 1983)

Chromatographic scanning was used to detect the position of activities of the reduced forms of ^{99}Tc and Rf values correlated with the U.V./visible spectra. The different species of ^{99}Tc that occur in radiopharmaceuticals when it is reduced by stannous chloride ($\text{SnCl}_2 \cdot 2\text{H}_2\text{O}$) appeared to be in the forms Tc(IV), Tc(V) and Tc(VII).

INTRODUCTION

Of the conveniently available radionuclide, technetium- ^{99m}Tc has by far the best nuclear properties for diagnostic imaging. With the advent of commercial generator systems, instant technetium, innovations in chelations, and new chelating agents, there has been a marked expansion in the use of ^{99}Tc labelled compounds. Chemical forms of ^{99}Tc are presently widely used radiopharmaceutical radionuclide imaging of the brain, liver, lung and skeleton.

A number of ^{99}Tc radiopharmaceutical preparations require the reduction of the pertechnetate ion (TcO_4^-), this reduction is often carried out using stannous chloride. Since the concentrations of ^{99m}Tc is very low in clinical work (10^{-9}M), meaningful analysis can be carried out only at higher concentration levels, using the carrier isotope ^{99}Tc .

The analytical methods that have been used to study the reduced forms of technetium-99 include polarography [1, 2 and 3], chromatography [4, 5 and 6]. None of these methods have proved useful in identifying the oxidation states of the reduced species of technetium-99 in radiopharmaceuticals. The present investigation, therefore, correlated the chromatographic behavior of ^{99}Tc chemical species with their U.V./visible spectra when $\text{NH}_4^{99}\text{TcO}_4$ is reduced by $\text{SnCl}_2 \cdot 2\text{H}_2\text{O}$ in order to know in what oxidation states technetium exists in radiopharmaceuticals.

MATERIALS AND METHOD

Ammonium pertechnetate ($\text{NH}_4^{99}\text{TcO}_4$) was purchased from the Radiochemical Centre, Amersham. It had a

*Present address: Department of Pharmacology and Therapeutics, University of Ibadan, Nigeria.

radioactive concentration of 0.63 mCi/ml on 1st April, 1976 ($t_{1/2} = 2.1 \times 10^5\text{h}$) and a chemical concentration of 36.9 mg. Tc per ml. The handling of this material was carried out in fume-cupboard with rubber gloves worn at all times. Solutions were not pipetted by mouth, there was no eating and hands were always washed before leaving the laboratory.

The stannous chloride solution and the 0.9% NaCl/0.1 M HCl buffer used in this investigation were purged with nitrogen for 30 mins before use.

10 microlitres of $\text{NH}_4^{99}\text{TcO}_4$ solution was taken with Hamilton syringes and placed in a scintillation vial. 10 ml. of 0.9 NaCl/0.1 M HCl buffer was added; 3 ml of this solution was taken into a U.V. cell (1 cm) and 10 microlitres of the solution spotted (in microlitre aliquots) onto Whatman 3MM paper. 10 microlitres of the stannous chloride solution was added to the cell content, the time of addition $t = 0$ was noted. 10 microlitres of this solution (after mixing) was immediately spotted on W 3 MM paper (in 1 microlitres aliquots).

The ultraviolet spectrum of the solution was determined before and after the addition of stannous chloride solution using the 0.9% NaCl/0.1 M HCl buffer as reference. The determination of the absorption spectrum and spotting of 10 microlitres were carried out at the following time: $t = 30\text{ mins}$, $t = 2\text{ hrs}$ and $t = 7\text{ days}$.

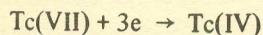
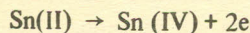
The spots were dried and the papers developed by the ascending elution technique in an atmosphere of nitrogen (oxygen free nitrogen) for 30 mins. After development, the papers were dried under I.R. light and scanned for radioactivity using the Panax for the detection of the reduced Tc species. The Rf values of the peaks that emerged were calculated.

RESULTS AND DISCUSSION

The pertechnetate ion in 9.0% NaCl/0.1 M HCl buffer absorbed at 246 and 289 nm before the addition of the stannous chloride solution and the solution was colorless (Fig. 1). It gradually turned yellow 30 mins after the addition of the stannous chloride solution. The yellow solution shows a new absorption in the U.V. region at 335 nm (Fig. 1). Stannous chloride solution in 0.9% NaCl/0.1 M HCl buffer absorbed in the U.V. region at 215 nm and hence the peak at 247 after the addition of stannous chloride solution was difficult to measure. The yellow solution was stable for several days.

The relative intensities and the Rf values of the peaks that emerged during the chromatographic scanning (Fig. 2) were calculated and the results tabulated in Table 1. The U.V./visible absorption maximum were also tabulated in this table.

The reduction of the pertechnetate ion in 0.9% NaCl/0.1 M HCl buffer with stannous chloride showed from the Rf values that there are 3 species of the reduced form Tc, the Rf values are 0.05, 0.73 and 0.92 for each spot. (Table 1), the Rf value of 0.73 is close to that obtained with the pertechnetate ion in 0.9% NaCl/0.1 M HCl buffer when no stannous chloride was added. This value (0.73) is probably due to the unreduced pertechnetate ion and the other Rf values of 0.05 and 0.92 must be due to the reduced forms of technetium. These values are close to the literature values for Tc(V) and Tc(IV) respectively [4]. The U.V. absorption spectrum of TcO_4^- showed maximum absorption at 247 and 289 nm and these values are close to the literature values [7,8]. The wavelength of maximum absorption (335 nm) obtained after the addition of stannous chloride solution (Fig 1) is close to the literature value of 338 nm for $TcCl_6^{=}$ [4]. It is, thus, possible that when the pertechnetate ion (TcO_4^-) is reduced in 0.9% NaCl/0.1M HCl buffer, $TcCl_6^{=}$ is formed in solution. The following reaction may be taking place:



The Tc(IV) probably formed a complex with HCl to give $TcCl_6^{=}$. The formation of this complex when Tc(VII) was reduced by ascorbic acid in the presence of HCl has been reported in the literature [2]. The absorption maximum of 338 nm was also reported for this complex. It can, therefore, be suggested that Tc(VII) was reduced to Tc(IV) in 0.9% NaCl/0.1M HCl buffer when reduction was carried

out with stannous chloride solution since the Rf value of 0.92 obtained in this work is close to the literature value of 0.90 for Tc(IV) [4] with the same chromatographic conditions.

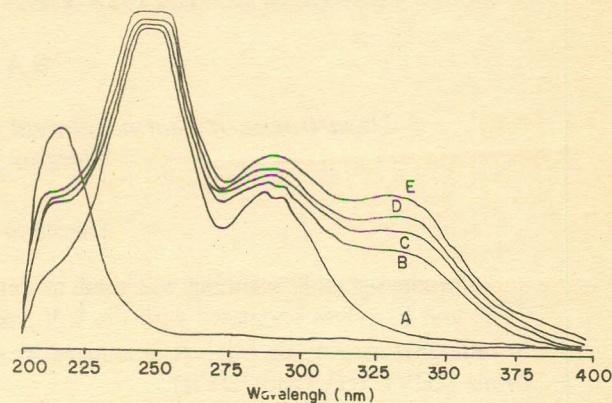


Figure 1:

The reduction of TcO_4^- with $SnCl_2$ in 0.9% NaCl/0.1M HCl in A represents the U.V. spectrum of TcO_4^- when no $SnCl_2$ solution was added

B represents the U.V. spectrum of TcO_4^- at time $t = 0$ min
 C " " " " " " " " " $t = 30$ min
 D " " " " " " " " " $t = 120$ min
 E " " " " " " " " " $t = 7$ days.

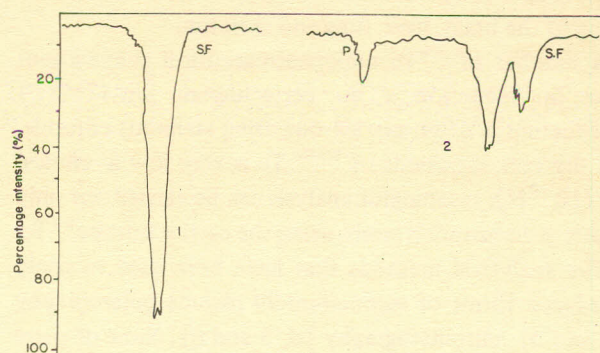


Figure 2

The radioactive scan of the chromatography paper for the detection of the activities of the reduced Tc species when TcO_4^- is reduced with $SnCl_2$ in 0.9% NaCl/0.1M HCl buffer.

1 represents the scan at time $T = 0$ min

2 represents the scan 7 days after the addition of $SnCl_2$ solution.

The scans for $t = 30$ and 120 min were the same as the one for the scan for 7 days.

S.F = solvent front,

P = point of application.

The variations in the Rf values obtained in this investi-

Table 1. Rf values of the reduced Tc species and the spectra data for the reduction of TcO_4^- with SnCl_2 in 0.9% NaCl/0.1M HCl buffer.

Time	Rf values	Relative intensities (%)	λ Maximum	Extinction
No SnCl_2	— 0.79 —	— 58 —	247 289	— 0.94
0 min.	0.07 0.75 —	6 40 —	247 289 335	— 0.95 0.6
30 min	0.07 0.79 0.97	17 35 39	247 289 335	— 0.97 0.7
120 min	0.06 0.76 0.92	14 39 50	247 289 335	— 0.98 0.8
7 days	0.05 0.77 0.91	8 70 35	247 289 335	— 0.98 0.7

gation (Table 1) for a particular spot is probably due to the fact that when Tc(VII) is reduced to different oxidation states, there are many possible products of each oxidation state due to the hydrolysis and complex formations, for example, the Tc(IV) has been shown to exist in solution as TcO_2 , TcCl_6^- , Tc(OH) , Tc(V)OCl_5^- and Tc(V)O(OH)Cl_4^- [4,2]. All these species will absorb differently in the U.V./visible region and will probably not have the same Rf values. The identification of the reduced species of Tc-99 when reduced by stannous chloride solution in Tc-99 radiopharmaceuticals is still faced with analytical problems because of various hydrolytic products that exist in solution during the reduction process; but to a good approximation it could be suggested from the results obtained in this investigation that the Tc-99 exists in the forms: Tc(VII), Tc(V) and Tc(IV) corresponding to the oxidations of 7, 5 and 4 when reduction is carried out with stannous chloride solution. The investigation also suggested that these various valency states of Tc-99 existing in solution during the reduction of pertechnetate ion with stannous chloride solution may be separated by ascending paper chromatography (using Whatman 3MM paper) using 0.3M HCl as solvent and the running time of at least 30 min. The paper can be scanned radioactively for the detection of the Tc-99 species by using the Rf values of the emerged

peaks.

It appears this will be a useful method for the determination of Tc-99 in Tc-99m radiopharmaceuticals.

Acknowledgements. I am grateful to Dr. T.L. Whately for allowing me to carry out this investigation in his laboratory at the Department of Pharmaceutical Chemistry of the University of Strathclyde, Glasgow during the preparation of my M.Sc thesis.

REFERENCES

1. R. Colton, J. Daiziel, W.P. Griffith and G. Wilkinson, *J. Chem. Soc.*, **1**, 71, (1960).
2. C. Bratu, G.H. Bratu, I. Galateanu, and M. Roman, *J. Radioanal. Chem.*, **26**, 5, (1975).
3. J. Steigman, G. Meinken, and P. Richards, *Int. J. Appl. Rad. Isot.*, **26**, 601 (1975).
4. S.K. Shukla, *J. Chromatog.*, **21**, 92 (1966).
5. L. Ossicini, F. Saracino, and M. Lederer, *J. Chromatog.*, **16**, 524 (1964).
6. P. Richard, *J. Inorg. Nucl. Chem.*, **38**, 817 (1976).
7. R.A. Armstrong, *Chemistry of Technetium in Low Oxidation States* Ph.D. thesis, Stanford University (1975).
8. G.E. Boyd, J.W. Cobble, C.M. Nelson, W.T. Smith, Jr., *J. Am. Chem. Soc.*, **74**, 556 (1952).