CHEMICAL STRUCTURE OF TECHNETIUM-99m-LABELED DIMETHYL-IDA (Tc-HIDA)

B.Zmbova, D.Živanov-Stakić, and A.Muk.
Institute for Radioisotopes, The Boris Kidrič Institute of Nuclear Sciences, Vinča, Belgrade, Yugoslavia.

The stability constants of the Tc-99m labeled radiopharmaceuticals are important thermodynamic quantities which define the ratio of ligand to the metal. The knowledge of the stability constants would aid greatly in predicting the radiopharmaceuticals in vivo stability and in optimization of their syntheses. The stability constant of the Sn-HIDA complex has been measured by the potentiometric method (1). Here we report the determination of the stability constant of the Sn-HIDA-Tc-99m complex by the same method.

The titration was performed on mole ratios of 10:3:1 of HIDA:Sn(II): :TcO_4^-, which provides reduced state of technetium and constant strenght of the electrolyte.

The stability constants of the Sn-HIDA-Tc-99m complex obtained are: logK = 9.0±0.1 and logK' = 6.94±0.06.

The absorption spectra of Sn(II), TcO_4^- and their HIDA complexes have been investigated by the spectrophotometric technique. The absorption maxima, the molar extinction coefficients and the composition of the complexes have been determined by the slope ratio method.

The spectrophotometric studies confirm the formation of a mixed Sn(II)-HIDA-Tc-99m complex and also shows that, depending on the Sn(II) concentration, technetium forms two types of complexes, being in different valence states.

(1) Zmbova B., Karanfilov E. and Jovanović V., Proceedings of 19th International Annual Meeting of the Society of Nuclear Medicine, Bern, 1981.

RECENT DEVELOPMENT IN THE SUBLIMATION GENERATOR OF $^{99}Tc^{m}$

L. Zsinka and L. Szirtes.
Institute of Isotopes of the Hungarian Academy of Sciences
Hungary, H-1525 Budapest, P.O.B. 77.

In the last twenty years the rapid growth in the utilization of $^{99}Tc^{m}$ and its compounds has led to the development of the separating methods of $^{99}Tc^{m}$ from its parent, ^{99}Mo.

The mostcommonly used separation is the elution of $^{99}Tc^{m}$ with saline from an aluminium oxide column to which the ^{99}Mo is complexed (1). The solvent extraction generator and the sublimation generator overcome the limitations inherent in the chromatographic generator and yield a suitable quality of $^{99}Tc^{m}$ from low flux irradiated natural molybdenum trioxide (2,3). In the routine production the separation efficiency of the sublimation method is about 25-30%, applying MoO_3 as target material (4,5). Several papers have described experimental studies of sublimation separation of $^{99}Tc^{m}$ and ^{99}Mo (6,7,8).

The aim of the work was to find a new target material with Mo content instead of MoO_3, in which due to the effect of the temperature near the sublimation temperature of technetium heptoxyd (311 C^{o}) some kind of reversible physical or chemical process contributes to the sublimation thus the separation efficiency could be increased and repeated. In this way the temperature of the separation will be very far from the sublimation temperature of MoO_3(780 C^{o}). The other elements in the compound of the new target material can give radionuclides with very short halflife after reactor irradiation because of the unnecessary radiation dose and the voltatile components. From this point of view the new target material was prepared from titanium and molybdenum compounds (9). The spectrophotometrically determined Mo/Ti ratios were found around 1.0. The Mo/Ti ratio was found to remain constant during the heating, thus the total weight loss of the samples can be attributed to the elimination of water.

The target materials were irradiated for 115 hours in VVRSz-2-M type reactor.

In order to apply the new target material an apparatus was constructed. To realize the original idea 27 various target materials were irradiated and 106 separations were carried out using this apparatus. The average efficiency value of these separations was found to be 63%.

Data concerning the new target material, apparatus and separation technics are collected in the paper.

(1) Richards P., BNL 9601.
(2) Lathorp K.A., Proceedings of a panel on Radiophermaceuticals from Generator-Produced Radionuclides, IAEA, Vienna, 1970.
(3) Perrier C., Segre E., J. Chem. Phys. 5, 712 (1937).
(4) Boyd R.E., Proceedings of a panel on Radiophermaceuticals from Generator-Produced Radionuclides IAEA, Vienna, 1970.
(5) Boyd R.E., Radiopharmacenticals and Labelled Compounds. Proc IAEA-WHO Symp. Copenhagen, 1973, p.3.
(6) Hallaba E., El-Asrag H.A., Isotopenpraxis, 11, 290 (1975).
(7) Rusek V., Vlcek J., Machán V., Rohacek J., Smejkal Z., Kokta L., Vitkova J., Radiochem. Radional. Letters 20 (1), 15-31 (1974).
(8) Levin V.I., Kozyreva-Alexandrova L.S., Sokolova T.N., Zalessky V.G., Radiochem. Radioanal. Letters 39 (2), 141-150 (1979).
(9) Zsinka L., Mándli J., Sugár I., Hungarian patent 169575, (1974).

SYNTHESIS OF CARRIER-FREE CATIONIC TECHNETIUM COMPOUNDS BY A KIT PROCEDURE

R.D. Neirinckx, K.A. Glavan, J.F. Kronauge and M.N. Eakins. Squibb Institute of Medical Research, Georges Road, New Brunswick, New Jersey 08903.

Cationic technetium compounds have recently attracted interest because they can image the myocardium of some animal species (1,2). Synthetic procedures for these cations were described for both carrier - ^{99}Tc(3,4) and no-carrier added ^{99m}Tc(1,2,5). Although the procedures are very valuable as a way of evaluating the compounds, they were complex, time consuming and employed high concentrations of potentially toxic ligands.

To make these Tc-compounds available in the typical radiopharmacy setting the parameters that govern their synthesis were studied and a simple kit synthetic procedure developed. The problem of the volatility of the ligands is solved by using solid metal complexes as sources of the ligands. Iron complexes were chosen because they are stable enough to be lyophilized but decompose when heated in an aqueous solution. To produce trans-dichloro technetium complexes sodium chloride was used as the chloride source. The complexant EGTA was added to the kits because it inhibits iron hydroxide formation and does not complex technetium in the reaction conditions that are proposed for the technetium-cation synthesis. Thus the procedure requires only the addition of a ^{99m}Tc generator eluate to a freeze dried kit and a 45' heating step. The product is pure enough for direct use.

As an example we synthesized $Tc(dmpe)_2Cl_2^+$ by means of $[Fe(dmpe)_2Cl_2]Cl\cdot H_2O$. The effects of NaCl concentration and amount of $[Fe(dmpe)_2Cl_2]Cl\cdot H_2O$ on the purity of $[Tc(dmpe)_2Cl_2]^+$ are summarized in Table I. The intermediate $Tc(dmpe)_2O_2^+$ was identified by FAB (Fig. 1) and IR. Figure 2 represents the formation of the desired cation as a function of time at 125°C. Two mg of EGTA was used in each reaction. Up to 200 mCi of $^{99m}Tc(dmpe)_2Cl_2^+$ is stable in the reconstituted kit solution for at least 24 hours. Toxicity studies have shown that the reconstituted kit product is safe to inject and in-vivo distribution of the $Tc(dmpe)_2Cl_2^+$ has shown its efficacy in several animal models, including monkey.

References:

1. Deutsch E., Glavan K.A., Fergusson D.L., et al: J Nucl Med 22: 897-907, 1981.

2. Deutsch E., Bushong W., Glavan K.A., et al: Science 214, 85 (1981).

3. Fergusson J.F., and Nyholm R.S., Nature, London, 183 1039 (1959).

4. Fergusson J.F., and Nyholm R.S., Chem. Ind. (London), 347-348 (1960).

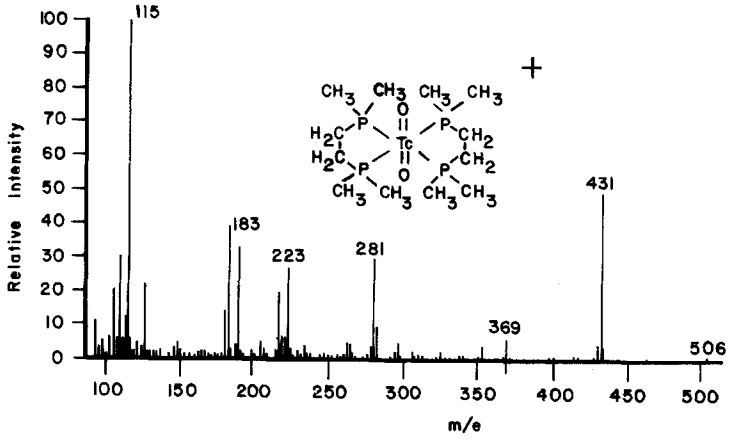
5. Deutsch E., Glavan K.A., Bushong W., et al: Applications of Nuclear and Radiochemistry (Pergamon, New York, in the press).

Table I

Purity of $[^{99m}Tc(dmpe)_2Cl_2]^+$ as a function of NaCl concentration and amount of $Fe(dmpe)_2Cl_2^+$ used.

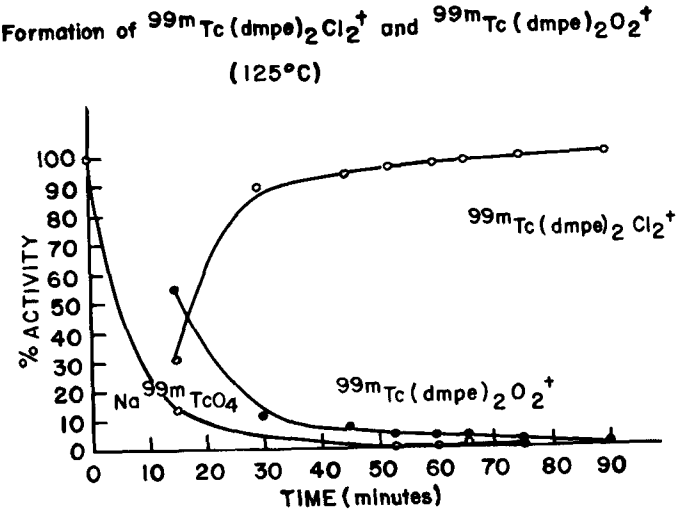
NaCl concentration (M)	$^{99m}Tc(dmpe)_2Cl_2$ purity	$[Fe(dmpe)_2Cl_2]Cl\cdot H_2O$ mg	$^{99m}Tc(dmpe)_2Cl_2^+$ purity (%)
0.15	87	1	47
0.30	89	2	76
0.45	89	4	89
0.62	90	6	88
0.90	91		

Figure 1



Positive Ion Fast Atom Bombardment Mass Spectrum of $[Tc(dmpe)_2O_2]$

Figure 2


Formation of 99mTc(dmpe)2Cl2+ and 99mTc(dmpe)2O2+
(125°C)
% ACTIVITY
100
90
80
70
60
50
40
30
20
10
0
0 10 20 30 40 50 60 70 80 90
TIME (minutes)
99mTc(dmpe)2 Cl2+
99mTc(dmpe)2 O2+
Na 99mTcO4

RADIOPHARMACEUTICALS LABELED WITH BROMINE RADIONUCLIDES

M.J. Welch and K.D. McElvany.
The Edward Mallinckrodt Institute of Radiology, Washington University School of Medicine, St. Louis, Missouri 63110.

Several radionuclides of bromine have been suggested for use in nuclear medicine studies; these nuclides include bromine-74, bromine-75, bromine-76, and bromine-77. The diversity of the available bromine radionuclides, and the greater strength of carbon-bromine bonds as compared with carbon-iodine bonds has led to increased interest in bromine-labeled radiopharmaceuticals over the past several years (1). Bromine-75 has a 95.5 min half-life and decays predominantly by positron emission; as such, this radionuclide can be used in conjunction with positron emission tomographs. Bromine-77 has a 57 hr half-life and decays >99% by electron capture. Although the decay scheme of bromine-77 is complex, phantom studies have demonstrated that reasonably good resolution can be achieved using a scintillation camera fitted with a high energy pinhole collimator (2). Many nuclear reactions have been proposed for the production of the four bromine radionuclides; these methods will be compared and contrasted (1).

Methods for labeling compounds with these bromine radionuclides can be divided into several general categories: (a) recoil labeling using the corresponding krypton to bromine parent-daughter system; (b) chemical labeling; and (c) enzymatic labeling. Recoil techniques have been utilized to label simple compounds in low yield (1), and an adaptation of the method in which the krypton is allowed to decay in the presence of Cl_2 or $KBrO_3$ can produce radiobrominated compounds at higher yields (3,4). Many chemical radiobromination methods, including the use of oxidizing agents such as N-chlorotetrafluorosuccinimide (5), N-chlorosuccinimide (6), organic hypohalites (7), acetic acid/hydrogen peroxide (8), and chloramine-T (9), have been utilized to label compounds with bromine. The majority of the available chemical techniques for no carrier added radiobromination involve conditions that are too harsh for labeling fragile biomolecules such as proteins. Enzyme-catalyzed bromination reactions offer an extremely gentle route for the preparation of radiolabeled molecules that are sensitive to these harsh conditions. The enzymes that have been utilized for this application are chloroperoxidase (10), bromoperoxidase (11), and myeloperoxidase (12).

In the last seven years the application of several brominated radiopharmaceuticals in animal studies or clinical trials has been described; these include 4-[^{77}Br]-2,5-dimethoxyphenylisopropylamine (13,14), [^{77}Br]-p-bromospiroperidol (15), [^{77}Br]-labeled fibrinogen (11), various radiobrominated fatty acids (16,17), and several radiobrominated estrogen analogs (8). The potential of these compounds for nuclear medicine imaging will be discussed.

(1) Stöcklin G., Int. J. Appl. Radiat. Isotop., 28, 131 (1977).
(2) Eckelman W.C. and Reba R.C., "Radiopharmaceuticals: Structure -- Activity Relationships", Grune and Stratton, New York, 1981, p. 449.
(3) Lambrecht R.M., Mantescu C., Redvanly C.S., and Wolf A.P., J. Nucl. Med., 13, 266 (1972).
(4) El-Garhy M. and Stöcklin G., Radiochem. Radioanal. Letters, 18, 28 (1974).
(5) Coenen H.H., Machulla H.-J., and Stöcklin G., J. Lab. Comp. Radiopharm., 16, 891 (1979).
(6) Wilbur D.S., Bentley G.E., and O'Brien H.A. Jr., J. Lab. Comp. Radiopharm., 18, 1693 (1981).
(7) Coenen H.H., El-Wetery A.S., and Stöcklin G., J. Lab. Comp. Radiopharm., 18, 114 (1981).
(8) Katzenellenbogen J.A., Senderoff S.G., McElvany K.D., O'Brien H.A., Jr., and Welch M.J., J. Nucl. Med., 22, 42 (1981).
(9) Petzold G. and Coenen H.H., J. Lab. Comp. Radiopharm., 18, 1319 (1981).

(10) Knight L., Krohn K.A., Welch M.J., Spomer B., and Hager L.P., "Radiopharmaceuticals", Society of Nuclear Medicine, New York, 1975, p. 149.
(11) McElvany K.D. and Welch M.J., J. Nucl. Med., 21, 953 (1981).
(12) McElvany K.D., Barnes J.W., and Welch M.J., Int. J. Appl. Radiat. Isotop., 31, 679 (1981).
(13) Sargent T., Kalbhen D.A., Shulgin A.T., Stauffer H., and Kusubov N., J. Nucl. Med., 16, 243 (1975).
(14) Coenen H.H., J. Lab. Comp. Radiopharm., 18, 733 (1982).
(15) Kulmala H.K., Huang C.C., Dinersten R.J., and Friedman A.M., Life Sciences, 28, 1911 (1980).
(16) Machulla H.-J., Stöcklin G., Kupfernagel Ch., Freunleib Ch., Hock A., Vyska K., and Feinendegen L.E., J. Nucl. Med., 19, 298 (1978).
(17) Coenen H.H., Harmard M.-F., Kloster G., Stöcklin G., J. Nucl. Med., 22, 891 (1981).

RECENT STUDIES OF RADIOBROMINATION AND -IODINATION (NCA) WITH CHLORAMINE-T AND DICHLORAMINE-T IN AQUEOUS AND ORGANIC SOLVENTS

H.H. Coenen, G. Petzold, and G. Stöcklin.
Institut für Chemie 1 (Nuklearchemie), Kernforschungsanlage Jülich GmbH, D-5170 Jülich, FRG.

Chloramine-T (CAT) has been widely used for the radioiodination of proteins and other compounds. In spite of the popularity of this reagent, there is little information available on the reaction mechanism involved. Hypochlorite (HOCl) is most often assumed to be the oxidizing agent and HOI, I_2, or simply "I^+" the reacting species. Recently CAT has also been used for radiobromination (1,2). Optimization studies for radiobromination exhibit striking pH-dependent differences between bromination and iodination (2).

Our selectivity studies on the halogenation of aniline and phenol now confirm these differences. The relative isomer distribution exhibits a complicated pH dependence, indicating a sensitive and complex reaction mechanism. Further detailed studies show that none of the above-mentioned species but rather H_2OX^+, or more likely ClX (X = I, Br), play a major role in halogenation at pH 1. At pH 7, N-halogenated species of toluene-sulfonamide such as RSO_2NClX and RSO_2NHX are considered. These were also discussed as reacting intermediates in reactions of bromamine-B at higher pH (3). The differences between bromination and iodination reactions must then be attributed to differences in the stability and reactivity of the intermediates.

Studies on chloramine-T (2) encouraged us to test dichloramine-T (DCT) as an oxidizing agent for radiohalogenation because of its solubility in organic solvents. Dichloramine-T (N,N-dichloro-p-toluene-sulfonamide) was found to be a very effective agent for fast radiobromination and radioiodination in many organic solvents. At room temperature, the reaction proceeds in less than one minute when using 10^{-3} M DCT. Tables 1 and 2 show the radiochemical yields and the relative isomer distribution obtained in the case of phenol and aniline. DCT can be used in protic (CH_3COOH), aprotic, polar (CH_2Cl_2) and nonpolar (CCl_4) solvents. For activated benzenes, high radiochemical yields ($\sim$ 75 %) are obtained, similar to those in aqueous solution using CAT. In addition, a solvent effect on the isomer distribution is found for iodination of phenol but is rather small for bromination of phenol and halogenation of aniline. A comparative study of radioiodination in polar solvents such as TFAA using various N-halosuccinimides and CAT (4) indicates that DCT and CAT are superior to the other reagents as far as the speed of the reaction and concentration of the oxidizing agent are concerned. The saturation yields are very similar for all reagents, with the exception of N-bromosuccinimide.

We also applied CAT/DCT to the radiohalogenation of unsaturated compounds (electrophilic addition) and compared the product distribution with that obtained using N-chlorosuccinimide (5). By choosing the appropriate solvent, the 1-bromo-2-methoxy- (in methanol), the 1-bromo-2-chloro- (in CH_2Cl_2), or 1-bromo-2-hydroxy- (in water or aqueous mixtures) addition products were formed in olefines such as cyclohexene (70 %) and tri-O-acetyl-D-glucal (75 %).

Table 1 Radiohalogenation (n.c.a.) of Phenol with DCT in Various Solvents at Room Temperature.

Solvent	Bromination		Iodination	
	Radiochem. Yield %	% Rel. Isomer Distr. (o/p)	Radiochem. Yield %	% Rel. Isomer Distr. (o/p)
AcOH	72	24/76	37	73/27
CH_2Cl_2	82	46/54	73	45/55
CCl_4	64	36/64	80	42/58

Table 2 Radiohalogenation (n.c.a.) of Aniline with DCT in Various Solvents at Room Temperature.

Solvent	Bromination		Iodination	
	Radiochem. Yield %	% Rel. Isomer Distr. (o/p)	Radiochem. Yield %	% Rel. Isomer Distr. (o/p)
AcOH	76	29/71	77	16/84
CH_2Cl_2	76	18/82	70	12/88
CCl_4	81	15/85	60	31/69

(1) Hadi U.A.M., Malcolme-Lawes I.J. and Oldham G., Int. J. appl. Radiat. Isotopes, 30, 709 (1979).
(2) Petzold G. and Coenen H.H., J. Lab. Comp. Radiopharm., 18, 1319 (1981).
(3) Hardy E.E. and Johnston J.P., J. Chem. Soc. Perkin II, p. 742 (1973).
(4) He Youfeng, Coenen H.H., Petzold G. and Stöcklin G., J. Lab. Comp. Radiopharm., in press.
(5) Wilbur D.S. and Anderson K.W., J. Org. Chem., 47, 358 (1982).

NEW METHODS OF RADIOHALOGENATION

M.R. Kilbourn, K.D. McElvany, and M.J. Welch.
The Edward Mallinckrodt Institute of Radiology, Washington University School of Medicine, St. Louis, Missouri 63110 USA.

The radioisotopes of iodine have seen widespread applications in nuclear medicine. Recently the radioisotopes of bromine have been suggested for nuclear medicine studies, as either replacements for radioiodine in presently used radiopharmaceuticals (1), or in the development of entirely new labeled compounds (2). As the heavy halogens are of such continued interest in nuclear medicine, we have developed new methods of radiohalogenation. We report here the development of two new methods for radiohalogenation via aliphatic nucleophilic substitution. Although to date these techniques have been utilized to label model compounds, the application of these reactions to label radiopharmaceuticals for myocardial and receptor studies is in progress.

Sodium Halide/Chlorotrimethylsilane. The treatment of an alcohol with chlorotrimethylsilane and either sodium bromide or sodium chloride is an excellent method for the synthesis of the corresponding alkyl halides. We have found that the use of sodium [^{77}Br]bromide or sodium[^{131}I]iodide allows for the simple synthesis of no-carrier-added radiolabeled alkyl halides.

$$RCH_2OH + (CH_3)_3SiCl + NaX \longrightarrow RCH_2X$$

$$X = {}^{77}Br,\ {}^{131}I$$

This new radiohalogenation reaction is very simple, consisting of adding alcohol, chlorotrimethylsilane, and acetonitrile (used as solvent) to the sodium halide, and refluxing the solution for 1-4 hours. Yields of bromine-77 or iodine-131 alkyl halides range from 26-86% (Table 1), and the reaction can be used on primary, secondary and benzylic alcohols. In all cases the radiolabeled alkyl halides are obtained as virtually the only labeled organic products. As examples of possible applications of this new radiolabeling method, we have converted 12-hydroxydodecanoic acid to [^{77}Br]12-bromododecanoic acid (12% yield, 87% radiochemical purity) and [^{131}I]12-iododododecanoic acid (31% yield, 93% radiochemical purity). This radiolabeling technique should be easily applied to the labeling of fatty acids for use as myocardial imaging agents.

Sodium Halide/Tetrahalomethane/Triphenylphosphine. The reaction of an alcohol with carbon tetrachloride and triphenylphophine yields the corresponding alkyl chloride. Similarly, use of carbon tetrabromide or carbon tetraiodide yields the alkyl bromide or alkyl iodine, respectively.

$$R\text{-}CH_2OH + (C_6H_5)_3P + CX_4 \longrightarrow R\text{-}CH_2X + (C_6H_5)_3PO + CHX_3$$

We have found that this reaction can be applied to radiolabeling of alkyl halides if [^{77}Br]bromide or [^{131}I]iodide ions are supplied as added external nucleophiles: the incorporation of external nucleophiles during the normal chlorination reaction has been recently reported (3). Using sodium [^{77}Br]bromide and carbon tetrachloride, we have prepared no-carrier-added [^{77}Br]benzyl bromide in 36-53% yields, with very high radiochemical purities (>97%). Using carbon tetrabromide and sodium [^{77}Br]bromide, the radiolabeled alkyl bromide is obtained as the carrier-added product. This radiobromination reaction can also be used to prepare labeled fatty acids; treatment of 16-hydroxyhexadecanoic acid ethyl ester with sodium [^{77}Br]bromide, carbon tetrachloride, and triphenylphosphine gave the desired [^{77}Br]16-bromohexadecanoic acid ethyl ester in 31% yield and 98% radiochemical purity. Radioiodination is also easily achieved, and we have prepared [^{131}I]1-iodohexane in 60% yield and 98% radiochemical purity (no-carrier-added synthesis using CCl_4). This radiohalogenation reaction is extremely fast (reaction times of 5-30 minutes) and proceeds under unusually mild conditions (0-5°C, no acid, base, or oxidizing agent). The combination of speed

and mildness should prove useful in the labeling of complex molecules with various radionuclides.

REFERENCES

(1) Stöcklin G., Int. J. Appl. Radiat. Isotop. 28, 131 (1977).
(2) Katzenellenbogen J.A., Senderoff, S.G., McElvany K.D., O'Brien H.A., and Welch M.J., J. Nucl. Med. 22, 42 (1981).
(3) Slagle J.D., Huang T., and Franzus B., J. Org. Chem. 46, 3526 (1981).

TABLE 1. Syntheses of bromine-77 or iodine-131 labeled alkyl halides using sodium halide/chlorotrimethylsilane/alcohol reaction

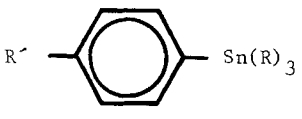
Product	Radiochemical Yield	Radiochemical Purity
$[^{77}Br]CH_3CH_2CH_2CH_2Br$	61	100
$[^{77}Br](CH_3)_2CHBr$	26	100
$[^{77}Br]CH_3(CH_2)_6CH_2Br$	59	100
$[^{77}Br]CH_3CH_2CHBrCH_3$	46	100
$[^{77}Br]C_6H_5CH_2Br$	86	100
$[^{77}Br]BrCH_2(CH_2)_{10}COOH$	12	87
$[^{131}I]CH_3CH_2CH_2CH_2I$	61	98
$[^{131}I]CH_3(CH_2)_4CH_2I$	48	97
$[^{131}I](CH_3)_2CHI$	51	98
$[^{131}I]ICH_2(CH_2)_{10}COOH$	31	93

SITE SPECIFIC RADIOBROMINATION OF AROMATIC COMPOUNDS

M.J. Adam, T.J. Ruth, B.D. Pate, and L.D. Hall.
TRIUMF and Department of Pharmaceutical Sciences,
University of British Columbia, Vancouver, Canada, V6T 2A3.

Recently a few new methods (1,2,3) for radiobrominating aromatic compounds have been described, which rely on the production of an electrophilic bromine species by oxidation of bromide ions. Unfortunately, these methods lack the required site specificity when applied directly to aromatics; however, this problem can be overcome by using aryl-tin reagents as substrates. There is ample precedent for cleavage of aryl-tin bonds by electrophiles such as halogens (4), under conditions such that alkyl-tin bonds are cleaved much more slowly than their aryl-tin counterparts (5). We recently reported the use of aryl-tin derivatives as precursors of radiofluorinated aromatic compounds (6), and have now extended that approach to the rapid synthesis of brominated aromatic compounds.

Reaction of tributylphenyltin (I), tributyl-p-anisoletin (II) or tetraphenyltin (III) with $NH_4{}^{82}Br$ were performed in 1N HCL in



(I) R = nBu, R´=H

(II) R = nBu, R´=OMe

(III) R = Ph , R´=H

EtOH/H_2O (2:1) with 10^{-3}M chloramine-T at 0°C for 5 min. and gave radiochemical yields of 96, 95 and 38% respectively.

Given both the speed and the high selectivity of cleavage of aryl-tin bonds in the presence of alkyl-tin groups, this route should be generally applicable for radiobrominating a wide variety of aromatic compounds with either ^{75}Br or ^{77}Br.

(1) D.J. Malcome-Lawes and S. Massey, J.C.S. Chem.Comm., 221 (1980).
(2) G. Petzold and H.H. Coenen., J. Lab. Comp. Radiopharm., 18,1319 (1981).
(3) H.H. Coenen, H.J. Machulla and G. Stöcklin., J. Lab. Comp. Radiopharm., 16, 891 (1979).
(4) R.K. Ingham, S.D. Rosenberg, and H. Gilman., Chem. Rev., 60, 459 (1960).
(5) A. Folaranmi, R.A.N. McLean, and N. Wadibia., J. Organomet. Chem., 73, 59 (1974).
(6) M.J. Adam, B.D. Pate, T.J. Ruth, J.M. Berry, and L.D. Hall., J.C.S. Chem. Comm., 733 (1981).

RADIOIODODESTANNYLATION: A METHOD FOR SPECIFICALLY LABELED RADIOPHARMACEUTICALS

R.N. Hanson and D.E. Seitz.
Departments of Medicinal Chemistry and Chemistry, Northeastern University, and Joint Program in Nuclear Medicine, Department of Radiology, Harvard Medical School, Boston, MA 02115.

The objective of this study has been the development of organotin chemistry and radioiododestannylation as a useful method for the synthesis of specifically substituted radiochemicals. The potential advantages of this approach include the ability to prepare a variety of stable organostannane intermediates of known structure, to convert them rapidly to organoiodides of the same chemical configuration, and then to easily separate them from the nonlabeled starting materials. The results described represent the initial investigations into the first two aspects.

One important feature of this methodology involves the facile synthesis of the bis(trialkylstannyl) intermediates I-III. The E-1,2-bis(tributylstannyl)ethylene I can be prepared in two steps in an overall yield of 90% starting from acetylene (1). The 2,5-bis(trimethylstannyl)thiophene II and -furan III are prepared in one step reactions with alkyllithium (2.1-2.5 equivalents) (2) and trimethyltin chloride (2.2 equivalents). The isolated yields are 82% and 68% respectively (3). The intermediates are solids which can be stored in the cold (0°C) for extended periods without decomposition.

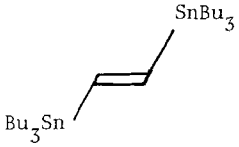
Another important feature is the ability of these intermediates to selectively undergo monotransmetalation. The addition of one equivalent of an alkyllithium gives the reactive monolithio derivative. Proof of the formation of this intermediate can be obtained by the addition of methyliodide. With methyllithium 75% of II or III is consumed to give isolated yields of 51% and 57% of the monomethyl monotrimethylstannylthiophene or -furan. The monolithio derivatives of I-III also add to ketones, such as hexanone and estrone, to give the allylic or benzylic alcohols in 30-80% yields.

Conversion of the functionalized trialkylstannyl ethylene or heteroarene to the corresponding iodide proceeds virtually instantaneously and quantitatively. The isolated yields of the iodinated products are in the 90-98% range. The position of iodination and its stereochemical orientation are identical to those of the trialkylstannyl intermediate. Whether iodine-125 labeled molecular iodine or iodine monochloride is used, comparable results are obtained. The use of no-carrier-added radioiodine, oxidizing agents, and more polar solvents also gives the identical radioiodinated product, but the yields and product mixtures are more variable. Current research efforts are focused on determining the optimal solvent and oxidant for the reaction.

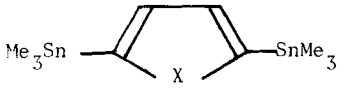
(1) Botarro, J.D., Hanson, R.N. and Seitz, D.E., J. Org. Chem., 46, 5221 (1981).

(2) Chadwick, D.J. and Willbe, C., J. Chem. Soc., Perkin Trans. 1, 887 (1977).

(3) Seitz, D.E., Lee, S.-H., Hanson, R.N. and Botarro, J.C., Tetrahedron Lett., (submitted).



I



II X = S

III X = O

REGIOSPECIFIC INCORPORATION OF RADIOIODINE INTO AROMATIC RINGS VIA ORGANOSILANES

D. S. Wilbur, K. W. Anderson, W. E. Stone, Z. V. Svitra, R. S. Rogers, and H. A. O'Brien.
Medical Radioisotope Research (CNC-3), Los Alamos National Laboratory, Los Alamos, New Mexico 87545.

Organosilanes are organometallic compounds, which unlike many other organometallic compounds, are quite stable and need no special care in handling. Organosilane derivatives can be readily synthesized via a number of methods (1) and are particularly noteworthy for their synthetic utility in electrophilic substitution reactions (2,3). The propensity of the aryltrimethylsilyl derivatives toward electrophilic cleavage, particularly by halogens, led to an investigation of the utility of such derivatives to introduce radioiodine into aromatic ring (4,5). The investigation utilized the three trimethylsilyl regioisomers of phenol and toluene as model compounds for highly activated and less activated aromatic rings.

Unlike the radiobrominations using aryltrimethylsilyl derivations (6), radioiodinations were found to be very slow in MeOH or EtOH using either N-chlorosuccinimide (NCS) or tert-butylhypochlorite to oxidize iodide to ICl. However, changing the solvent to acetic acid increased the rate of reaction such that carrier-added radioiodinations (Iodine-131) could be accomplished in reasonable times with very good radiochemical yields. No-carrier-added radioiodinations (I-131) were found to be very slow under the same reaction conditions, but elevation of the reaction temperature to 60°C again gave reasonable radiochemical yields in short reaction times (Table 1).

The ortho and para activating effect in the trimethylsilyl phenols causes the addition of electrophilic halogens to these positions to be more rapid than the cleavage of the meta-trimethylsilyl group (6). Thus, the meta substituted radioiodine labeled phenolic rings are not directly accessible via this method. However, derivatization of the phenol by the electron withdrawing acetoxy group decreased the activation to ortho and para substitution to the extent that regiospecific cleavage of the meta-TMS derivative by iodine or bromine could be accomplished in high yield.

The utility of the trimethylsilyl derivatives to introduce radioiodine (or radiobromine) into specific compounds of interest is being investigated. Currently, the trimethylsilyl derivations of hippuric acid, haloperidol, and ω-phenyl fatty acids are being synthesized.

(1) Häbich D. and Effenberger F., "Preparation of Aryl- and Heteroaryltrimethylsilanes", Synthesis 841-876 (1979).

(2) Chan T. H. and Fleming I., "Electrophilic Substitution of Organosilicon Compounds - Applications to Organic Synthesis", Synthesis 761-786 (1979).

(3) Eaborn C., "Cleavage of Aryl-Silicon and Related Bonds by Electrophiles", J. Organometallic Chem. 100, 43-57 (1975).

(4) Félix G., Dunogués J., Pisciotti F., and Calas R., "Regiospecific Synthesis of Mono- and Polyiodo Derivatives of Benzene", Angew. Chem. Int. Ed. Engl. 16, 488-489 (1977).

(5) Félix G., Dunogués J., and Calas R., "Regiospecific Synthesis of Disubstituted Benzene Derivatives", Angew. Chem. Int. Ed. Engl. 18, 402-404 (1979).

(6) Wilbur D. S., Anderson K. W., Stone W. E., and O'Brien H. A., "Aromatic Radiobrominations Using Aryltrimethylsilyl Derivatives", Abstract. Society of Nuclear Medicine 29th Annual Meeting. J. Nucl. Med. in press.

TABLE 1: Radioiodinations[a]

	[b]Rxn Time	NCS/^{131}I(ca)/HOAc	NCS/^{131}I(nca)HOAc/60°C
para-TMST[c]	5 min	81%	56%[d]
	1 hr	97%	72%
meta-TMST	5 min	61%	75%[d]
	1 hr	88%	93%
ortho-TMST	5 min	92%	65%[d]
	1 hr	96%	81%

[a]All radioiodinations were conducted at room temperature unless otherwise stated; all represent regiospecific cleavages; percentages recorded are of total activity seen by HPLC radiochromatography.

[b]Measured from time of addition of oxidizing agent.

[c]TMST=trimethylsilyltoluene.

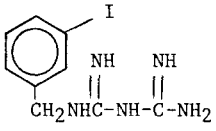
[d]20 minutes from addition of reagents; ~15 minutes at 60°C.

MW^2 RADIOIODIDE EXCHANGE METHOD: IMPROVEMENTS AND APPLICATION TO NO-CARRIER-ADDED SYNTHESES

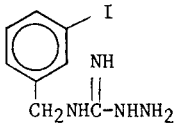
D.M. Wieland, T.J. Mangner, M. Inbasekaran, and C.A. Otto.
University of Michigan Medical Center, Ann Arbor, Michigan 48109.

Recently our laboratory reported a convenient solid-phase technique for radioiodide exchange labeling of aryl iodides not activated by electron-donating substituents (1,2). We report here modifications of this reaction which increase its versatility.

The original technique (MW^2 method) involves the facilitation of radioiodide exchange by mildly acidic, oxidizing conditions provided by the in situ thermal decomposition of $(NH_4)_2SO_4$ in the presence of oxygen. Reactions are generally run with 0.25-2.0 mg of aryl iodide, 1-10 mCi of NaI-125, and 2-4 mg of $(NH_4)_2SO_4$ at 140-160°. The water initially used to dissolve the reactants is rapidly driven off by subsequent heating. Although a wide variety of aryl iodides have been radiolabeled by this technique (2), we have observed hydrolysis, acid catalyzed reactions, and thermal decomposition with certain compounds. For example meta-iodobenzylbiguanide 1 undergoes considerable hydrolysis to meta-iodobenzylamine under the usual reaction conditions. This can be avoided by initially dissolving the reactants in 95% methanol and then evaporating the solvent



1

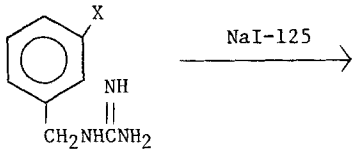


2

with a stream of argon at ambient temperature. The reaction temperature is then raised to 140° to give a 45% radiochemical yield (isolated) of pure product within 90 minutes. This low temperature elimination of water via methanol/argon is also the key to successful exchange with aryl iodides thermally unstable at 140-160°. For example N^3-amino-meta-iodobenzylguanidine (2) is exchange labeled at 108° in 53% radiochemical yield (isolated).

The ease with which isotopic exchange occurs prompted us to apply this technique to aryl bromides and chlorides. Preliminary results shown in Table 1 suggest that this technique could be used for no-carrier-added syntheses. The radiochemical purity of the products in Table I has been confirmed by radio-HPLC using reverse phase columns. In most cases a 2-3 minute separation between the respective aryl bromides and iodides can be obtained by gradient elution. Radiochemical yields are moderate due to loss of some radioactivity as volatile radioiodine. Solutions to this problem are under investigation. Also by this technique, I-125-para-iodophenylacetic acid has been synthesized from the respective bromide in 34% isolated yield. The reaction is being applied to longer chain ω-phenylfatty acids.

TABLE 1



	Radiochemical Yield (%)		
X	o	m	p
Br	-	55	50
Cl	10	<1	-

(1) Mangner T.J., Wu J.L., Wieland D.M. and Beierwaltes W.H., J. Nucl. Med. 22, P12 (1981).
(2) Mangner T.J., Wu J.L. and Wieland D.M., J. Org. Chem., In press (1982).

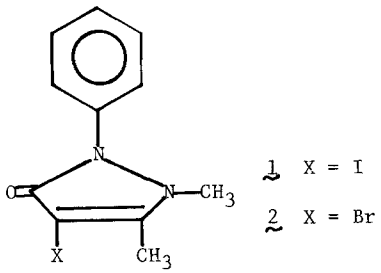
SYNTHESIS OF RADIOBROMINATED 4-BROMOANTIPYRINE FOR THE MEASUREMENT OF CEREBRAL BLOOD FLOW

C.-Y. Shiue and A.P. Wolf.
Department of Chemistry, Brookhaven National Laboratory, Upton, NY 11973.

Radioisotopically labeled antipyrines, labeled with ^{14}C, ^{131}I, ^{125}I or ^{123}I have been used to study the symmetry of brain perfusion using autoradiography (1), gamma camera (2), and single photon tomography (3). However, 4-iodoantipyrine (1) is unstable in vivo and the half-life for radioactive iodine is relatively long ($t_{1/2}$ = 13.1 hrs for ^{123}I; $t_{1/2}$ = 8 days for ^{131}I). In order to overcome these problems, we have recently synthesized ^{18}F-4-fluoroantipyrine (a positron emitter, $t_{1/2}$ = 110 min) and used it successfully as a regional cerebral blood flow indicator (4,5). In extending the availability of radiopharmaceuticals such as ^{18}F-4-fluoroantipyrine, we have synthesized ^{82}Br-4-bromoantipyrine (2).

^{82}Br-4-Bromoantipyrine was synthesized by three methods: the melt method, isotopic exchange in acidic medium (6) and the silica gel catalyzed method (7). Among these methods, the melt method was the simplest one and gave the highest radiochemical yield (~ 90%). Isotopic exchange in acidic medium gave ^{82}Br-4-bromoantipyrine in ~ 15% radiochemical yield while the silica gel catalyzed method gave ^{82}Br-4-bromoantipyrine in poor yield.

In a typical experiment, a solution of 4-bromoantipyrine (14.3 mg, 0.054 mmol) and ^{82}Br-NH_4Br (33.7 mCi) in methanol (1 ml) was placed in a V-shaped flask and was heated briefly under a gentle stream of nitrogen to dryness. The residue was allowed to melt (140°C) and remain as a melt for 5 min and then cooled to room temperature. The mixture was dissolved in water (1 ml) and extracted with chloroform (3 x 1 ml). The organic layer was separated, passed through an anhydrous sodium sulfate column (4 x 1 mm) and evaporated to dryness to give 29.7 mCi (88.1%) of ^{82}Br-4-bromoantipyrine. The synthesis time was ~ 20 min. Radiochemical purity of ^{82}Br-4-bromoantipyrine was determined to be > 99% by thin layer chromatography on silica gel (toluene-ethyl acetate, 1:1), Rf = 0.61. The partition coefficient of ^{82}Br-4-bromoantipyrine in octanol/phosphate buffer was 9.78 which was between ^{131}I-4-iodoantipyrine (11.34) and ^{18}F-4-fluoroantipyrine (5.18).



1 X = I

2 X = Br

Fig. 1 Structures of 4-haloantipyrine

This method can readily be adapted for the synthesis of ^{75}Br-4-bromoantipyrine (a positron emitter, $t_{1/2}$ = 98 min) and used for the measurement of cerebral blood flow by positron emission tomography.

This research was carried out at Brookhaven National Laboratory under contract with the U. S. Department of Energy and the Office of Health and Environmental Research.

(1) Eckman, W.W., Phair, R.D., Fenstermacher, J.D. et al., Am. J. Physiol. 229, 215 (1975).
(2) Uszler, J.M., Bennett, L.R., Mena, I., et al., Radiology 115, 197 (1975).
(3) Kuhl, D.E., Edwards, R.Q., Ricci, A.R., et al., Radiology 121, 405 (1976).
(4) Shiue, C.-Y. and Wolf, A.P., J. Label. Cmpds. Radiopharm. 18, 1059 (1981).
(5) Duncan, C.C., Shiue, C.-Y., Wolf, A.P., et al., J. Cerebral Blood Flow and Metab. I (Suppl. 1), S78-S79 (1981).
(6) Robinson, G.D. and Lee, A.W., J. Nucl. Med. 17, 1093 (1976).
(7) Boothe, T.E., Campbell, J.A., Djermouni, B., et al., Int. J. Appl. Radiat. Isot. 32, 153 (1981).

THE CHEMISTRY OF TECHNETIUM

C.J.L. Lock.
Institute for Materials Research, McMaster University, Hamilton, Ontario
Canada L8S 4M1.

The chemistry of technetium in the reduced oxidation states relevant to radiopharmaceuticals will be discussed and compared to the better known chemistry of rhenium.

Specific areas to be discussed will be complexes of Tc(V) containing the TcO^{3+}, TcO_2^{+} and $TcO(OR)^{2+}$ groups and Tc(IV) and Tc(III) octahedral complexes stabilized with S, N, O and P containing ligands. Attempts to characterize technetium compounds by use of ^{99}Tc NMR will also be discussed with examples from Tc(VII)d^0, Tc(V)d^2, and Tc(I)d^6 complexes.

A NEW CLASS OF WATER SOLUBLE LOW VALENT TECHNETIUM UNIPOSITIVE CATIONS: HEXAKISISONITRILE TECHNETIUM(I) SALTS

A.G. Jones, A. Davison, M.J. Abrams, J.W. Brodack, C.E. Costello, A.I. Kassis, R.F. Uren, M. Simon, L. Stemp, and B.L. Holman.
Department of Radiology, Harvard Medical School, Boston, MA 02115.

A new class of cationic technetium(I) complexes containing various isonitrile ligands has been identified. These materials can be readily prepared in aqueous media at both carrier and no carrier added concentrations by reduction of pertechnetate ion in the presence of the ligand. A number of this class have now been characterized by elemental analysis, UV-VIS, IR, and NMR spectroscopy and field desorption mass spectrometry. Preliminary biological studies with several of these cations (e.g. methyl, isopropyl, n-butyl and t-butyl isonitriles) show that they each behave differently in vivo. Of these, the hexakis-(t-butylisonitrile)technetium(I) cation has proven to possess some remarkable properties.

The table below shows data obtained using a standardized in vitro method for measuring uptake of radioactivity in Chinese hamster V79 lung fibroblasts. Following a ten minute incubation the uptake of the complex was measured as 29 pCi/cell, as compared to 1.2 pCi/cell for ^{111}In-oxine and 0.01 for TcO_4^-. Retention studies indicated that 50% of the radiolabel remained with the cells after 16 hours in culture. At the end of this time, the cells appeared viable by inspection and seemed to have divided relatively normally. For example, the cell population in one experiment increased from an initial 235,000 cells/ml to 325,000 cells/ml.

When injected intravenously into rats and dogs, the t-butyl complex gave good images of the heart after initial clearance of activity from the lungs. In one dog with an experimental myocardial infarction, the remaining viable tissue was well delineated. Comparison of the distribution of the ^{99m}Tc complex and ^{201}Tl in the isolated heart from this animal by analyzing tomographic slices obtained on a SPECT detector showed the two to be essentially similar.

More interestingly, in two ostensibly normal animals, as well as visualizing the heart, distinct sites of localization were observed in the lungs. On the supposition that these could be blood clots, the complex was then tested in a canine model of pulmonary embolism. Autologous blood was allowed to clot in vitro, samples introduced into the lungs via a catheter placed in the inferior vena cava, and their final positions determined. Most of these were visualized after injection of the complex. In one animal, clots trapped in the abdominal field by a filter placed in the inferior vena cava were also visualized. Further experiments are now being conducted to determine the effectiveness of this complex in labeling cells for imaging the heart, and for the detection of vascular emboli.

Table. Comparative Whole Cell Uptake Levels in Chinese Hamster V79 Lung Fibroblasts.

Agent	External Conc. (uCi/ml)	Internal Conc. (pCi/cell)
^{111}In-Oxine	15	1.2
^{51}Cr-Chromate	15	9.0
$^{99m}TcO_4^-$	100	0.01
$^{99m}Tc(CN\text{-}t\text{-}C_4H_9)_6^+$	13	29.1

Characterization of [^{99}Tc]-hexakis-(t-butylisonitrile)technetium(I) phosphate salt.

Elemental analysis: $C_{30}H_{54}N_6F_6PTc$

	C	H	N
Calcd.:	48.50	7.34	11.32
Found:	48.61	7.24	11.32

Infrared: C-N 2080 $cm.^{-1}$
PF_6 840 $cm.^{-1}$

NMR (in CD_2Cl_2): complex singlet 1.65 ppm with respect to TMS
ligand triplet 1.2 ppm with respect to TMS

FDMA (positive ion mode): m/z = 597 C^+ (strong)
m/z = 742 M^+ (= CA^+) (weak)

(1) Kassis A.I. and Adelstein S.J., J. Nucl. Med., 21, 88 (1980).

THE PREPARATION OF TECHNETIUM(III) COMPOUNDS IN AQUEOUS MEDIA

M.J. Abrams, A. Davison, J.W. Brodack, A.G. Jones, R. Faggiani, and C.J. Lock.
Department of Chemistry, Massachusetts Institute of Technology, Cambridge, MA 02139.

The orange-red complex that is formed when acid solutions of TcO_4^- are heated with thiourea has formed the basis of a colorimetric test for the element for over 20 years (1). Two salts have now been isolated from such solutions and fully characterized. The treatment of NH_4TcO_4 with thiourea in ethanolic HCl gives the red hexakis-(S-thiourea)technetium(III) chloride trihydrate in 90% yield. A similar reaction in 48-50% tetrafluoroboric acid gives the tetrafluoroborate salt $[Tc(S\text{-thiourea})_6][BF_4]_3$ in 82% yield. The characterization of these two salts is given below. A single crystal x-ray determination showed the chloride salt to contain an octahedral arrangement of six S-bonded thiourea ligands around the technetium atom.

The complexes can function as synthetic precursors for other low valent technetium complexes. For example, reaction with t-butylisonitrile in methanol gives a 60% yield of hexakis-[t-butylisonitrile]technetium(III) hexafluorophosphate. With 1,2-bis-diphenylphosphinoethane (diphos) the known complex trans-[dichlorobis(diphos)]technetium(III) chloride (2) is obtained.

A number of technetium(III) complexes with isoleptic ligands are well characterized, for example $K_4[Tc(CN)_7]$ (3), $[n\text{-}Bu_4N]_3[Tc(NCS)_6]$ (4), and tris-(2,4-pentanedionato)technetium(III) (5). The last complex was first identified by Mazzi et al. using a nonaqueous synthesis starting with $[TcCl_6]^{2-}$. We have now isolated this complex at both carrier and no carrier added concentrations after direct reduction of pertechnetate with $Na_2S_2O_4$ in aqueous ethanolic base.

These results, and those obtained by Deutsch in producing mixed ligand phosphine and arsine complexes, clearly demonstrate that stable technetium(III) complexes can be readily obtained in aqueous solution with a variety of ligand types and reducing agents.

Characterization of hexakis-(S-thiourea)technetium(III) chloride trihydrate.

Elemental analysis: $C_6H_{24}N_{12}S_6Cl_3Tc$

	C	H	N	S
Calcd.:	10.88	3.66	25.39	29.05
Found:	10.84	3.59	25.30	29.04

UV/VIS (in MeOH): 493 nm $\varepsilon = 5.8 \times 10^3$ l. mol.$^{-1}$ cm.$^{-1}$
428 nm $\varepsilon = 7.4 \times 10^3$ l. mol.$^{-1}$ cm.$^{-1}$

Conductivity: 10^{-3} M (in MeOH) 260 ohm.$^{-1}$ cm.^{2} mol.$^{-1}$

Magnetic susceptibility (in CH_3CN): $\mu_{eff}(308^oK) = 2.7$ BM

Characterization of hexakis-(S-thiourea)technetium(III) tetrafluoroborate.

Elemental analysis: $C_6H_{24}B_3N_{12}F_{12}S_6Tc$

	C	H	N	S
Calcd.:	8.83	2.97	20.60	23.57
Found:	8.63	3.19	20.19	23.19

UV/VIS (in MeOH): 493 nm $\varepsilon = 6.8 \times 10^3$ l. mol.$^{-1}$ cm.$^{-1}$
428 nm $\varepsilon = 8.2 \times 10^3$ l. mol.$^{-1}$ cm.$^{-1}$

Conductivity: 10^{-3}M (in MeOH) 230 ohm.$^{-1}$ cm.^{2} mol.$^{-1}$

Magnetic susceptibility (in CH_3CN): $\mu_{eff}(308^{\circ}K) = 2.7$ BM

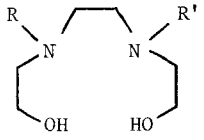
(1) Jasim F., Magee R.J. and Wilson C.L., Talanta, 2, 93 (1959).
(2) Hurst R.W., Heineman W.R. and Deutsch E., Inorg. Chem., 20, 3298 (1981).
(3) Trop H.S., Jones A.G. and Davison A., Inorg. Chem., 19, 1993 (1980).
(4) Trop H.S., Davison A., Jones A.G., Davis M.A., Szalda D.J. and Lippard S.J., Inorg. Chem., 19, 1105 (1980).
(5) Mazzi U., Clemente D.A., Bandoli G. and Magon L., Transition Met. Chem., 4, 151 (1979).

SYNTHESIS AND CHARACTERIZATION OF NEUTRAL AMINOETHANOL COMPLEXES OF TECHNETIUM

A.V. Kramer, L.A. Epps, N. Ranganathan, H.T. Ravert, and H.D. Burns.
Divisions of Nuclear Medicine and Radiation Health Sciences, The Johns Hopkins University, 615 North Wolfe Street, Baltimore, Maryland 21205-2179.

The identification of stable, neutral and lipophilic Tc-99m agents capable of passive diffusion across the blood brain barrier and cell membranes has been the goal of several recent investigations (1-4). It has been suggested that such complexes might be useful for measuring brain blood flow (1-3) or for incorporation into bifunctional radiotracers designed to mimic metabolically active substrates or localize at intracellular receptors (4).

As part of our program to develop such radiotracers, we have found that N-substituted N,N'-diethanolethylenediamines (I) react with $TcOCl_4^-$ (5) in methanol to form neutral technetium (V) complexes formulated as TcOLX (II), where X = OH, OCH_3 or Cl depending upon conditions. Similar results have been observed in the $SnCl_2$ reduction of TcO_4^- in the presence of excess ligand.

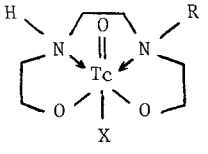


Ia R = H, R' = CH_3 MeDADAH$_2$

Ib R = H, R' = CH_2CH_2OH DATAH$_3$

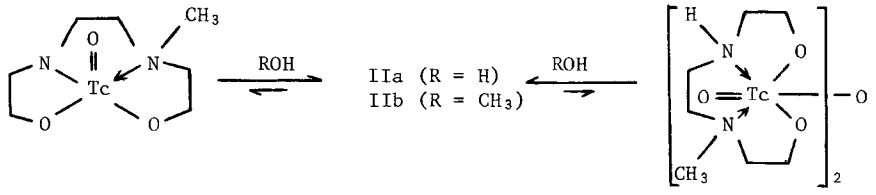
The Tc-99 N-methyl-N,N'-diethanolethylenediamine complex (Tc-MeDADA) was isolated as dark red crystals. Elemental analysis, infrared spectroscopy and mass spectrometry suggest it exists in the solid state as the N-deprotonated complex (IV) although the oxo-bridged dimer (V) cannot be ruled out. Although sparingly soluble in aprotic solvents, it dissolves readily in water and methanol to give yellow-green solutions of the corresponding monomers (II). In aqueous media, complex IIa is stable in the pH range 5-8 and is a non-electrolyte as determined by ion exchange chromatography, electrophoresis and conductivity measurements. NMR spectroscopy shows a single isomer of undetermined structure dominates in solution.

The N,N,N'-triethanolethylenediamine reacts with $TcOCl_4^-$ and TcO_4^- under similar conditions to yield a yellow-green solid (Tc-DATA) formulated as TcOLCl (IIIc) in which only two OH groups are deprotonated.



IIa R = CH_3, X = OH
IIb R = CH_3, X = OCH_3
IIc R = CH_3, X = Cl
IIIa R = CH_2CH_2OH, X = OH
IIIb R = CH_2CH_2OH, X = OCH_3
IIIc R = CH_2CH_2OH, X = Cl

Complexes II and III can be prepared on the Tc-99m level from TcO_4^- using similar reaction conditions. Work is currently underway to (a) synthesize N-substituted analogs of the parent complexes to enhance lipophilicity and impart physiological specificity and (b) evaluate their in vivo stability and distribution for possible use in bifunctional radiopharmaceuticals.



IV V

(1) Oldendorf W.H., J. Nucl. Med. 19, 1182 (1978).
(2) Loberg M.D., Corder E.H., Fields A.T. and Callesy P.S., J. Nucl. Med. 20, 1181 (1979).
(3) Kung H.F. and Blau M., J. Nucl. Med. 21, 147 (1980).
(4) Burns H.D., Dannals R.F., Marzilli L.G. and Wagner H.N. Jr., J. Nucl. Med. 20, 641 (1979).
(5) Cotton F.A., Davison A., Day V.W., Gage L.D. and Trop H.S., Inorg. Chem. 18, 3024 (1979).

Analytical Data

Elemental analysis:

		C	H	N
Tc-MeDADA	Calculated (IV)	30.67	5.52	10.22
	Calculated (V)	29.70	5.70	9.89
	Found	29.78	5.79	9.33
Tc-DATA	Calculated	28.21	5.32	8.22
	Found	27.92	5.17	7.98

IR spectroscopy (KBr):

	Possible $\nu_{Tc=O}$ (cm^{-1})
Tc-MeDADA (IV or V)	915(s), 935(s), 985(s)
Tc-DATA	928(s), 940(s)

Mass spectrometry:

		m/e	Assignment
Tc-MeDADA	(FD-MS)	274	$O=Tc(MeDADA-H)^+$
	(FAB-MS)	275	$O=Tc(MeDADA)^+$
Tc-DATA	(FD-MS)	304	$O=Tc(DATA-H)^+$

UV/VIS spectroscopy (MeOH):

	λ_{max} (nm)	ε
Tc-MeDADA	260	---
	400	450
Tc-DATA	260	---
	402	500

REACTION OF CYCLAM WITH REDUCED NO-CARRIER-ADDED Tc-99m

D.E. Troutner, W.A. Volkert, C. Reid, T. Hoffman, and R.A. Holmes.
Department of Chemistry, University of Missouri, Columbia, Missouri 65211.

Macrocyclic amines readily form complexes with technetium-99m (Tc) in high yields in basic aqueous media and exhibit exceptional stability. Because of these characteristics, this type of ligand is well suited to be used as a basis to formulate new Tc radiopharmaceuticals. Tc complexes of one of the simplest macrocyclic amines, cyclam (1,4,8,11-tetraazacyclotetradecane), is being extensively investigated in this laboratory. Radiochemical properties (1) of Tc-cyclam (Tc-CYC) and the crystal structure (2) of Tc-99-CYC $[Tc(V)O_2(cyclam)]^{+1}$ have been described. Most of these studies were performed at cyclam concentrations of 10^{-3} M or greater. Cyclam-based-radiopharmaceuticals, however, may have limited solubility in water and complexation at low concentrations will require more knowledge of the conditions necessary to maximize labeling yields. For this reason a study to investigate the factors affecting labeling of cyclam at low concentrations in aqueous media by direct reduction of TcO_4^- by stannous ion was initiated.

In initial experiments, Tc-CYC complexes were prepared in open vials. To 5 ml of cyclam (10^{-3} to 10^{-5} M) were added 0.5 ml of 0.1M NaOH and 0.3 ml of a no-carrier-added Tc solution which had been eluted from a generator with 0.9% saline and diluted 10-fold with water. To this solution was added 0.2 ml of a saturated stannous tartrate solution (10^{-4}M) and the time of this addition taken as the start of the reaction. Concentrations after this mixing were: cyclam, (8-800) x 10^{-6}M; NaOH, 8 x 10^{-3}M; NaCl, 8 x 10^{-4}M; $SnC_4H_4O_6$, 3 x 10^{-6}M, and TcO_4^-, (5-20) x 10^{-10}M. Samples were withdrawn after 1, 5, 10 and 50 min and analyzed for reduced Tc(R), free TcO_4^-(T), and Tc-CYC(C) by paper chromatography using the acetone-saline method of Colombetti, et al. (3). Other experiments were done in a sealed system under N_2-purge. Samples were withdrawn by glass capillary tubes through a rubber septum. Some typical results for the two methods are shown in Tables 1 and 2.

Table 1. Percent of reduced Tc, TcO_4^-, and Tc-CYC as a function of time at pH 12 in air.

[CYC]	8 x 10^{-4}			8 x 10^{-5}			8 x 10^{-6}			0	
Time (min)	R	T	C	R	T	C	R	T	C	R	T
1	1	0	99	38	3	59	89	3	8	97	2
5	0	0	100	5	1	94	59	12	29	81	15
10	0	0	100	2	1	97	44	24	32	25	67
50	0	0	100	1	1	98	4	57	39	4	91

Table 2. Percent of reduced Tc, TcO_4^-, and Tc-CYC as a function of time at pH 12 under N_2.

[CYC]	8 x 10^{-5}			4 x 10^{-5}			8 x 10^{-6}			0	
Time (min)	R	T	C	R	T	C	R	T	C	R	T
1	35	1	64	68	2	30	85	2	13	96	2
5	6	0	94	27	1	72	74	2	24	93	2
10	3	0	97	14	1	85	63	2	35	88	3
50	3	0	97	6	2	92	37	3	60	92	5

Other experiments were performed at pH 8, 10 and 13 for reaction times up to three hrs. Two linear amines, 1,5,8,12-tetraazadodecane(I) and 1,4,8,11-tetraazaundecane(II) were tested as well as two other macrocyclics, 1,4,8,11-tetramethyl-1,4,8,11-teraazacyclotetradecane(III) and racemic 5,7,7,12,14,14-hexamethyl-1,4,8,11-tetraazacyclotetradecane(IV).

Results in Tables I and II show that the rate of formation of Tc-CYC at pH 12 is dependent on cyclam concentration and that at low cyclam concentrations the reoxidation to TcO_4^- is a competing reaction. The reduced product (R) identified by chromatography using saline at neutral pH as the solvent is a combination of a "reactive-reduced"-intermediate, which can be either oxidized or be complexed by the amine ligands, and an "unreactive-reduced" species (presumably hydrolyzed-reduced-Tc). Under N_2 there was little reoxidation to TcO_4^-, but there was what appeared to be a slow conversion to an "unreactive-reduced" species of Tc which was not complexed by cyclam. When TcO_4^- was reduced in the absence of cyclam followed by the addition of cyclam after 30 min complexing occurred within a few mins. However, when excess cyclam was added to solutions in which reduced Tc persisted after 3 hrs no further complexing was observed. The extent of the formation of this "unreactive reduced" Tc species increased at lower pH's, at reduced concentrations of cyclam, and at increased concentrations of $SnC_4H_4O_6$.

At cyclam concentrations of 4×10^{-5} M or higher it appeared that the rate of complexation of the "reactive-reduced" Tc species could be approximated by a first order expression with a half-time approximately equal to $(9 \times 10^{-5}/[CYC])$ min. Estimated half-times for experiments at lower concentrations are consistent with this value if only early times are considered. At pH 8 or 10, the rates of complexation were approximately 10-fold lower which may be due to increased protonation of the ligand and/or increased rate of formation of the "unreactive reduced" Tc species. At pH 13 there was no change in the rate. The rate of reaction of ligands I and II was estimated to be about 20-40 times faster than that of cyclam and that for ligand III about 40 times slower. No estimate could be made for ligand IV.

In summary, when TcO_4^-, at no-carrier-added levels, undergoes direct reduction by $SnC_4H_4O_6$ at pH 12 or higher, a meta-stable "reactive-reduced" species is formed which complexes with cyclam with a half-time approximately equal to $(9 \times 10^{-5}/[CYC])$ min. This species is entirely oxidized to TcO_4^- in air saturated solutions with no ligand present and partially oxidized at 8×10^{-6} M cyclam. In the absence of oxygen, less than 3 percent TcO_4^- is formed and any Tc not complexed to the cyclam within 2 hrs has been converted into an "unreactive-reduced" form. At lower pH, the rate of conversion of the "reactive-reduced" species to the unreactive form appears to increase while the rate of complexation decreases. The use of Tc-99 should help clarify the nature and perhaps the structure of this meta-stable reduced form. Finally, these results suggest that radiopharmaceuticals based on cyclam can be prepared in high yield at pH 12 at a cyclam concentrations of 4×10^{-5} M or higher.

(1) Simon, J., D.E. Troutner, W.A. Volkert, and R.A. Holmes, Radiochem. Radioanal. Letters 47, 111 (1981).
(2) Zuckman, S.A., G.M. Freeman, D.E. Troutner, W.A. Volkert, R.A. Holmes, D.G. Van Derveer, and E.K. Barefield, Inorg. Chem. 20, 2386 (1981).
(3) Colombetti, L. G., S. Moerlein, G.C. Patel, et al., J. Nucl. Med. 17, 805 (1976).

SYNTHESIS OF ^{99m}Tc: 2,6-DIMETHYL-, 2,6-DIETHYL-, 2,6-DIISOPROPYL-, AND 4-n-BUTYL-PHENYLCARBAMOYLMETHYL-IMINODIACETIC ACID

A.E.A. Mitta, C. Archiprete, and E.G. Gros.

Gerencia de Investigaciones, Comisión Nacional de Energía Atómica, Av. Libertador 8250, 1429 Buenos Aires, Argentina.

Since Harvey et al.(1) reported the synthesis of 2,6-dimethyl-phenyl carbamoylmethyl iminodiacetic acid (I) and tested it as a ^{99m}Tc complex formation agent, several authors informed about the preparation of iminodiacetic acid derivatives to be used for the same purposes (2-6).

We wish to report here the synthesis, physical properties and assays as complex agent for the mentioned isotope of compound I, and of 2,6-diethyl-phenylcarbamoylmethyl iminodiacetic acid (II), 2,6-diisopropyl-phenylcarbamoylmethyl iminodiacetic acid (III), and 4-n-butyl-phenylcarbamoylmethyl iminodiacetic acid (IV).

The synthesis of I, II, III, and IV were accomplished as indicated in Scheme 1. Their physical properties (m.p., analysis, ^{1}H-NMR spectra and mass spectra) are presented in Tables 1, 2, and 3.

SCHEME I

SYNTHESIS OF COMPOUNDS I, II, III, IV

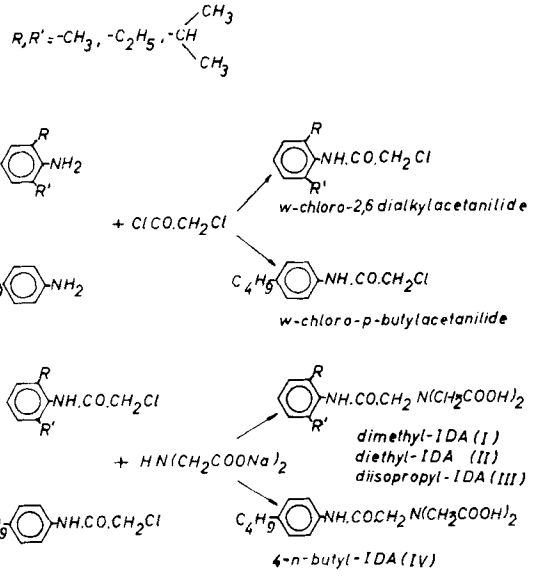
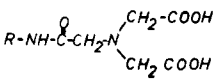


Table 1 MICROANALYSIS

Compound	Yield%	m.p.		Calculated (%) C	H	N	Found (%) C	H	N
I	74	218°	$C_{14}H_{18}N_2O_5$	57.13	6.17	9.52	57.15	6.39	9.55
II	79	186-188°	$C_{16}H_{22}N_2O_5$	59.61	6.88	8.69	59.70	7.01	8.67
III	75	197-198°	$C_{18}H_{26}N_2O_5$	61.70	7.48	8.00	61.66	7.55	7.78
IV	86	196-198°	$C_{16}H_{22}N_2O_5$	59.61	6.88	8.69	59.84	7.02	8.89

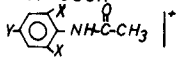
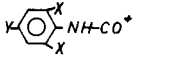
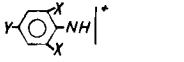
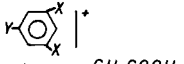
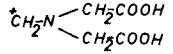
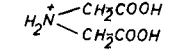
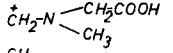
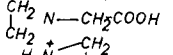
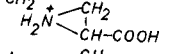
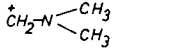
TABLE 2

MASS SPECTRA



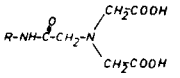
1) R =  2) R = 

3) R =  4) R = n-Bu 

ASSIGNATION	m/e (%)			
	1 X = Me, Y = H	2 X = Et, Y = H	3 X = iPr, Y = H	X = H, Y = n-Bu
M^+	294(0)	322(0)	350(0)	322(0)
$M^+ - COOH$	249(2)	277(3)	305(3)	277(3)
	163(37)	191(15)	219(11)	191(17)
	148(9)	176(20)	204(39)	176(11)
	120(49)	148(47)	176(16)	148(10)
	106(11)	134(21)	162(14)	134(29)
	146(100)	146(100)	146(100)	146(100)
	134(21)	134(21)	134(4)	134(3)
	102(68)	102(82)	102(70)	102(70)
	101(42)	101(19)	101(10)	101(53)
	88(99)	88(68)	88(47)	88(72)
	58(62)	58(50)	58(41)	58(74)

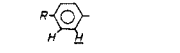
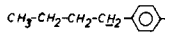
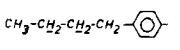
PMR SPECTRA

TABLE 3



1) R =  2) R = 

3) R =  4) R = n-Bu 

ASSIGNATION	δ (ppm)			
	1	2	3	4
-COOH	9,38 (s)	9,30 (s)	9,42 (s)	10,36 (s)
$-N-CH_2-COOH$	3,56 (s)	3,48 (s)	3,58 (s)	3,58 (s)
$-\overset{O}{\overset{\|}{C}}-CH_2-N$	3,48 (s)	3,40 (s)	3,52 (s)	3,50 (s)
	2,13 (s)	—	—	—
	—	0,98 (t)	—	—
	—	2,40 (c)	—	—
	—	—	1,12 (d)	—
	—	—	3,05 (m)	—
	7,00 (s)	6,98 (s)	7,18 (s)	—
	—	—	—	7,55 (d)
	—	—	—	7,15 (d)
	—	—	—	2,55 (t)
	—	—	—	1,45 (bd)
	—	—	—	0,90 (t)

(1) Harvey, E., Loberg, M.D. and Cooper, M., J.Nucl.Med. 16, 533 (1975).
(2) Loberg, M.D., Cooper, M., Harvey, E., Callery, P. and Faith, W., J.Nucl.Med., 17, 633 (1976).
(3) Callery, P., Faith, W., Loberg, M.D., Fields, A., Harvey, E. and Cooper, M., J.Med.Chem., 19, 962 (1976).
(4) Ryan, J., Cooper, M., Loberg, M.D., Harvey, E. and Sikorski, S., J.Nucl.Med., 18, 997 (1977).
(5) Loberg, M.D. and Fields, A., Int.J.Appl.Radiat.Isot., 29, 167 (1978).
(6) Fields, A., Porter, D.W., Callery, P., Harvey, E. and Loberg, M.D., J.Lab.Compds.Radiopharm., 15, 387 (1978).

THE ROLE OF THE PROTON IN TECHNETIUM REDUCTION AND COMPLEXATION

J.B. Slater, J.F. Harwig, and W. Wolf.
Radiopharmacy Program, University of Southern California, Los Angeles, CA 90033.

A primary requirement in the rational design of Tc radiopharmaceuticals is an understanding of the reduction and complexation processes of Tc. A major problem in defining these aspects of Tc chemistry is the formation of reduced hydrolyzed species of Tc in the presence or absence of ligands. Electrochemical studies of Tc in nonaqueous systems, on the other hand, show a one-electron reduction of Tc, to oxidation state VI ($TcO_4^- + e^- \longrightarrow TcO_4^{=}$), with no indication of other reduced species (1,2). In the presence of a ligand with a suitable proton and coordinating group, quantitative reduction of Tc below VI and accompanying complexation with the ligand occur (2). To further characterize these processes, the present work focuses on the key role of the proton.

The studies were performed by polarography with a PAR Model 174A polarographic analyzer in both the sampled DC and differential pulse modes. A dropping Hg electrode served as the working electrode, with a Pt auxillary electrode and a SCE reference electrode. A Hg flow rate of 0.98 mg/sec was used with a drop time of 0.5 sec. Oxidation states were determined by the Ilkovic equation. The solvents used were acetonitrile (CH_3CN), dimethylformamide (DMF), dimethylsulfoxide (DMSO), and tetramethylurea (TMU), with the supporting electrolyte $(CH_3)_4NPF_6$. The ligands studied were oxine and its $(CH_3)_4N^+$ salt, the ortho and meta isomers of aminophenol and aminothiophenol, catechol, resorcincol, and quinoline-8-carboxylic acid. The external proton sources included NH_4^+, phenol, and 2,4,6-trimethylphenol. Tc was used in the form of the NH_4^+ and $(CH_3)_4N^+$ salts of $^{99}TcO_4^-$.

In the absence of external proton sources Tc reduction below VI and complexation occurred with oxine in all solvent systems, corresponding to a Tc state of IV and a 3:1 molar ratio of oxine:Tc. Tc reduction below VI and complexation also resulted with both the ortho and meta isomers of aminothiophenol, but only in CH_3CN. For the ortho isomer a single four-electron Tc reduction wave was found, while for the meta isomer two Tc reduction waves, with an overall transfer of three electrons, were observed. In the presence of catechol in CH_3CN the Tc reduction wave shifted to a less negative potential, but Tc remained in the VI state. A 3:1 molar ratio of catechol:Tc shifted all the Tc to the new potential. With resorcinol, quinoline-8-carboxylic acid, and the ortho and meta isomers of aminophenol Tc reduction below VI and complexation did not occur, although a wave attributable to reduction of the labile proton was observed in each case. In DMF and DMSO the aminothiophenol isomers also displayed this behavior. No effect at all was seen with the oxine salt.

When external proton sources were added to the above ligands with which Tc reduction below VI and complexation had not been observed, a change in behavior occurred in only one case, the oxine salt. The proton sources NH_4^+ and phenol caused the orange color of the oxine anion to disappear, and a complex essentially identical to the oxine complex was formed. With 2,4,6-trimethylphenol as the proton source the color of the oxine anion remained unchanged, and Tc reduction below VI and complexation were not observed.

The present results further support our previous conclusion (2) that reduction of Tc below VI requires, in addition to the reducting agent itself, both a suitable proton source and an appropriate coordinating group, with the proper spatial relationship, to remove oxygen from TcO_4^-. Oxine, with its internal hydroxylic proton and aromatic ring nitrogen, in fixed steric arrangement, seems to fulfill these requirements perfectly in all the solvent systems, while the aminothiophenol isomers and catechol exhibit a solvent dependence. The other ligands appear completely unable to promote Tc reduction below VI and complexation, possible reasons for which include: pKa of ligand internal proton too low (proton reduces too easily), pKa of ligand internal proton too high (proton not sufficiently acidic), coordinating group of ligand not a satisfactory electron pair donor. An external proton source with proper pKa, although potentially able to obviate the first two problems, requires an unfavorable

ternary reaction with the TcO_4^- and the ligand, the latter two species being negatively charged. Only with the oxine salt was a complex observed in the presence of external proton sources, and only because these proton sources simply protonated the oxine anion, producing oxine itself. These results demonstrate some of the requirements and limitations of the proton in its role as a key factor in the chemistry of Tc in both nonaqueous and aqueous systems.

(1) Astheimer L. and Schwochau K., J. Inorg. Nucl. Chem., 38, 1131 (1976).
(2) Slater J.B., Harwig J.F., and Wolf W., Submitted to 29th Annual Meeting of the Society of Nuclear Medicine, Miami Beach, June, 1982.

TABLE 1 Tc Reduction Potential In Absence of Ligand

Solvent	Reduction Potential	Oxidation State
	$(CH_3)_4N^+$ Salt of TcO_4^-	
CH_3CN	-1.75	VI
DMF	-1.83	VI
DMSO	-1.86	VI
	NH_4^+ Salt of TcO_4^-	
CH_3CN	-1.82	VI
DMF	-1.88	VI
DMSO	-1.86	VI
TMU	-2.18	VI

TABLE 2 Reduction Potentials of Ligands

Ligand	Solvent	Reduction Potential	pKa In Water	
			1	2
o-amino-phenol	CH_3CN	-1.63*	9.71	
	DMF	-1.77*		
	DMSO	-1.68*		
m-amino-phenol	CH_3CN	-1.61*	9.87	
Catechol	CH_3CN	-1.42*	9.48	12.08
	DMF	-1.73*		
	DMSO	-1.71*		
Resorcinol	CH_3CN	-1.53*	9.44	11.32
Quinoline-8-COOH	CH_3CN	-1.29*		
o-amino-thiophenol	DMF	-1.62*		
	DMSO	-1.65*		
	CH_3CN	-1.89**		
	DMF	-1.89**		
m-amino-thiophenol	CH_3CN	-1.81**		
Oxine	CH_3CN	-1.91**	5.02	9.81
	DMF	-1.88**		
	DMSO	-1.95**		
	TMU	-2.3**		
Oxine $(CH_3)_4N^+$ Salt	CH_3CN TMU	Does not reduce " "		

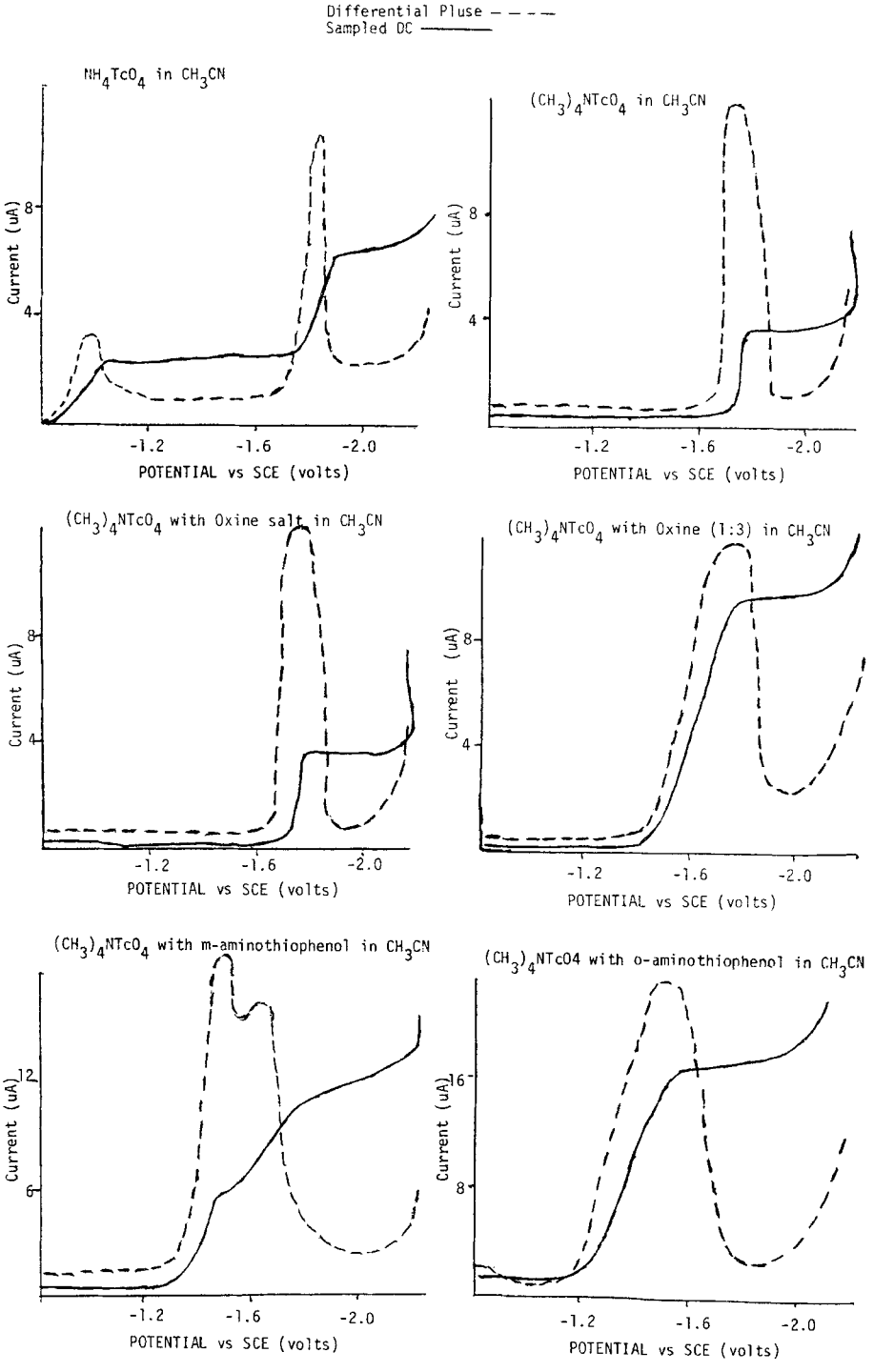
*In the presence of TcO_4^-
**In the absence of TcO_4^-

TABLE 3 Tc Reduction Potential In Presence of Ligand

Ligand	Solvent	Reduction Potential	Oxidation State
	$(CH_3)_4N^+$ Salt of TcO_4^-		
Oxine	CH_3CN	-1.70	IV
	DMF	-1.72	IV
	DMSO	-1.73	IV
Catechol	CH_3CN	-1.67	VI
o-amino-thiophenol	CH_3CN	-1.38	III
m-amino-thiophenol	CH_3CN	-1.45	
		-1.65	IV
	NH_4^+ Salt of TcO_4^-		
Oxine	CH_3CN	-1.66	IV
	DMF	-1.75	IV
	DMSO	-1.74	IV
	TMU	-1.65	IV

TABLE 4 Reduction Potentials of External Proton Sources (In the Presence of TcO_4^-)

Proton Source	Solvent	Reduction Potential	pKa In Water
Phenol	CH_3CN	-1.70	9.99
	DMF	-1.74	
2,4,6-Tri-methylphenol	CH_3CN	-1.70	10.88-10.99
	DMF	-1.78	
NH^+	CH_3CN	-0.95	9.25
	DMF	-1.57	
	DMSO	-1.57	
	TMU	-1.75	


Differential Pluse - - - -
Sampled DC ————
NH_4TcO_4 in CH_3CN
$(CH_3)_4NTcO_4$ in CH_3CN
$(CH_3)_4NTcO_4$ with Oxine salt in CH_3CN
$(CH_3)_4NTcO_4$ with Oxine (1:3) in CH_3CN
$(CH_3)_4NTcO_4$ with m-aminothiophenol in CH_3CN
$(CH_3)_4NTcO4$ with o-aminothiophenol in CH_3CN
Current (uA)
POTENTIAL vs SCE (volts)
-1.2
-1.6
-2.0
4
8
12
16

OXOTECHNETIUM COMPLEXES CONTAINING $TcON_2S_2$ CORES

A.G. Jones, A. Davison, J.W. Brodack, D.E. Brenner, J. Lister-James, C.E. Costello, C.J.L. Lock, K.J. Franklin, M.R. LaTegola-Graff, C. Orvig, and M. Sohn.
Department of Radiology, Harvard Medical School, Boston, MA 02115.

Synthesis of the tetradentate ligands N,N'-ethylenebis-(2-mercaptoacetamide), N,N'-bis(2-mercaptoethyloxamide), and 2-mercaptoacetylglycylcysteamine has allowed the isolation of three isomeric oxotechnetium(V) anions with the general formula $[TcC_4H_4N_2O_3S_2]^{1-}$. The ligands and hence the resulting complexes differ only in the positions of the two carboxyl groups on the ligand backbone. These materials can be prepared at carrier and no carrier added concentrations by reduction of pertechnetate with sodium dithionite in aqueous ethanolic base in the presence of the bis-S-benzoyl protected ligands.

The complex oxo[N,N'-ethylenebis(2-mercaptoacetimido)]technetate(V) has been fully characterized by elemental analysis, UV/VIS, IR and NMR spectroscopy, field desorption mass spectrometry (both positive and negative ion mode), and by a single crystal x-ray determination. The technetium is coordinated to an oxygen atom and to the two sulfur and nitrogen atoms of the ligand, forming a distorted square pyramid with the oxygen at the apex. The Tc-O bond length is 1.679(5) Å, almost identical to the 1.672(8) Å seen in cis-$[TcO(SCH_2CH_2S)_2]^{1-}$ (1). The distance of the technetium atom above the square plane is also similar, 0.771(5) Å versus 0.791(8) Å, respectively. As reported previously (2,3) this material undergoes rapid renal excretion with 70-75% of the injected dose appearing in urine by 60 minutes. A small proportion (5-7%) is cleared through bile. Using a combination of reverse-phase ion pair HPLC and field desorption mass spectrometry, it has been shown that the complex is excreted unchanged through both pathways. Preliminary studies with the other two isomers indicate similar rates of renal excretion, but with somewhat less of the radioactivity passing into bile.

These basic ligand backbones can be readily substituted, and this can lead to marked changes in distribution. Functionalization also produces syn and anti isomers that are enantiomeric pairs of diastereomers, except where the side-chain is introduced as an optically pure reagent without racemization. The diastereomers can be separated by HPLC and these also show differences in their biological properties (4).

(1) DePamphilis B.V., Jones A.G., Davis M.A. and Davison A., J. Am. Chem. Soc., 100, 5570 (1978).
(2) Davison A., Sohn M., Orvig C., Jones A.G. and LaTegola M.R., J. Nucl. Med., 20, 641 (1979).
(3) Fritzberg A.R., Klingensmith W.C., III, Whitney W.P. and Kuni C.C., J. Nucl. Med., 22, 258 (1981).
(4) Fritzberg A.R., private communication (1981).

SYNTHESIS, CHARACTERIZATION, AND ELECTROCHEMISTRY OF TRANS DIHALO TECHNETIUM PHOSPHINE COMPLEXES: POTENTIAL MYOCARDIAL IMAGING AGENTS

K.A. Glavan, J.F. Kronauge, E. Blaubaugh, R.D. Neirinckx, M.N. Eakins, and M.D. Loberg. Squibb Institute of Medical Research, Georges Road, New Brunswick, New Jersey 08903.

There has recently been considerable interest in technetium chemistry as it is applied to the development of new Tc-99m radiopharmaceuticals. Much of this interest has focused on cationic technetium complexes having the formula trans-$[TcD_2X_2]^+$ where D is bis(1,2-dimethylarsino)benzene (diars) or bis(1,2-dimethylphosphino)ethane (dmpe) and X is Cl, Br, or I. This interest is predicated in part on the ability of these complexes to concentrate in normal myocardium (1-3). An investigation was therefore conducted into the synthesis, electrochemistry, and in vivo distribution of a series of Tc-phosphine complexes having the formula trans-$[TcD2Cl_2]^+$ where D is bis(1,2-dimethylphosphino)ethane (dmpe), bis(1,2-diethylphosphino)ethane (depe), bis(1,2-di-n-propylphosphino)ethane (dppe), bis(1,2-di-n-butylphosphino)ethane (dbpe), bis(1,2-diphenylphosphino)ethane (dpe), and bis-(1,2-dimethylphosphino)benzene (diphos). These complexes were synthesized using both Tc-99 and "no carrier added" Tc-99m. The Tc-99 complexes were characterized by UV/VIS, IR, elemental analysis, fast atom bombardment mass spectroscopy (e.g., Figure I and II), and single crystal X-ray analysis. The Tc-99m complexes were characterized by HPLC. Rat distribution studies were completed for both the Tc-99 and Tc-99m complexes. Since the $[Tc(dmpe)_2Cl_2]^+$ complex exhibited the most promising myocardial uptake, the series of complexes $[Tc(dmpe)_2X_2]^+$ where X is Cl, Br, or I was also studied. Table I presents an example of the in vivo distribution data for the $[^{99}Tc(dmpe)_2Cl_2]^+$, $[^{99}Tc(dmpe)_2Br_2]^+$, and $[^{99}Tc(depe)_2Cl_2]^+$ complexes. The n-octanol/phosphate buffer partition coefficients for the Tc-99 dichloro complexes are listed in Table II. This investigation demonstrated the importance of structure and possibly lipophilicity as influencing factors in the myocardial uptake of these cationic technetium complexes.

Technetium electrochemistry has also received considerable attention recently as it relates to the body's ability to differentiate among these types of cationic Tc-arsine and Tc-phosphine complexes. (4). The electrochemical behaviour of an expanded series of Tc-phosphine complexes was investigated, and Table III lists the reduction potentials for the corresponding Tc(III)/Tc(II) couples. As seen from the table, reduction to the Tc(II) state for these Tc-phosphine complexes occurs at potentials which are accessible to biological systems. Thus, the biologically accessible redox couples of these complexes may be important as a source of biological differentiation among these cationic Tc-phosphine complexes.

References

1. Deutsch E., Glavan K.A., Sodd V.J., Nishiyama H., Fergusson D.L., and Lukes S.J.: Cationic Tc-99m Complexes as Potential Myocardial Imaging Agents. J Nucl Med 22: 897 (1981).
2. Deutsch E., Bushong W., Glavan K.A., Elder R.C., Sodd V.J., Scholz K.L., Fortman D.L., and Lukes S.J.: Science 214, 85 (1981).
3. Deutsch E., Glavan K.A., Bushong W., and Sodd V.J.: Applications of Nuclear and Radiochemistry (Pergamon, New York, in press).
4. Hurst R.W., Heineman W.R., and Deutsch E.: Inorg. Chem. 20: 3298 (1981).

Table I. Uptake of ^{99}Tc-labelled complexes in rats at 5 minutes post injection

	% Injected Dose/g Tissue		
Tissue	$Tc(dmpe)_2Cl_2^+$	$Tc(dmpe)_2Br_2^+$	$Tc(depe)_2Cl_2^+$
Blood	0.17	0.13	0.24
Heart	2.4	1.5	1.1
Lung	0.77	0.74	1.3
Liver	1.2	1.2	2.5
Kidney	4.9	4.0	3.3
Muscle	0.56	0.18	0.23
Brain	0.09	0.06	0.05

Table II. Octanol/0.01 M Phosphate-Buffered Saline, pH 7.01 Partition Coefficients for the Tc-99 Complexes

Complex	Partition Coefficient
$[Tc(dmpe)_2Cl_2]^+$	2.65
$[Tc(depe)_2Cl_2]^+$	52.3
$[Tc(dppe)_2Cl_2]^+$	56.9
$[Tc(dbpe)_2Cl_2]^+$	78.9
$[Tc(dpe)_2Cl_2]^+$	119
$[Tc(diphos)_2Cl_2]^+$	106

[a]The octanol-to-buffered-saline partition coefficients were normalized to a constant weight of solvent.

Table III. Formal Reduction Potentials for the Tc(III)/Tc(II) Couples in N,N-Dimethylformamide[a]

Tc(III) complex	E°'-[Tc(III)/Tc(II)]
$Tc(dmpe)_2Cl_2^+$	- 0.250
$Tc(depe)_2Cl_2^+$	- 0.336
$Tc(dppe)_2Cl_2^+$	- 0.257
$Tc(dbpe)_2Cl_2^+$	- 0.290
$Tc(dpe)_2Cl_2^+$	- 0.063[b]

[a]Given in volts vs. NaSCE. The supporting electrolyte is 0.5 M tetraethylammonium perchlorate. E°' determined by averaging $E_{p(a)}$ and $E_{p(c)}$ values obtained by cyclic voltammetry. The redox couples are reversible unless otherwise noted. [b]Irreversible.

Fig 1 Positive Ion Fast Atom Bombardment Mass Spectrum of $[Tc(dmpe)_2Cl_2]^+$

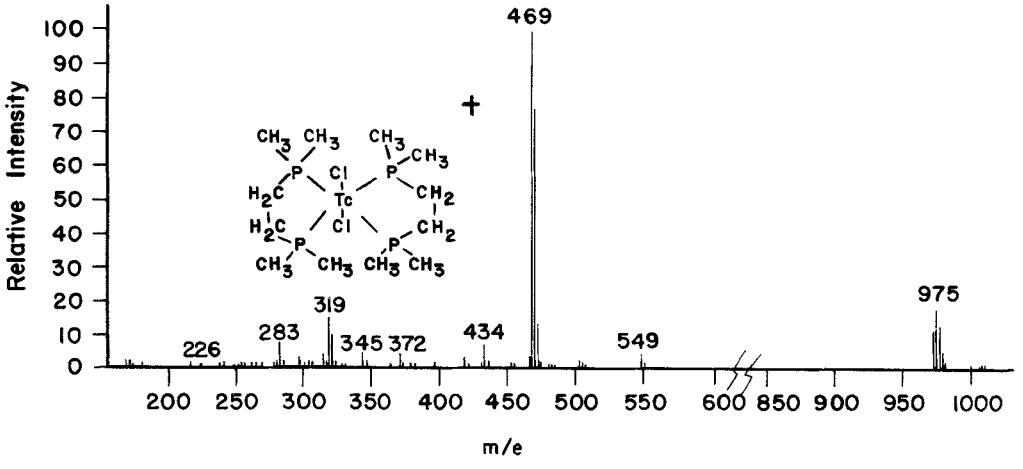
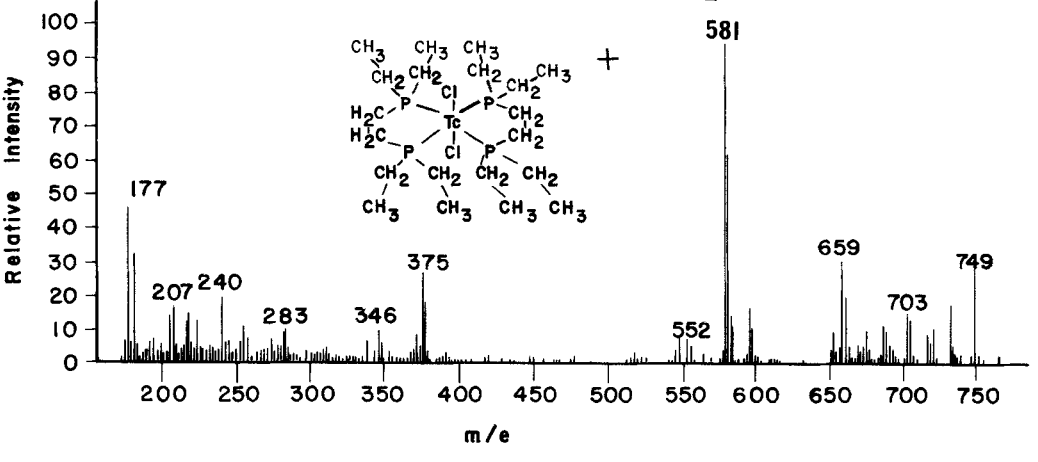


Fig 2 Positive Ion Fast Atom Bombardment Mass Spectrum of $[Tc(depe)_2Cl_2]^+$



$N_2{}^{15}O$ SYNTHESIS FOR MEDICAL USE

C. Crouzel and J.C. Baron.
Service Hospitalier Frédéric Joliot, CEA Département de Biologie, 91406 Orsay, France.

The most accessible and best developed method available today for cerebral blood flow (CBF) measurement by positron emission tomography is the continuous inhalation technique using CO_2 labeled with oxygen-15[1]. Among the theoretical limitations of the technique, the most important is the assumption that oxygen-15 labeled water (the actual tracer of blood flow which is produced in vivo after inhalation of labeled CO_2) is freely diffusible across the blood-brain barrier. This assumption has been demonstrated to be inaccurate in several studies using both animals[2] and man[3].

The use of $N_2{}^{15}O$, an inert and freely diffusible gas at any flow rate, would allow an improved measurement of CBF even with its other limitations which arise from its short half-life.

It was therefore of interest to investigate the continuous production of nitrous oxide labeled with oxygen-15.

This has been accomplished by oxidation of ammonia using a stream of ^{15}O-labeled oxygen [4].

$$4\ NH_3 + 4\ O_2 \rightarrow 2\ N_2O + 6\ H_2O$$

The oxygen-15 was produced by irradiation of an N_2 + 2 % O_2 mixture with 8 MeV deuterons[5]. Ammonia was added to the target gases as they emerged from the target and the mixture was passed through a furnace containing a platinum on alumina catalyst at 300°C.

The oxidation conditions : gas flow over the catalyst, quantities of ammonia and oxygen, dimensions of the furnace, and quantity of the platinum catalyst were studied as well as the stability of the catalyst. Analysis of the reaction products was performed using gas chromatography ; 50 - 80 % of the activity leaving the catalyst was found to be $N_2{}^{15}O$ according to the conditions used.

The purification of the gas to remove unreacted oxygen gas and nitrogen oxides that might have formed is under study.

Using a flow rate at the target of 450 ml/min, a 25 µA irradiation yielded 40 µCi/ml of $N_2{}^{15}O$ with 3 % N_2O carrier at the outlet of the system located 20 metres from the target.

(1) Jones T., Chesler D.A., Ter Pogossian M.M., Brit. J. Radiol., April 1976, 49, 339-343.
(2) Eichling J., Raichle M., Grub R., Ter Pogossian M., Circ. Research, 35, 1974, 358-364.
(3) Paulson O., Hertz M., Bolwig T., Lassen N., Micro. Vascular Research 13, 1977, 113-124.
(4) De Laney J.E., Manogue W.H., Cat. Proc. Int. Congr. 5th, 267-277, 1972.
(5) Clark J.C., Buckingham P.D.,"Short-lived radioactive gases for clinical use",Butterworth, 1975, 135-140.

This work was supported in part by a grant N°120006 from Institut National de la Santé et de la Recherche Médicale.

ON-LINE PRODUCTION OF (^{13}N)-NITROGEN FROM SOLID ENRICHED (^{13}C)-TARGETS, AND ITS APPLICATION TO (^{13}N)-AMMONIA SYNTHESIS USING MICROWAVE RADIATION

R.A. Ferrieri, D.J. Schlyer, A.P. Wolf, and B. Wieland.
Department of Chemistry, Brookhaven National Laboratory, Upton NY 11973.

The unique application of ^{13}N as a tracer for the metabolic fate of various nitrogen containing molecules (1-3) has led to an interest in methods for preparing ^{13}N-labelled nitrogen gas from either solid ^{12}C targets or gas targets consisting of some volatile form of ^{12}C or ^{16}O (cf. 4 for review). Recent work has shown that much higher yields of high specific activity (~ 20 mCi/ml) ^{13}N-labelled nitrogen gas can be produced from an enriched carbon-13 powder target utilizing the ^{13}C(p,n)^{13}N reaction, however, only a batch process target was described (5). In the present work, an on-line method to produce (^{13}N)-nitrogen gas from a carbon-13 powder target has been developed where the heat deposited from a high current, high energy proton beam (10 μA, 25 MeV) anneals the carbon-13 powder and releases (^{13}N)-nitrogen gas into a helium sweep. This technique has the distinct advantage of separating the radionuclide from the target matrix on-line and, since the target is reusable, eliminates the cost/effort problem of recovering the ^{13}C.

The present target design consists of a (^{12}C)-graphite cylinder packed with the (^{13}C)-powder and fixed within a quartz flow-through tube. The proton beam is then focused through a 3 mil molybdenum foil onto the graphite. Graphite was selected as the primary holder because of its rapid heat conductance property.

In preliminary studies, the effluent was found to be essentially free of radioactive species other than (^{13}N)-N_2 when the target was heated in a helium sweep prior to irradiation. An unheated target yielded predominantly ^{13}N-labelled oxides of nitrogen. Even with this pre-irradiation treatment, chemical impurities in a relative distribution of 87% O_2, 13% CO_2 were released into the target effluent at a rate of ~ 40 μmol/min for a standard helium sweep flowrate of 100 cc/min.

Direct application of the above target was found in the synthesis of (^{13}N)-NH_3, perhaps the most widely used ^{13}N-labelled precursor in ^{13}N-labelled compound synthesis. The synthesis of NH_3 through exposure of $N_2 + H_2$ to microwave radiation (6) has been successfully adapted to the synthesis of (^{13}N)-NH_3. The technique of using microwave radiation as an excitation method to synthesize labelled compounds has been employed in past studies at this laboratory (7,8). In the present study, no-carrier-added (^{13}N)-N_2 was recycled with excess H_2 through a standard Evenson microwave discharge cavity (cw microwave source of 100 watts at 2-3 GHz), and (^{13}N)-NH_3 was condensed out of the gas stream as it was produced. As seen in the Table, up to ~ 65% conversion of the (^{13}N)-N_2 extracted from the ^{13}C-powder has been achieved, but this yield was sensitive to the concentration of the chemical impurities (relative to H_2) eluting from the ^{13}C-powder.

This research was carried out at Brookhaven National Laboratory, in part, under contract with the U. S. Department of Energy and supported by its Office of Basic Energy Sciences and also supported by NIH Grant No. 15380.

(1) Meeks, J. C., Wolk, C. P., Lockau, W., et al. J. Bacteriol., 134, 125 (1978).
(2) Thomas, J., Meeks, J. C., Wolk, C. P., et al. J. Bacteriol. 129, 1545 (1977).
(3) Krohn, K.A. and Mathis, C.A. "The Use of Isotopic Nitrogen as a Biochemical Tracer," in Short-Lived Radionuclides in Chemistry and Biology, (J. Root and K. Krohn, Eds.) ACS Advances in Chemistry Series Monography, American Chemical Society, Washington, DC (1981).
(4) Hoop, B. Jr., Laughlin, J.S., and Tilbury, R.S. in Instrumentation in Nuclear Medicine (G. J. Hine and J. A. Sorensen, Eds.) Academic Press, New York, Vol. II, p. 407 (1974).
(5) Austin, S.M., Galonsky, A. and Bortins, G., Nucl. Instr. and Methods, 126, 373 (1975).
(6) Lalo, C., Vermeil, C., Bull. Soc. Chim. de France Partie I, 320 (1979).
(7) Wolf, A.P., "Labelling of Organic Compounds by Irradiation and Other Non-Synthetic Methods", Proc. of the 2nd Intl. Conf. on Methods of Preparing and Storing Labelled Compounds. Brussels, 907 (1966).
(8) Martinotti, F.F., Welch, M.J. and Wolf, A.P., Chem. Comm., 115 (1968).

Table of the % conversion of (^{13}N)-N_2 to (^{13}N)-NH_3 with the variation in the partial pressure ratio of target impurities to hydrogen

Partial Pressure Ratio of Target Impurities to H_2	% Conversion of (^{13}N)-N_2 to (^{13}N)-NH_3
4.850	13
3.950	16
1.780	31
1.320	23
1.110	33
0.400	34
0.390	34
0.090	60
0.075	74
0.048	56
0.040	63
0.036	49
0.027	63

THE REACTION OF $^{11}CO_2$ IN A MICROWAVE DISCHARGE

K. NIISAWA, J. SAITO, K.TAKI, T.KARASAWA, and T. NOZAKI.
Faculty of Industrial Hygiene, Kitasato University, 1-15-1, Kitasato, Sagamihara, Kanagawa, 228, Japan.

By the proton bomdardment of N_2-H_2 and N_2-O_2 systems, $^{11}CH_4$ and $^{11}CO_2$ are produced respectively (1,2), and can be used as precursors for various ^{11}C-radiopharmaceuticals.
The decomposition of ordinary amount of CH_4 in the microwave discharge in N_2 carrier gas is known to give various compounds such as C_2H_2 and HCN(3). On the other hand, the decomposition of no-carrier-added $^{11}CH_4$ gave only $^{11}CO_2$ in our experiments. This difference seems to be ascribable to the oxidation of $^{11}CH_4$ by trace O_2 as an impurity, whose amount is much larger than that of the no-carrier-added $^{11}CH_4$.
We therefore tried the microwave discharge in a gas mixture of no-carrier-added $^{11}CH_4$, N_2 and H_2 , the last being used for removal of impurity O_2 , and for the first time a small amount of $H^{11}CN$ was obtained.

The reaction vessel (ca. 270 ml volume) with a fan for the circulation of the gases was used as is shown in Fig. 1.
The products were analyzed by radiogaschromatography equipped with NaI(Tl) scintillation detector.

In order to improve the yield of $H^{11}CN$, its dependence on the reaction time, pressure and composition of the gases was investigated. The result is shown in Tab. 1.
It is clear that $H^{11}CN$ is produced as the main product under a suitable condition.
The microwave discharge has also been carried out for the production of a few other ^{11}C-precursors. The discharge in a mixture of $^{11}CO_2$, H_2S and Ar was found to give efficiently $^{11}CS_2$. This compound can be used widely for the syntheses of sulfur-containning ^{11}C-compounds.
Typical radio-chromatograms for $H^{11}CN$ and $^{11}CS_2$ are shown in Fig.2.

The formation processes of the products will be also discussed.

The authors are grateful to Dr. M. Iioh, the chief of the radiological section of National Hospital of Nakano, for the use of cyclotron.

(1) Christman D.R., Finn R.D., Karlstrom K.I. and Wolf A.P., Int. J. appl. Radiat. Isotopes, 26, 435 (1975).
(2) Clark J.C. and Buckingham P.D., "Short-lived Radioactive Gases for Clinical Use", Butterworths, London, 1975.
(3) Takahashi S., Nippon Kagaku Zashi, 81, 987 (1960).

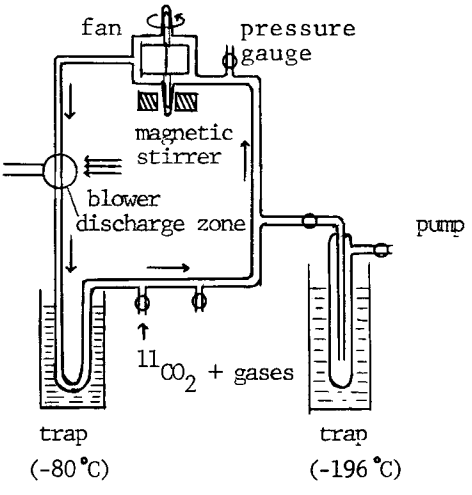


Fig.1. Reaction Vessel

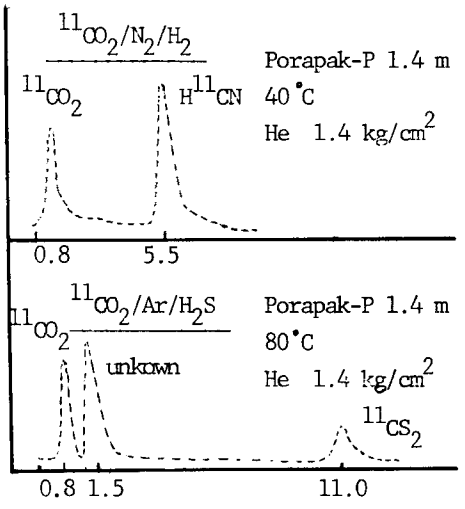


Fig. 2 Radio-gas-chromatograms of reaction products

Tab.1 Product Distribution from $^{11}CO_2$ in a Microwave Discharge in N_2– H_2 and Ar– H_2S Systems*

carrier gases (H_2/N_2)	discharge time(min.)	reaction pressure (torr)	product (%) remaining $^{11}CO_2$	$H^{11}CN$	volatile fraction at -196°C	solid fraction
	5	20	37.4	15.5	43.9	3.2
3/1	10	20	19.8	22.6	42.1	15.5
	15	20	14.0	39.0	48.0	0
2/1	5	20	30.0	17.0	37.0	16.0
	10	20	33.0	40.0	27.0	0
1/3	15	20	24.5	7.5	47.0	21.0
1/1	15	20	32.6	32.4	27.8	7.2
3/1	15	20	14.0	39.0	48.0	0
10/1	15	12	25.7	31.0	15.7	27.6
50/1	15	6.7	21.0	16.0	9.2	53.8
(Ar/H_2S)**			$^{11}CS_2$			
1/5.4			16.6	12.0	26.4	26.2

* : Incident power, 50 W for N_2– H_2, and 43 W for Ar – H_2 systems

** : The percent yield of unknown product in the Ar– H_2 systems was 18.4%

IN-TARGET PRODUCTION OF ^{11}C-PRECURSORS IN NITROGEN CONTAINING SOLIDS

K. Rössler, H. Lattke, C. Mathias, L.M. Al-Shukri, and M. Vogt.
Institut für Chemie 1 (Nuklearchemie) der Kernforschungsanlage Jülich GmbH, D-5170 Jülich, FRG.

Previous studies on the in-target production of ^{11}C-synthesis precursors via the $^{14}N(p,\alpha)^{11}C$ nuclear reaction in solid ammonium halides (NH_4X; X = Cl, Br, I) (1,2) have been extended to a variety of nitrogen containing compounds: NH_4F, $Co(NH_3)_6Cl_3$, $LiNH_2$, frozen NH_3 (77 K), and methyl substituted ammonium chlorides such as CH_3NH_3Cl, $(CH_3)_2NH_2Cl$, $(CH_3)_3NHCl$ and $(CH_3)_4NCl$. A dose range of 13 MeV protons from 10^{-2} to 10^{2} eV/molecule was studied in order to elucidate the mechanisms of primary reactions as well as the conditions for production of ^{11}C-precursors with high radioactivities. A 2π water-cooled target array was used. For experiments with frozen NH_3, a special liquid nitrogen cooled cryostat was applied.

The kind of ^{11}C-products formed via nuclear recoil was similar to that obtained in the systems studied previously; however, the radiochemical yields of the individual products and their dose dependence differ significantly. Besides minor amounts of $^{11}CO_2$, $^{11}CH_4$, halogen derivatives of ^{11}C-methane, ^{11}C-formiate and $^{11}CN^-$, the main products were: ^{11}C-methylamine, ^{11}C-formamidine, ^{11}C-cyanamide, and ^{11}C-guanidine. In $Co(NH_3)_6Cl_3$ labeled complex ions, such as $[Co(NH_3)_5{}^{11}CH_3NH_3]^{3+}$, are also formed with radiochemical yields up to 45 %. The methyl substituted ammonium chlorides showed chain elongation and increased substitution of hydrogen by $^{11}CH_3$-groups. However, at higher doses (> 10 eV/molecule) the radiolysis and polymerization of matrix material and products prevented production of ^{11}C-precursors with good yields.

The maximum radiochemical yields of ^{11}C-precursors obtained by a 5 μAcm^{-2} 13 MeV proton beam are reported in Table 1. The N/H ratio is calculated from the number of nitrogen over that of hydrogen atoms which are bound directly to the ^{11}C atom, averaged over all products. Thus, it represents the ability of ^{11}C to build up larger molecules by reaction with matrix constituents. According to the N/H ratio the following sequence of target materials is obtained: $NH_4Cl < NH_4F < NH_3$ (77 K) $< Co(NH_3)_6Cl_3 < NH_4Br < NH_4I < LiNH_2$.

Even if the formation of larger nitrogen containing molecules is easier in NH_4I, frozen NH_3 provides an interesting target material for practical precursor production, since the target material and volatile ^{11}C-products can be evaporated, whereas the heavier ^{11}C-products are adsorbed on the walls and can easily be eluted by rinsing the target chamber with water solution. In older, relatively low-dose hot-atom experiments (3), the major ^{11}C-products were methylamine and methane. About 4 g of frozen NH_3 irradiated with 30 μAcm^{-2} gave rise to several 100 mCi of total ^{11}C-radioactivity corresponding to some 10 mCi of ^{11}C-cyanamide, ^{11}C-formamidine and ^{11}C-guanidine. Compared to the irradiation of liquid NH_3 (4,5), the solid target provides the advantage of a higher pressure inside the chamber and a greater (dynamical) radiation stability due to more efficient radical recombination processes.

(1) Rössler K., Vogt M. and Stöcklin G., J. Lab. Comp. Radiopharm., 18, 190 (1981).
(2) Vogt M., Report Jül-1960, KFA Jülich, FRG (1980).
(3) Cacace F. and Wolf A.P., J. Amer. Chem. Soc., 87, 5301 (1965).

(4) Iwata R., Ido T. and Tominaga T., J. Lab. Comp. Radiopharm., 18, 187 (1981).
(5) Iwata R., Ido T. and Tominaga T., Int. J. appl. Radiat. Isotopes, 32, 303 (1981).

Table 1. Maximum Radiochemical Yields of Some ^{11}C-Synthesis Precursors Formed in a 5 μAcm^{-2} 13 MeV Proton Beam.

Target	% Radiochem. Yield				
	Methyl-amine	Formamidine	Cyanamide	Guanidine	N/H Ratio
NH_4Cl	80	10	0	0	0.4
NH_4F	70	10	30	0	0.7
$NH_3(s)$*	60	20	10	20	0.9
$Co(NH_3)_6Cl_3$**	5	10	10	50	1.5
NH_4Br	35	20	45	35	2.1
NH_4I	30	15	10	65	2.6
$LiNH_2$	5	5	80	0	8.8

* also up to 10 % $^{11}CH_4$

** also up to 45 % ^{11}C-labeled complex compounds

A CF_4-H_2-Ne GAS TARGET FOR REPRODUCIBLE HIGH YIELDS OF ANHYDROUS $H^{18}F$

R.A. Ferrieri, R.R. MacGregor, S. Rosenthal D.J. Schlyer, J.S. Fowler, and A.P. Wolf.
Department of Chemistry, Brookhaven National Laboratory, Upton, NY 11973.

Anhydrous $H^{18}F$ is a potentially useful precursor due to its synthetic versatility and the possibility of its production at very high specific activity. Various $H^{18}F$ targets have been described utilizing the $^{20}Ne(d,\alpha)^{18}F$ reaction on a mixture of neon: hydrogen (1-5). However, near quantitative recovery or even reproducibility of poor recoveries of NCA $H^{18}F$ generated from these targets have not been achieved. It is therefore advantageous to develop a production system which would allow the maximization of both production and recovery of the anhydrous $H^{18}F$.

The effect of carrier HF in the target gas during irradiation (4.4% HF: 95.6% Ne) is apparent from the 100% theoretical yield of recovered $H^{18}F$ without post-irradiation heating of the target. However, the obvious problems of handling tank HF as well as the potential introduction of unacceptable quantities of carrier make this method unsatisfactory in routine $H^{18}F$ production. The handling of tank HF can be avoided by a novel approach where HF is radiolytically generated in situ from the deuteron bombardment of target gas mixtures containing $CF_4/H_2/Ne$. The mechanism of HF formation is through radiolytic decomposition of CF_4 to $[CF_n]$ radical species and F-atoms. H-abstraction by the F-atoms produces the unlabeled HF. The $[CF_n]$ species are either lost through surface polymerization or stabilize through H-abstraction to produce trace fluoromethanes (CF_3H, CF_2H_2 and CFH_3). The important hot atom processes observed include F- and H-substitution reactions on CF_4 and trace $CF_nH_{(4-n)}$ to produce $CF_3{}^{18}F$, $CF_2{}^{18}FH$ and $CF{}^{18}FH_2$ in a relative distribution of 1.0:0.6:0.2 at a dose of 0.84 μA-hr and H-abstraction from H_2 to produce $H^{18}F$. F-abstraction to produce ^{18}F-F_2 was insignificant.

A plot of the relative distribution of ^{18}F-fluoromethanes and $H^{18}F$ as a function of H_2:CF_4 mole fractions (see Figure) indicates conditions for optimized $H^{18}F$ formation when the H_2:CF_4 ratio was > 3. These results were based on the recovered $H^{18}F$ yield from an unheated target. Using this information, the total amount of H_2:CF_4 was varied relative to neon (while maintaining at least a 3:1 ratio, respectively), and the total dose varied for a constant amount of H_2:CF_4 to determine these effects on the recovered yields of ^{18}F-labelled and radiolytic HF. The results presented in the Table can be summarized as follows: (i) the recovered yield of $H^{18}F$ increases and plateaus at about 43% total ^{18}F when the amount of H_2:CF_4 exceeds 2% of the target gas; (ii) the radiolytic yield of HF surprisingly decreases from essentially 100% to a level of about 20% at the same 2% break-point in the gas mixture; and (iii) a x10 increase in applied dose resulted in a x2.8 increase in the radiolytic HF yield, but with very little change in the recovery of $H^{18}F$.

These results show that while reproducibly high $H^{18}F$ yields are obtainable from this target, the specific activity is poor. However, some optimization of this is possible by controlling the radiolysis through variation of gas composition and applied dose.

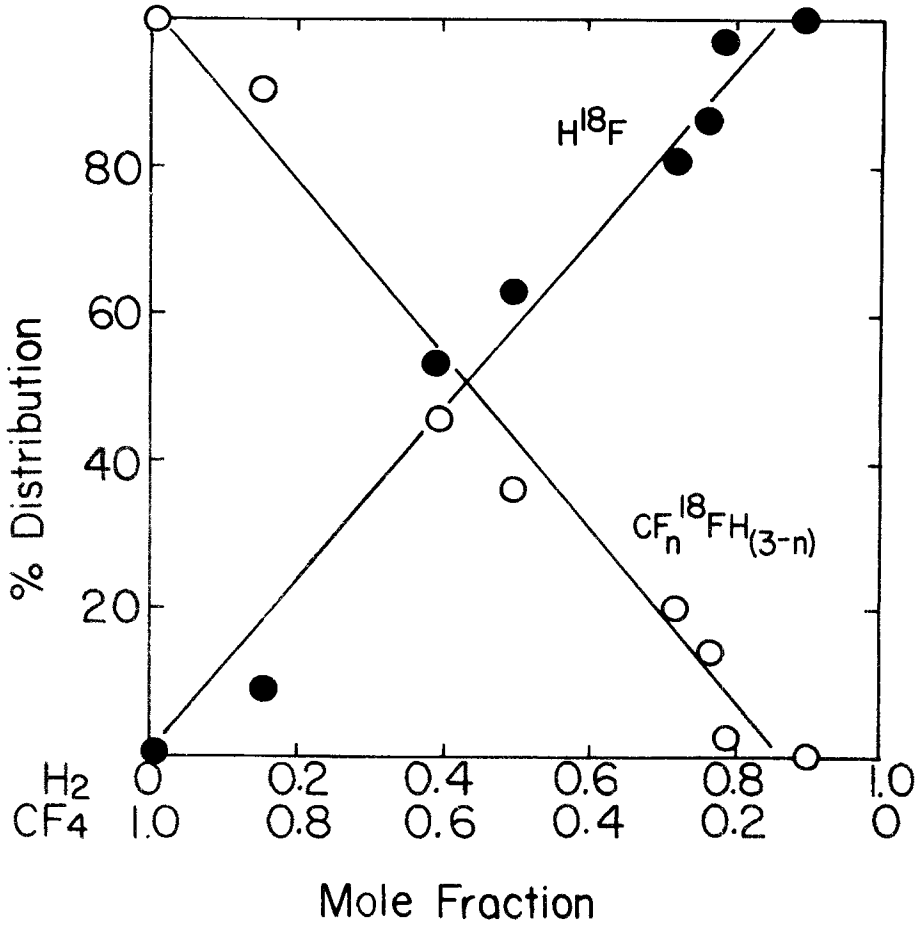
This research was carried out at Brookhaven National Laboratory under contract with the U. S. Department of Energy and supported by its Office of Basic Energy Sciences and also by the Office of Health and Environmental Research. The authors wish to acknowledge the assistance of Elinor Norton in the yield measurements of HF.

(1) Winchell, H.S., Wells, D.K., Lamb, J.F. and Beaudry, S.B., U.S. Patent 3, 981, 769 (1976).
(2) Straatmann, M.G. and Welch, M.J., J. Nucl. Med., 18, 151 (1977).
(3) Lambrecht, R.M., Neirinckx, R. and Wolf, A.P., Int. J. Appl. Radiat. Isot., 29, 175 (1978).
(4) Ehrenkaufer, R.L.E., Fowler, J.S., MacGregor, R.R., Ruth, T.J., Wieland, B. and Wolf, A.P., "Production of Carrier-Free, Anhydrous $H^{18}F$ by Deuteron Irradiation of H_2/Ne Target Gas." Part I, 2nd Chemical Congress of the North American Continent, San Francisco, California, ISBNO-8412-0583-3. (Full manuscript in preparation).
(5) Dahl, J.R., Lee, R., Schmall, B. and Bigler, R. E., J. Label. Cmpds. Radiopharm., XVIII, 34 (1981).

Table of the ^{18}F-Labelled and Radiolytic HF Yields From Varied CF_4:H_2 Amounts and Varied Applied Dose

Gas Run	Composition (mmol)[a] CF_4	H_2	Applied Dose (μA-hr)[b]	Radiolytic Amount of HF (mmol)	% Recovery of F as HF[d]	$H^{18}F$ Yield % Total ^{18}F
1	0.02	0.08	0.84	0.08	100	10
2	0.16	0.48	0.84	0.35	55	29
3	0.47	1.18	0.84	0.36	19	44
4[c]	1.16	3.56	0.84	1.18	25	42
5	1.16	3.56	0.14	0.80	17	49
6	1.16	3.56	1.40	2.20	47	53

[a] Total target pressure maintained at 380 psia.
[b] Doserate was maintained at 10 μA for all runs.
[c] Run 4 was an average of 5 separate runs.
[d] % recovery of F as HF equals 100 x [(mmol HF)/(4 x mmol CF_4)].



PLOT OF THE RELATIVE DISTRIBUTION OF $H^{18}F$(●) AND ^{18}F-LABELLED FLUOROMETHANES (O) AS A FUNCTION OF H_2:CF_4 MOLE FRACTIONS

SYNTHESES OF RADIOBROMINATED 2-DEOXY-2-BROMO-D-GLUCOSE AND 2-DEOXY-2-BROMO-D-MANNOSE FOR THE MEASUREMENT OF CEREBRAL GLUCOSE METABOLISM IN VIVO

Y.-G. Zhou, C.-Y. Shiue, and A.P. Wolf.
Department of Chemistry, Brookhaven National Laboratory, Upton, NY 11973.

The glucose analogs, 2-deoxy-2-[^{18}F]fluoro-D-glucose (1-4), 3-deoxy-3-[^{18}F]fluoro-D-glucose (5) and 2-deoxy-D-[1-^{11}C]glucose (6) have been used as tracers for the measurement of cerebral glucose metabolism under different pathological states in humans non-invasively (7-10). 2-Deoxy-2-[^{18}F]fluoro-D-glucose (2-^{18}FDG), in particular, has generated wide interest in the biomedical community and is now produced regularly in the centers around the world for use in clinical studies. Nevertheless, the relatively low yield (~ 20%) of this radiopharmaceutical has imposed a limitation on the capabilities for many centers to synthesize sufficient 2-^{18}FDG for their own needs. Therefore, we have searched for methods which will improve the yield of 2-^{18}FDG (11), or analogs which will behave like 2-^{18}FDG. In extending the availability of radiopharmaceuticals such as 2-^{18}FDG, we have synthesized 2-deoxy-2-[^{82}Br]bromo-D-glucose (5) and 2-deoxy-2-[^{82}Br]bromo-D-mannose (6) from bromine chloride (1).

Unlabeled 2-deoxy-2-bromo-D-glucose (5) and 2-deoxy-2-bromo-D-mannose (6) have been synthesized by the reaction of 3,4,6-tri-O-acetyl-D-glucal (2) with N-bromosuccinimide and hydrogen fluoride, followed by acid hydrolysis (12,13). Bromination of 2 with bromine gave a mixture of 2-deoxy-2-bromo-3,4,6-tri-O-acetyl-α-D-glucopyranosyl bromide and 2-deoxy-2-bromo-3,4,6-tri-O-acetyl-α-D-mannopyranosyl bromide in a ratio of 2:1 (14). However, these methods would entail the inherent lose of 50% of the bromine activity as that for the synthesis of 2-^{18}FDG from 2 and [^{18}F]F_2. It is therefore advantageous to synthesize compounds 5 and 6 from "Br^+".

Bromine chloride (BrCl) has been prepared by condensing chlorine gas into liquid bromine (15). However, this method is not suitable for the radiopharmaceutical synthesis because of the difficulty in quantitating the amounts of each halide added. We have synthesized ^{82}Br-labeled bromine chloride *in situ* by oxidation of ^{82}Br-bromide with chloramine-T (16) or N-chlorosuccinimide (17).

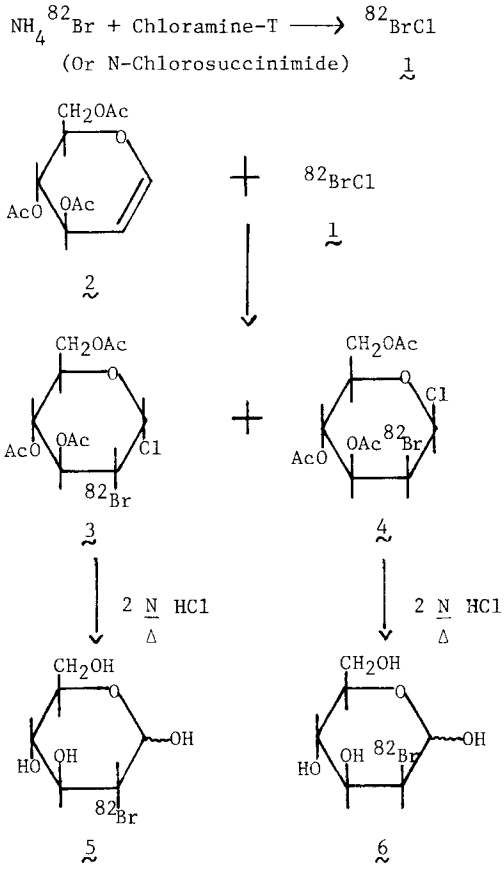
Reaction of 3,4,6-tri-O-acetyl-D-glucal (2) with ^{82}Br-BrCl (1) in THF at room temperature gave 2-deoxy-2-[^{82}Br]bromo-3,4,6-tri-O-acetyl-α-D-glucopyranosyl chloride (3) and 2-deoxy-2-[^{82}Br]bromo-3,4,6-tri-O-acetyl-β-D-mannopyranosyl chloride (4). Compounds 3 and 4 were separated by silica gel column (ether-hexane, 1:1). The ratio of 3 to 4 was ~ 3:1 and the overall radiochemical yield was ~ 70% (based on ^{82}Br activity). Hydrolysis of 3 and 4 with 2 N HCl followed by silica gel column purification gave 5 and 6 respectively (Scheme 1). The identities of 5 and 6 were confirmed by comparing the glc retention times of their silylated derivatives with authentic samples (12).

In summary, we report here the syntheses of 2-deoxy-2-[^{82}Br]bromo-D-glucose and 2-deoxy-2-[^{82}Br]bromo-D-mannose from ^{82}Br-bromine chloride without the inherent lose of 50% of the bromine activity which will occur with the use of $^{82}Br_2$. This method can readily be adapted for the syntheses of ^{75}Br-2-BrDG and ^{75}Br-2-BrDM (a positron emitter,

$t_{1/2}$ = 98 min) and use for the measurement of cerebral glucose metabolism *in vivo* by positron emission tomography.

This research was carried out at Brookhaven National Laboratory under contract with the U. S. Department of Energy and the Office of Health and Environmental Research.

(1) Ido, T., Wan, C.-N., Casella, V., et al., J. Label Cmpds. Radiopharm., 14, 175-183 (1978).
(2) Fowler, J.S., MacGregor, R.R., Wolf, A.P., et al., J. Nucl. Med., 22, 376-380 (1981).
(3) Barrio, J.R., MacDonald, N.S., Robinson, G.D., et al., J. Nucl. Med. 22, 372-375 (1981).
(4) Shaughnessy, W.J., Gatley, S.J., Hickwa, R.D., Int. J. Appl. Radiat. Isot. 32, 23-29 (1981).
(5) Tewson, T.J., Welch, M.J., Raichle, M.E., J. Nucl. Med. 19, 1339-1345 (1978).
(6) MacGregor, R.R., Fowler, J.S., Wolf, A.P., et al., J. Nucl. Med. 22, 800-803 (1981).
(7) Reivich, M., Kuhl, D., Wolf, A.P., et al., Circ. Res. 44, 127 (1979).
(8) Greenberg, J., Reivich, M., Alavi, A., et al., Science 212, 678 (1981).
(9) Phelps, M., Kuhl, D., Mazziotta, J.C., Science 211, 1445 (1981).
(10) Alavi, A., Reivich, M., Greenberg, J., et al., Sem. Nucl. Med. 11, 24 (1981).
(11) Shiue, C.-Y., Salvadori, P.A., Wolf, A.P., et al., manuscript in preparation.
(12) Kent, P.W., Robson, F.O., Welch, V.A., J. Chem. Soc. 3273 (1963).
(13) Kent, P.W., Barnett, J.E., J. Chem. Soc. 6196 (1964).
(14) Lemieux, R.U., Fraser-Reid, B., Can. J. Chem. 42, 532 (1964).
(15) de la Mare, P.B.D., "Electrophilic Halogenation", Cambridge University Press: New York, 1976, p. 23.
(16) Kabalka, G.W., Sastry, K., Hsu, H., et al., J. Org. Chem. 46, 3113 (1981).
(17) Wilbur, D., Anderson, K., J. Org. Chem. 47, 358 (1982).



Scheme 1 Syntheses of ^{82}Br-2-Deoxy-2-Bromo-D-Glucose and ^{82}Br-2-Deoxy-2-Bromo-D-Mannose

^{75}Br- AND ^{123}I-ANALOGUES OF D-GLUCOSE AS POTENTIAL RADIOPHARMACEUTICALS

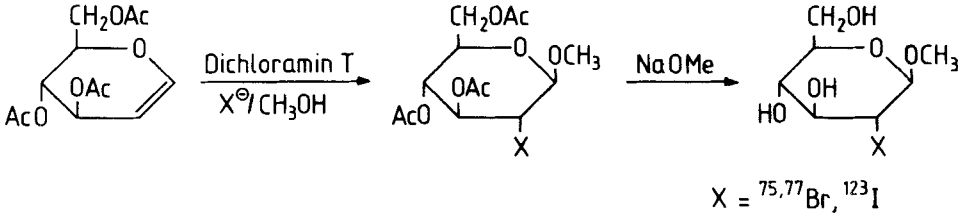
G. Kloster, P. Laufer, W. Wutz, and G. Stöcklin.
Institut für Chemie 1 (Nuklearchemie), Kernforschungsanlage Jülich GmbH, D-5170 Jülich, FRG.

Sugar analogues are potential radiopharmaceuticals for investigations of brain and heart glucose utilization. Apart from ^{11}C-D-glucose (1), 2-^{18}F-fluorodeoxy-D-glucose (2), 3-^{18}F-fluorodeoxy-D-glucose (3) and 3-^{11}C-methyl-D-glucose (4) have been investigated as glucose tracers up to now. Since both ^{11}C and ^{18}F pose some synthetic problems due to their half-lives or their chemical reactivity, we chose to prepare some ^{123}I- or ^{75}Br-labelled D-glucose analogues and study their biodistributions in order to find analogues that may also be used by institutions other than those having a cyclotron. At this time, only 3-deoxy-3-iodo-D-glucose (5) has been prepared, but no biodistribution data have been reported. An unsuccessful attempt to prepare 2-deoxy-2-iodo-D-glucose (6) was also reported.

We synthetized ^{123}I-3-deoxy-3-iodo-D-glucose (3-IDG) and ^{75,77}Br-3-bromo-3-deoxy-D-glucose (3-BDG) from the commercially available 1,2:5,6-diisopropylidene-D-allofuranose via its triflate and the corresponding halide. After hydrolysis of the ketals and chromatographic separation of the product, both compounds were obtained in 10 % overall radiochemical yield in a reaction time of 2 h.

Biodistribution in animals showed that 3-BDG and 3-IDG did not accumulate significantly in the brain (maximum uptake: 38 % mean body concentration [% MBC] for 3-IDG and 50 % MBC for 3-BDG). Accumulation in the heart was higher (> 200 % MBC for each), but activity levels in blood and lung were still higher. Thus, IDG and BDG do not seem to be promising tracers of D-glucose.

We attempted to produce ^{123}I-2-deoxy-2-iodo-D-glucose (2-IDG) and ^{75,77}Br-2-bromo-2-deoxy-D-glucose from tri-O-acetyl-D-glucal using pathways described in the literature for the inactive compounds (7). All of our attempts to isolate either 2-IDG or 2-BDG failed completely.



During one of these attempts, we produced ^{123}I-ß-methyl-2-deoxy-2-iodo-D-glucoside (MIDG) and ^{77}Br-ß-methyl-2-bromo-2-deoxy-D-glucoside (MBDG). The overall yield of these compounds from tri-O-acetyl-D-glucal and XCl generated from $X^{\ominus}$ and dichloramin-T (8) was 20 % for MIDG and 15 % for MBDG. Both compounds cochromatographed with standards prepared according to Lemieux and Fraser-Reid (9). As these compounds have the all-trans, all-equatorial structure required for transport on the hexose carrier, they may be tracers for D-glucose, even though they are not expected to be substrates for hexokinase.

Biodistribution studies in animals showed that MBDG and MIDG are also not significantly accumulated in the brain (maximum uptake: 28 % MBC for MIDG and 52 % for MBDG). Accumulation in the heart was also higher for both (around 200 % MBC), with heart-to-blood and heart-to-lung ratios around 1. Thus, MIDG and MBDG also do not seem to be promising tracers of D-glucose.

More promising results were obtained with the intermediate triacetyl derivatives of MIDG and MBDG, namely ^{123}I-methyl-2-deoxy-2-iodo-tri-O-acetyl-ß-D-glucopyranoside (MITG) and ^{75}Br-methyl-2-deoxy-2-bromo-tri-O-acetyl-ß-D-glucopyranoside (MBTG). Both compounds were prepared in 35-40 % radiochemical yield after less than 1.5 h synthesis time including chromatographic separation.

Biodistribution data in mice showed (Fig. 1) that both MITG and MBTG rapidly enter the brain, reaching 100 % MBC for MITG 1 min after application and 160 % MBC for MBTG 0.25 min after application. At the same time the blood concentration reached 190 % MBC. For MBTG, the brain-to-blood concentration ratio stayed fairly constant at about 0.8 during the first 10 min, while the absolute concentration fell to 106 % MBC at 10 min. Large amounts of radioactivity were observed in the kidneys, indicating possible renal excretion. Radiochemical analysis of the brain showed that MBTG is not altered in the brain. Nothing is known about the mechanism by which MBTG enters the brain. If it were a substrate for the hexose carrier of the blood brain barrier, MBTG would be a promising alternative to 3-^{11}C-methyl-D-glucose (4,10) as a tracer for hexose transport.

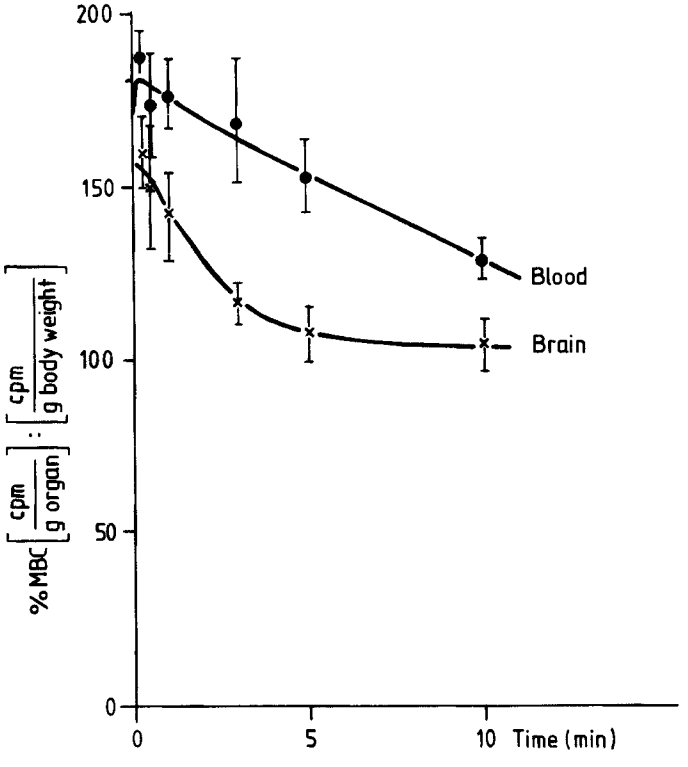


Fig. 1 Radioactivity concentration in blood and brain after i.v. injection of MBTG.

(1) Raichle M.E., Welch M.J., Grubb R.L. Jr., Higgins C.S., Ter-Pogossian M.M. and Larson K.B., Science, 199, 986 (1978).
(2) Gallagher B.M., Ansari A., Atkins H., Casella V, Christman D.R., Fowler J.S., Ido T., MacGregor R.R., Som P., Wan C.-N., Wolf A.P., Kuhl D.E. and Reivich M., J. Nucl. Med., 18, 990 (1977).
(3) Tewson T.J., Welch M.J. and Raichle M.E., J. Nucl. Med., 19, 1339 (1978).
(4) Kloster G., Müller-Platz C.M. and Laufer P., J. Lab. Comp. Radiopharm., 18, 855 (1981).
(5) Tsuya A. and Shigematsu A., Patent Application 28 17 336 (Fed.Rep.Germany) 1979.
(6) Fowler J.S., Lade R.E., MacGregor R.R., Shiue C., Wan C.-N. and Wolf A.P., J. Lab. Comp. Radiopharm., 16, 7 (1979).
(7) Honda S. and Takiura K., Carbohydr. Res., 34, 45 (1974).
(8) Coenen H.H., Petzold G. and Stöcklin G., this meeting, see also He Youfeng, Coenen H.H., Petzold G. and Stöcklin G., J. Lab. Comp. Radiopharm., in press.
(9) Lemieux R.U. and Fraser-Reid B., Can. J. Chem., 42, 532 (1964).
(10) Kloster G., Stöcklin G., Vyska K., Freundlieb C., Höck A., Feinendegen L.E., Traupe H. and Heiss W.-D., in: P.H. Cox (ed.) Progress in Radiopharmacology, Vol. 3, Nijhoff, Den Haag, in press.

AN ALTERNATIVE SYNTHESIS OF 2-DEOXY-2-FLUORO GLUCOSE

T.J. Tewson. Cardiology Department, University of Texas Health Science Center at Houston, Houston, Texas, USA 77030.

The current synthesis of fluorine-18 2-deoxy-2-fluoro glucose(1), based upon ^{18}F-F_2, is inherently limited to a maximum possible yield of 50%, based upon fluorine-18, as only one of the two fluorine atoms in the reagent is utilized in the product. A synthesis based upon fluoride ion would be advantageous as, in principle, all the fluorine-18 produced could be incorporated into the final product. However, nucleophillic displacement at the two position of carbohydrates is a notoriously intractable procedure(2) because of competing re-arrangement and elimination reactions. The use of cyclic leaving groups such as sulfite(3) and sulfate esters have potential to overcome these problems as the energy of the transitions states for the non-productive reactions should be prohibitive. Although these compounds have the potential for substitution at each end of the ring, sterochemical requirements of the transition state and the product, i.e. linear relationship of the incoming fluoride to the departing oxygen and the least stearic interactions of the product sulfate should lead to largely or exclusively substitution at one of the two possible carbon atoms.

The 2,3-sulfite and sulfate esters of 4,6-benzylidene α-and β-methyl-mannopyranoside were prepared and reacted with tetramethylammonium fluoride. Both sulfite esters gave low yields of fluorinated materials, the major reaction being attack at sulfur and the α-methyl sulfate gave a fragmentation product which had lost the 1-O methyl group. However, the 4,6-benzylidene-1-O-β-methylmannopyranoside reacts rapidly(25 mins at room temperature) in acetonitrile to give 4,6-benzylidene-2-deoxy-2-fluoro-1-O-β-methyl-3-O-sulfate-gluco-pyranoside as the major product, by HPLC.

Treatment of the reaction mixture with acetone/trifluoroacetic acid gives 4,6-benzylidene-2-deoxy-2-fluoro-1-O-β-methyl-glucopyranoside 94% pure by gas chromatography.

Two other benzylidene-fluoro-hexopyranosides, amounting to 6% of the product can be detected by G.C./M.S. but it is not yet clear whether these are products of fluorination reaction or produced during the removal of the sulfate. Strong acid hydrolysis of the reaction mixture removes all the protecting groups to give 2-deoxy-2-fluoroglucose, apparently pure, although the sensitivity of detection is much lower at the stage.

The synthesis will be performed using fluorine-18 to determine if the high yield and simplicity of the procedure are duplicated when working with the radionuclide.

(1) Ido T. et al., Journal of Labelled Cmpds and Radiopharmaceuticals 14, 175 (1978).
(2) Pengalis A.E., Advances in Carbohydrate Chemistry and Biochemistry, 38, 195 (1981).
(3) Tewson T.J., Welch M.J., J. Nucl. Med. 21, 559 (1980).

Radioactivity Labeling of C-1 Position of Glucose and Mannose —— Synthetic Methods, Metabolism, and Application of the Compounds for in vivo Determination of Regional Glucose Utilization Rate in the Brain and for Detection of Tumors in the Body

T. Hara and T. Nozaki.
National Nakano Chest Hospital, 3-Egota Nakanoku Tokyo, Japan.

^{11}C-labeled glucose and mannose are thought to be useful radiopharmaceuticals for physiological study and diagnosis of diseases. The ^{11}C-labeling of C-1 position of the aldoses is possible by chain extension of the next lower sugar, arabinose, via the Kiliani-Fischer cyanohydrin synthesis employing sodium ^{11}C-cyanide. As a result of chirality of the starting aldose, unequal amounts of diastereomeric aldonitriles are produced, the ratio being dependent on the pH. At neutral pH (7.5-8.0) aldonitriles are rather stable and fit for catalytic reduction with H_2 to give the desired aldoses directly (glucose $\leq$ mannose)[1] (Method 1). At alkaline pH (11.5-12.0) the initially produced epimeric aldonitriles are hydrolyzed in situ to the corresponding aldonates. Separation of the epimeric aldonates, lactonization, and reduction afford the final object (glucose $\geq$ mannose)[2] (Method 2). We examined the optimal condition for synthesis of glucose-mannose mixture using $Na^{14}CN$ in regard to the time requirement, radiochemical yield, and radiochemical purity (Tables 1 and 2). The whole procedure was finished in 30 min in both methods. Method 1 was found simpler and more convenient for automatic processing in spite of the lower yield (30 %) as compared with Method 2 (45 %). We already have an automatic $H^{11}CN$ producing system, and it will be connected to an automatic aldose synthetic system which is now under construction and nearing completion. A whole system will further include a high-performance liquid chromatography for separating glucose and mannose in the form of sugar-borate complexes, from which boric acid is easily removed as volatile methylborate.

It is assumable that glucose and mannose are utilized in the body chiefly as the energy source and finally exhaled in breath as CO_2. The 1-position carbon (as well as C-6) of glucose and mannose stays in the body for a longer period than the other carbons of the same compounds (C-1 and C-6 are converted to CO_2 after they turn twice the TCA cycle). This fact may facilitate a steady accumulation of radioactivity for a sufficiently long duration in some organ after i.v. bolus injection of the C-1 labeled glucose and mannose. This is an absolute requisite for the measurement of regional glucose (or mannose) utilization rate with ^{11}C and a positron CT, where the initial velocity of uptake should be determined. Figs.1 and 2 show the time course of ^{14}C distribution we determined in normal awake rats after i.v. injection of glucose-1-^{14}C and mannose-1-^{14}C. The brain, both the gray matter and the white matter, exhibited the above-mentioned steady accumulation of radioactivity. The "normalized ^{14}C concentration" means

$$\frac{^{14}C \text{ dose in a particular tissue}}{\text{wet weight of the tissue}} \Big/ \frac{\text{injected } ^{14}C \text{ dose}}{\text{body weight}} .$$

At the peak time of accumulation after injection of both compounds, brains of rats were freeze-blown and extracted into perchloroacetic acid-alcohol at -20°. Amino acid analyses revealed that more than 80 % of the brain activity was associated with glucogenic amino acids (glutamate, aspartate, and glutamine) which are known to constitute large metabolic pockets of TCA cycle in the brain[3,4]. Figs.3 and 4 show the result with tumor-bearing (Walker 256 carcinoma) rats. The tumor tissue presented also an initial gradual uptake of radioactivity. This tumor-specific characteristic (the delay of peak-time) may be visualized with ^{11}C in a scintillation camera after an appropriate data processing.

(1) Isbell H.S., Karabinos J.V., Fresh H.L., Holt N.B., Schwebel A. and Galkowski T.T., J. Res. Nat. Bureau Standards 48, 163 (1952).
(2) Serianni A.S., Nunez H.A. and Barker R., Carbohyd. Res. 72, 71 (1979).
(3) Busch H., Fujiwara E. and Keer L.M., Cancer Res. 20, 50 (1960).
(4) Van den Berg C.J., Kržalić LJ., Mela P. and Waelsch H., Biochem. J., 113, 281 (1969).

Table 1 (Method 1)

$K^{14}CN$ + NaCN 25mg in 4ml of water.
+ Arabinose 150mg in 1ml of water.
+ 0.25M acetic acid to pH 8.0 (pH stat); 25°, 10 min.
+ 0.25M acetic acid to pH 4.2 (pH stat).
Transfer the reaction solution to an autoclave containing 5% palladium-barium sulfate 500mg in 5ml of water previously exposed to H_2 at 7kg/cm^2, 25°, 10 min.
+ H_2 at 7kg/cm^2; 50°, 10 min.
Filtration.
Vacuum evaporation and dissolve in water.

Table 2 (Method 2)

$K^{14}CN$ + arabinose 400mg in 0.05M NaOH 2ml.
Dowex-1 chromatography: water-wash then elute with 5M acetic acid.
Vacuum evaporation to complete dryness.
+ Gluconolactone 10mg + trifluoroacetic acid 5ml; 70°, 5 min.
Vacuum evaporation.
Dissolve in 3ml of water.
+ 5% Na(Hg) 500mg twice keeping pH at lower than 3.5 with oxalic acid (pH stat).
Neutralize with 1N NaOH.
+ 4 vols of methanol and filtration.
Vacuum evaporation.
Dowex-1 (acetate form) chromatography: elution with water.

Distribution of ^{14}C in normal rats (Figs.1 and 2) or tumor-bearing rats (Figs.3 and 4) after i.v. injection of glucose-1-^{14}C or mannose-1-^{14}C.

Fig.1

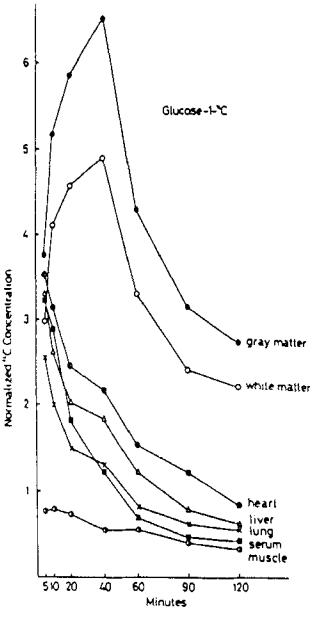
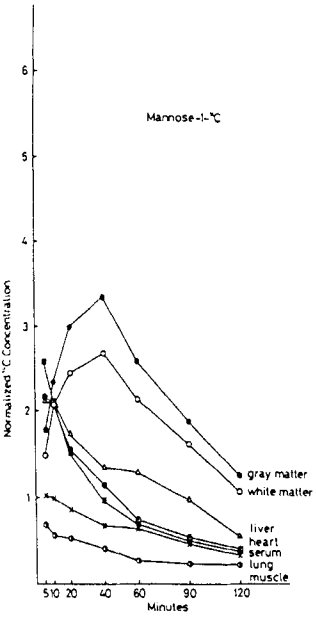
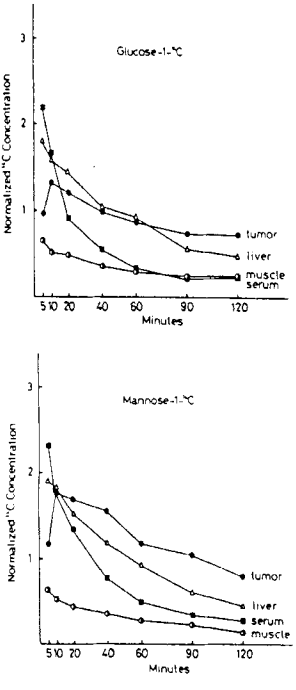


Fig.2



Figs.3 and 4



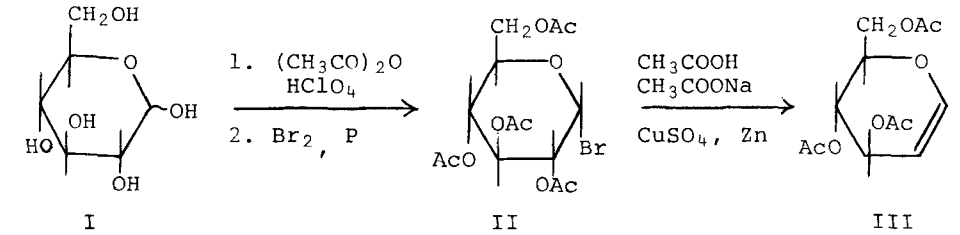
BLOOD-BRAIN TRANSPORT OF NEW GLUCOSE ANALOGS AND THEIR EFFECT ON YEAST HEXOKINASE

Yoshio Homma, Yuko Murase, Michiko Ishi, and Yukio Murakami.
Laboratory for Radiopharmaceutical Chemistry, Kyoritsu College of Pharmacy, Minato-ku, Tokyo, 105 Japan.

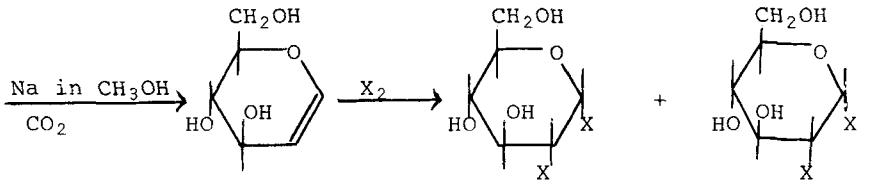
D-Glucal, a kind of unsaturated sugar derivatives derived from D-glucose, contains a double bond which lies between carbon atoms 1 and 2, therefore, it adds two atoms of halogen or hydrogen *etc.* In course of a study concerning the relative permeability of blood-brain barrier (BBB) to a number of sugars including D-glucose and 2-deoxy-D-glucose, we found that the dihalogen addition products D-glucal have reasonably greater brain uptake than D-glucose and 2-deoxy-D-glucose.

The synthetic route adopted for the preparation of the D-glucal dihalides is as follows. Tetra-O-acetyl-α-D-glucopyranosyl bromide (II) was prepared from D-glucose (I) by a method analogous to that described by Bárczai-Martos and Kőrősy (1). The treatment of (II) with zinc dust and $CuSO_4.5H_2O$ in the mixture of glacial acetic acid and sodium acetate gave tri-O-acetyl-D-glucal (III), which was dissolved in dry methanol, and sodium was added. The solution was allowed to remain at room temp. for 24 h, treated with carbon dioxide and evaporated under diminished pressure. The residue was extracted with hot ethyl acetate. The combined extracts were concentrated to afford D-glucal (IV), m.p. 57 - 59°, $[\alpha]_D^{19}$ - 8.0 (c 1.88 in water).

Dihalogen addition products of D-glucal were formed quite readily by the direct addition of halogen to the unsaturated linkage of (IV) within a few minutes in a 98 % yield.



I II III



IV V VI

A mixture was prepared containing 0.07 - 0.10 μCi of [^{82}Br]-D-glucal dibromides, [^{36}Cl]-D-glucal dichlorides, D-[6-^{14}C]glucose and, 2-deoxy-D-[1-^{14}C]glucose and mixed with approximately 1.36 μCi of tritiated water (^{3}HOH), which was used as an internal standard of brain uptake. The left common carotid artery of male Donryu rats was surgically exposed and 0.2 ml of the buffered Ringer solution

containing the radioactive mixture described above was injected within 0.2 sec.

The brain uptake index (BUI) of the D-glucal dihalides was measured and compared with those of D-glucose and 2-deoxy-D-glucose according to the procedure of Oldendorf (2-4). The BUI of [^{82}Br]-D-glucal dibromides was 59.4 ± 2.4 % for the midbrain and 60.1 ± 2.4 % for the cortex. The BUI of [^{36}Cl]-D-glucal dichlorides resulted in the following data : 55.2 ± 2.2 % for the midbrain and 59.5 ± 2.4 % for the cortex. On the other hand, the average BUI of 2-deoxy-D-[1-^{14}C] glucose was 49.4 ± 2.0 % for the midbrain and 49.2 ± 2.0 % for the cortex. Those of D-[6-^{14}C]glucose was 32.9 ± 1.3 % for the midbrain, 33.6 ± 1.3 % for the cortex (Table 1).

Since the halogenated product is a mixture of stereoisomers termed 2-halogeno-2-deoxy-α-D-glucopyranosyl halide(V) and 2-halogeno-2-deoxy-α-D-mannopyranosyl halide(VI), the sum of the BUI for α-D-gluco (V) and α-D-manno(VI) compounds (Table 1) and individual BUI for these stereoisomers were studied.

Investigation with yeast hexokinase, phosphoglucose isomerase and fructo-6-phosphate kinase have shown that D-glucal dihalides is a good substrate for hexokinase but not enter into subsequent metabolic steps of glycolysis. The property has been extremely useful for the quantitative determination of local glucose metabolism in brain.

The halogenation reaction is sufficiently rapid and usable activity from D-glucal dihalides is twice as much as those from equal moles of ^{18}F-2-deoxy-2-fluoro-D-glucose and ^{18}F-3-deoxy-fluoro-D-glucose. D-Glucal can be readily available for months without any detectable chemical alteration.

Table 1. Brain uptake index of radiolabeled hexoses for midbrain and cortex

	BUI %*	
	midbrain	cortex
^{3}HOH reference	100	100
^{82}Br-D-Glucal dibromides	59.4 ± 2.4	60.1 ± 2.4
^{36}Cl-D-Glucal dichlorides	55.2 ± 2.2	59.5 ± 2.4
2-deoxy-D-[1-^{14}C]Glucose	49.4 ± 2.0	49.2 ± 2.0
D-[6-^{14}C]Glucose	32.9 ± 1.3	33.6 ± 1.3

* Brain uptake index value are means + SD. For each mean value n=5 except that for ^{36}Cl-D-glucal dichlorides, n=10.

(1) Bárczai-Martos M. and Kőrösy F., Nature, 165, 369 (1950).
(2) Oldendorf W.H., Brain Res., 24, 372 (1970).
(3) Oldendorf W.H., Am. J. Physiol., 221, 1629 (1971).
(4) Oldendorf W.H., Am. J. Physiol., 224, 1450 (1973).

SYNTHESIS OF ^{18}F-LABELED ACETYL HYPOFLUORITE FOR RADIOTRACER SYNTHESIS

J.S. Fowler, C.-Y. Shiue, A.P. Wolf, P.A. Salvadori, and R.R. MacGregor. Chemistry Department, Brookhaven National Laboratory, Upton, NY 11973.

Acetyl hypofluorite (CH_3CO_2F) is a newly characterized electrophilic fluorination reagent, the synthesis and reactivity of which was recently described by Rozen (1). The unlabeled compound is synthesized from sodium acetate, acetic acid and elemental fluorine in freon-11 at -78°.

$$F_2 + CH_3CO_2Na \xrightarrow[-78^\circ]{CFCl_3\text{-}CH_3CO_2H} CH_3CO_2F + NaF \qquad \text{(Eq. 1)}$$

This reagent appears to be milder and more selective than elemental fluorine. Its reactions with olefins are characterized by syn addition as well as regiospecificity (1). It has also been used in the direct fluorination of activated aromatic rings (2).

We report here the synthesis of ^{18}F-labeled acetyl hypofluorite in ~ 80% yield (based on $[^{18}F]F_2$) by purging the contents of the Ne/F_2 target through a solution of ammonium acetate in acetic acid. Yields were determined and conditions optimized by quenching the oxidant formed with

$$[^{18}F]F_2 + CH_3CO_2NH_4 \xrightarrow{CH_3CO_2H} CH_3CO_2{}^{18}F + NH_4{}^{18}F \qquad \text{(Eq. 2)}$$

KI and titrating the I_2 liberated with standard thiosulfate solution. Factors influencing the yield include efficiency of dispersal of the neon/$[^{18}F]F_2$ gas mixture, solution volume, substrate concentration, nature of the cation, and purge time.

Optimization of yields was carried out by using a known amount of F_2 in neon simulating target conditions. In yield optimization studies, the target gas was purged through a vessel ((0.43 in x 9.0 in) with a teflon frit for dispersal) containing acetic acid solutions of various salts and salt concentrations. Purge time was 23-25 minutes and was not varied except to study the influence of this parameter on yield. The gas exiting the reaction vessel was passed through a solution of 1 M KI with starch indicator to trap any oxidant not retained by the first vessel. At the end of purge the acetic acid solution was added to an excess of aqueous KI with starch indicator. This solution and the KI solution into which the exit gas was purged, were each titrated. The oxidant contained in the acetic acid solution was not F_2 as demonstrated by the addition reactions described below.

As can be seen from Eq. 2, the maximum radiochemical yield of $CH_3CO_2{}^{18}F$ from $[^{18}F]F_2$ is 50% since each molecule of F_2 produces one molecule of fluoride salt in addition to one molecule of acetyl hypofluorite. The 80% chemical yield therefore translates into a maximum radiochemical yield of 40%. The influence of the cation on yield is shown in Table 1 where it can be seen that NH_4^+, K^+ and Cs^+ give significantly greater yields than Na^+. The influence of purge time and salt concentration was studied using sodium as the cation and showed that approximately 23 minutes (flow rate = ~ 70 ml/min) is required to unload the target under these experimental conditions without decreasing yields. The influence of sodium acetate concentration on acetyl hypofluorite yield is shown in Figure 1, where it can be seen that increasing amounts of sodium acetate decreased the acetyl hypofluorite yield and significantly increased the

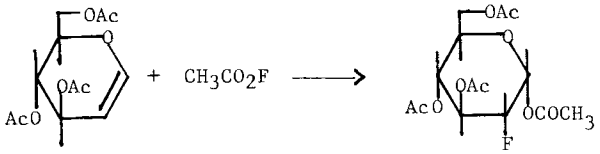
amount of oxidant which could not be accounted for in the acetic acid solution and KI trap.

Table 1. Yield data for CH_3CO_2F synthesis.[a]

Cation[b]	Purge Time (min)	Yield[c] of CH_3CO_2F(%)
Na^+	13	52 (n=1)
Na^+	23	64.0 ± 2.5 (n=4)
Na^+	46	67 (n=1)
NH_4^+[d]	23	78.9 ± 1.6 (n=5)
K^+	23	76.7 ± 1.8 (n=5)
Cs^+	23	71.0 ± 2.5 (n=3)
None	23	43.6 ± 2.4 (n=3)

[a] All runs used a total volume of 15 ml of HOAc.
[b] Cation concentrations varied from 60-150μmol, the yields reported here being optimized in this concentration range.
[c] Theoretical (100%) yield was determined by purging the Ne/F_2 mixture directly into 1M KI solution and titrating the liberated I_2 with standard thiosulfate solution.
[d] NH_4OH (58%).

The identity of the oxidant as acetyl hypofluorite was confirmed by its addition to 3,4,6-tri-O-acetyl-D-glucal to form 2-deoxy-2-fluoro-1,3,4,6-tetra-O-acetyl-D-glucose and to 1,2-difluorostilbene to produce 1,1,2-trifluoro-2-acetoxystilbene.



$C_6H_5CF{=}CFC_6H_5 + CH_3CO_2F \longrightarrow C_6H_5CF_2{-}CFC_6H_5$ (with $OCOCH_3$)

^{18}F-Acetyl hypofluorite is currently being used in our laboratory to produce ^{18}F-2-deoxy-2-fluoro-D-glucose (^{18}FDG) for PETT studies (3). The yield of ^{18}FDG is ~ 20%, a factor of 2 higher than the synthesis using $[^{18}F]F_2$ and the experimental setup is considerably simpler because of the selectivity of the reagent.

In summary, large quantities (~ 144 mCi of $CH_3CO_2{}^{18}F$ from 360 mCi of ^{18}F at EOB) of $CH_3CO_2{}^{18}F$ are available, on line, from $[^{18}F]F_2$ after a 20-25 minute purge of the target gas through a solution of acetic acid/ammonium acetate. This ^{18}F-labeled precursor is readily available to institutions now producing $[^{18}F]F_2$ with no change in targetry required and is used in a new high yield synthesis of ^{18}FDG.

This research was supported in part by the Department of Energy, Division of Health and Environmental Research, and Public Health Service Grant NS-15380-06.

(1) Rozen, S., Lerman, O. and Kol, M., J. C. S. Chem. Comm., 443 (1981).
(2) Lerman, O., Tor, Y. and Rozen, S., J. Org. Chem. 46, 4629 (1981).
(3) Shiue, C.-Y., Salvadori, P., Wolf, A. P. et al., Manuscript in preparation.

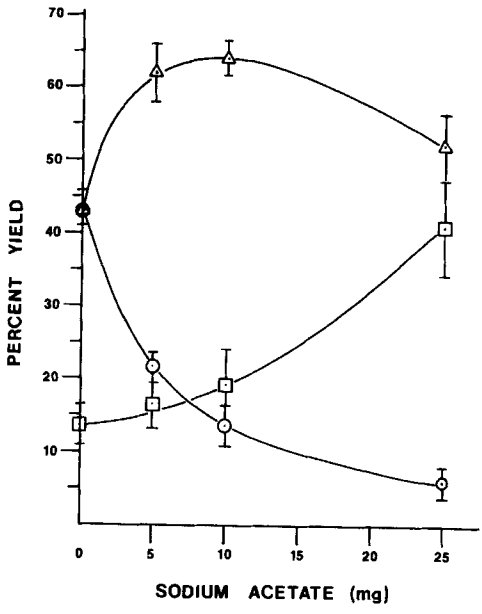


Fig. 1 The influence of sodium acetate concentration on the yield of acetyl hypofluorite (△——△) is shown along with the oxidant contained in the gas exiting the vessel (KI trap, ○——○). Each of these values is measured. The oxidant loss (□——□) is calculated and is the difference between the amount of F_2 purged through the acetic acid and the KI solutions and the sum of the oxidant in the acetic acid and KI solutions. Each point is the average ± SDM of 4 determinations.

F-18 PERCHLORYL FLUORIDE: SYNTHESIS AND REACTIONS

R.E. Ehrenkaufer and R.R. MacGregor.
University of Michigan Hospitals, Division of Nuclear Medicine, Ann Arbor, Michigan 48109.

Current investigations have demonstrated the need for continued development of methods for aromatic fluorination which are adaptable to radiopharmaceutical applications (1). Previous studies have shown that $FClO_3$ would react with unfunctionalized aryl-lithiums (such as phenyllithium) to produce moderate yields of aryl fluorides (i.e. fluorobenzene) (2). However, its use in the synthesis of functionalized aryl fluorides has not been reported. The goal of this work therefore, was to synthesize $[^{18}F]$-$FClO_3$ from a readily accessible ^{18}F precursor and demonstrate its usefulness in the fluorination of aryl lithiums containing pharmacologically interesting functional groups.

The synthesis of $^{18}FClO_3$ was carried out by the reaction of $[^{18}F]$-F_2 with $KClO_3$ (3). The $[^{18}F]$-F_2, which was formed by deuteron irradiation (4) of neon gas [$^{20}Ne(d,\alpha)^{18}F$, T 1/2 110 m, β^+ emitter] containing carrier amounts of added elemental fluorine (0.1 to 1.0%), was purged rapidly (200 cc/m) from the irradiated target through a columnn of granular $KClO_3$ at 90°-150° yielding $^{18}FClO_3$. Rapid on line purification of the $^{18}FClO_3$ was accomplished by passing the gas stream effluent from the $KClO_3$ reactor through a series of two solid phase scrubbers containing crushed NaOH pellets and granular $Na_2S_2O_3$ respectively. These effectively remove any unreacted F_2 and potential chlorine oxides that may have formed in the $KClO_3$ reactor. The effluent gas was then passed through a trap at liquid nitrogen temperature to isolate the $^{18}FClO_3$ (bp-47°, mp-148°). Under these conditions $^{18}FClO_3$ was rapidly prepared (less than 10 minutes) in yields averaging 23%. It should be noted that a maximum yield of 50% based $[^{18}F]$-F_2 is possible since 50% of the activity is consumed as $K^{18}F$.

$$[^{18}F]\text{-}F_2 + KClO_3 \rightarrow {}^{18}FClO_3 + K^{18}F$$

The $^{18}FClO_3$ was analyzed gas chromatographically both on a halocarbon/Kel-F column and Porapak-Q and was shown to be 91 to > 99% pure. The only radiochemical impurities present were determined to be ^{18}F labeled CF_4 and NF_3, which are known to be formed during ^{18}F-F_2 production within the target from impurities in the target gas mixture (5).

The $^{18}FClO_3$ was transferred into a reaction vessel (-78°) containing the desired aryl lithium by flushing the trap at room temperature with an inert-dry carrier gas (N_2, neon). The reactions were conveniently carried out in the solvents in which the aryl lithiums were initially formed (Et_2O, THF, hexane, etc) and were complete in 5 to 10 minutes. Workup consisted of an initial quenching of unreacted anion with CH_3I followed by aqueous extraction of the organic phase containing the aryl fluoride (as in the case of anisole and veratrole). With benzaldehyde (which was protected as the 1,3-dimethylimidazolidine) and aniline (protected as the N-t-Boc derivative) acidic workup at this point rapidly yielded the unblocked labeled aryl fluorides (6,7). Results are shown below in the table.

(1) Rosenfeld M.E. and Widdowson D.A., J.C.S. Chem. Comm., 914 (1979); Tewson T.J. and Welch M.J., ibid., 1149 (1979), Schrobilgen G., Firnau G., Chirakal R. and Garnett E.S., ibid., 198 (1981); Adam M.J., Pate B.D., Ruth T.J., et al., ibid, 733 (1981). Cacace F, Speranza M. and Wolf A.P., J. Labelled Comp. & Radiopharm., 18, 1721 (1981).
(2) Khutoretskii V.W., et al., Russ. Chem. Rev., 36, 145 (1967); Schlosser M. and Heinz G., Chem. Ber., 102, 1944 (1969).
(3) Englebrecht A. and Atzwanger H., J. Inorg. Nucl. Chem., 2, 348 (1956);
(4) Casella V., Ido T., Wolf A.P., et al., J. Nucl. Med., 21, 751 (1980).
(5) Bida G.T., Ehrenkaufer R.E., Wolf A.P., et al., J. Nucl. Med., 21, 758 (1980).
(6) Harris T.D. and Roth G.P., J. Org. Chem., 44, 2004 (1979).
(7) Muchowski J.M. and Venuti M.C., J. Org. Chem., 45, 4798 (1980).

TABLE 1. [^{18}F] Yields of the Aryl Fluorides

Starting Compound	Product	% Yield(^{18}F)+
anisole*	2-Fluoroanisole	34
veratrole*	3-Fluoroveratrole	21
N-(t-Boc)aniline	2-Fluoroaniline	24
	N-methyl-2-Fluoroanilinet	2
1,3-dimethyl-2-phenyl-imidazolidine	2-Fluorobenzaldehyde	3

*Anisole and veratrole are methoxybenzene and 1,2-dimethoxybenzone (or dimethyl catechol) respectively.

+Yields were based on $^{18}FClO_3$ radioactivity and determined by radiogas chromatography. These do not represent optimized yields. Except in the case of the aniline derivative no other volatile ^{18}F labeled peaks were observed.

†Since the dianion of t-Boc-aniline is formed by reaction with t-butyl-lithium, sequential reaction with $^{18}FClO_3$ followed by CH_3I is expected to yield this product. Identity was confirmed by GC/MS analysis (M/e 125).

NO-CARRIER-ADDED ^{18}F-FLUORIDE IN ORGANIC SOLVENTS: PRODUCTION AND LABELING RESULTS

M. Berridge, C. Crouzel, and D. Comar.
Commissariat a l'Energie Atomique, Departement de Biologie, Service Hospitalier Frederic Joliot, 91406 Orsay France.

While labeling reactions using displacement by F18-fluoride ion have enjoyed increasing interest (1,2,3), the production of usable fluoride activity has involved use of carrier, water, or solid salts which are insoluble in organic solvents. We report here the cyclotron production of anhydrous F18-fluoride with no added carrier, and handling methods which allow its delivery as a solution in organic solvents. The reactivity of this fluoride has been compared with certain other reported fluoride preparations, and the reaction conditions for its use in different labeling reactions have been investigated.

The activity was produced by the He3 bombardment of Ne20 to produce Nel8 using a mixture of 2% hydrogen in neon in a non-passivated nickel target (4). The Nel8 decays to F18 with a half- life of 1.5 sec while it is swept from the target at a flow rate of 2 cubic meters per hour. The F18-HF is trapped using a PTFE tube (5mm x 1m) cooled at -15 degrees. The yield of F18-HF by this method is 5.4 mCi/μA hr.

The HF contained in the large PTFE cold trap was flushed with dry nitrogen into a 1mm id PTFE tube while heating the large trap over 80°C. Over 90% of the activity was easily transferred. A small quantity (0.1-1ml) of the organic solvent of choice ($CHCl_3$, CH_2Cl_2, benzene, bromobenzene, DMSO, pyridine) was then used to wash the activity into a teflon reaction vessel of 2ml volume which could be tightly sealed. The activity recovery in solution was 95-100% when glycol sulfite, pyridine, or solutions of sulfonic acids, soluble salts, or triazine in other solvents were used, and 70-90% when the dry, distilled solvents were used alone. Use of the pre-mixed reaction solution for the collection of activity was therefore preferable to adding other reagents later to an F18-HF solution.

It has been noted in early experiments that while HF in solution was a good reagent for labeling via triazine decomposition, it was not an effective reagent for labeling by nucleophilic displacement. The addition of an alkalai metal carbonate however gave displacement yields which exceeded those obtained by other methods.

The disappointing yields and numerous by-products given by the triazine decomposition reaction was an incentive to investigate other methods of labeling aromatic rings. An exchange of F18 for aromatic iodine was successfully attempted. The iodinated compounds were easily prepared by the Sandmeyer procedure in 50-70% yield from the same aromatic amines used to prepare the aryl triazines. Heating for 10-40 min in DMSO containing no-carrier-added fluoride gave good yields of the desired labeled compound and no other labeled products.

Table I shows the early radiochemical yields obtained using this general method for the preparation of some simple molecules which serve as a comparison. The yields in each case equal or exceed former results when conditions are unchanged, and the yields of the new aromatic fluorination reaction are considerably higher than other methods.

Labeled Compound	Precursor	Solvent	Other	Yield	Ref. Yield
Fluoroethanol	Glycol Sulfite	-	K_2CO_3	60 %	60 % (5)
p-Fluorobenzonitrile	Piperidyldiazo-benzonitrile	Benzene	-	11 %	4 % (6)
p-Fluorobenzonitrile	p-Iodobenzonitrile	DMSO	$CsCO_3$	75 %	-
p-Fluoroaniline	p-Bromoaniline	DMSO	$CsCO_3$	10 %	-
Fluorobenzene	Iodobenzene	DMSO	$CsCO_3$	20 %	-
7-Fluoropalmitic acid	Benzyl-7-Mesyl-palmitate	DMSO	K_2CO_3	10 %	6 % (7)

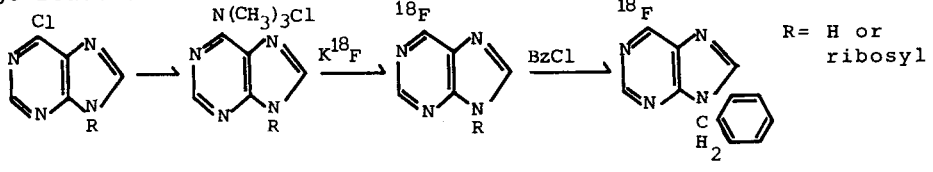
1) Tewson T., Welch M. and Raichle M. J. Nucl. Med. 19, 12, 1339 (1978).
2) Karim H. and Stöcklin G. J. Lab. Comp. Radiopharm. 13, 4, 519 (1977).
3) Gatley S., Hichwa R., Shaughnessy W. and Nickles R. Int. J. Appl. Radiat. Isot. 32, 211 (1981).
4) Crouzel C. and Comar D. Int. J. Appl. Radiat. Isot. 29, 407 (1978).
5) Tewson T. and Welch M. J. Nucl. Med. 21, 559 (1980).
6) Maeda M., Tewson T. and Welch M. in preparation.
7) Berridge M., Tewson T. and Welch M. J. Lab. Comp. Radiopharm. 18, 1, 240 (1981).

Work partly supported by INSERM contract #120006 and an NSF/CNRS scientific exchange award to MB.

^{18}F-LABELLED 6-FLUORO-PURINE DERIVATIVES AS A NEW TYPE BRAIN SCANNING AGENT

T. Irie, K. Fukushi, O. Inoue, T. Yamasaki, and Y. Kasida.
Division of Clinical Research, National Institute of Radiological Sciences, Anagawa, Chiba-shi(CHIBA), 280 Japan.
T. Ido.
Cyclotron Radioisotope Center, Tohoku University, Sendai-shi(MIYAGI).
T. Nozaki.
the Institute of Physical and Chemical Research, Wako-shi(SAITAMA).

6-Fluoro-9-benzylpurine-^{18}F has been found to be a hopeful brain scanning agent. We succeeded in synthesizing this compound by the following process, which proved to be superior to the halogen-exchange reaction:



Anhydrous K^{18}F prepared from aqueous ^{18}F was dissolved in DMF containing 18-Crown-6, to which trimethylpurin-6-ylammonium chloride was added. The solution was heated to give 6-fluoropurine-^{18}F, from which 6-fluoro-9-benzylpurine-^{18}F was prepared by benzylation. 6-fluoro-9-β-D-ribofuranosylpurine-^{18}F was prepared by similar fluorination from 9-β-D-ribofuranosylpurin-6-yltrimethylammonium chloride. Their yields for various preparation conditions are shown in Table 1.

The biodistribution of the three ^{18}F-labelled purine derivatives in mice were measured; they are shown in Fig. 1. A high brain concentration is obviously seen in the 9-benzyl compound. Chemical form of the ^{18}F-activity in the brain was shown to be F^- ion by the analysis of the brain homogenate with an alumina column. Enzymatic cleavage of this compound was proved to take place rapidly by its in vitro treatment with adenosine deaminase.

It is thus highly probable that 6-fluoro-9-benzylpurine penetrates the B.B.B. fairly freely owing to its lipophilic property and that its C-F bond is then cleaved enzymatically to give F^- ion whose clearance through the B.B.B. is slow. A remarkably increased clearance of ^{18}F-activity from the brain is observed for experimental animals whose B.B.B. were modified by infusion of mannitol into the carotid artery after administration of 6-fluoro-9-benzylpurine-^{18}F.

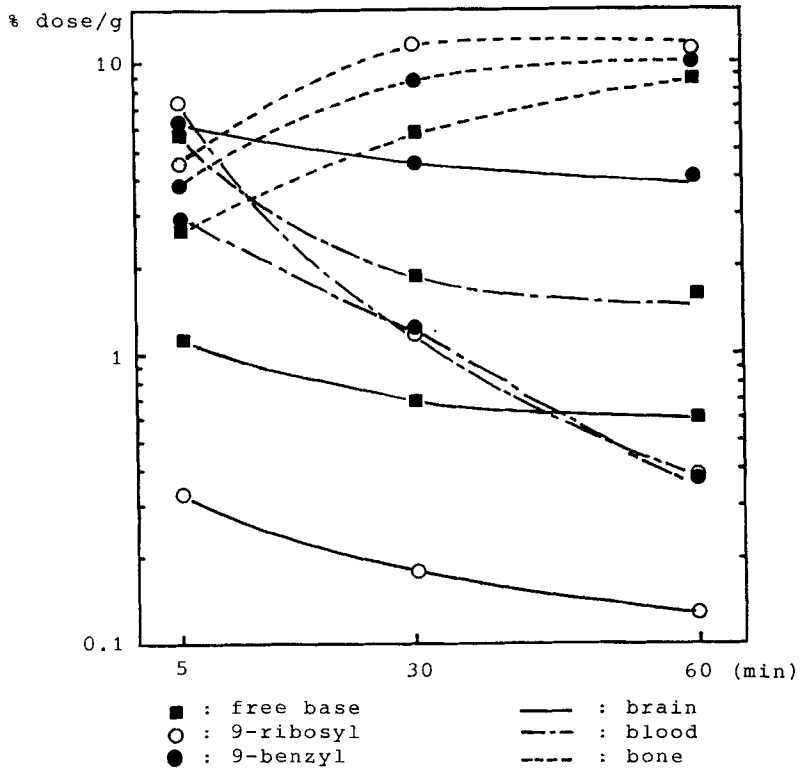
6-Fluoro-9-benzylpurine-^{18}F, therefore, is regarded as a potential radiopharmaceutical for the measurement of cerebral activities such as blood flow and/or adenosine deaminase activity. Studies on purine derivatives labelled with other radiohalogens and in the other positions are in progress, in order to develop radiopharmaceuticals with more desirable properties.

Table 1 Yields of (^{18}F)6-fluoropurine derivatives

Compound	Substrate (μmol)	^{18}F-Fluoride (μmol)	Reaction time and temp.	Radiochemical yield (%)
Free base	10	KF (5)	25 min, 80°C	13.9
	10	KHF_2 (5)	25 min, 80°C	12.3
	34	KF (5)	45 min, 80°C	37.7
9-Ribosyl	30	KF (5)	30 min, 60°C	63.5
	30	KOH (5)	30 min, 60°C	35.2

	Yield of 6-F-Purine (%)	Benzylation condition	Yield of N-9 benzylate	Radiochemical yield (%)
9-Benzyl	38.1	DMSO 200 μl BzCl 50 μl K_2CO_3 30 mg	44.2	16.9
	23.6	DMSO 200 μl BzCl 20 μl K_2CO_3 30 mg	11.4	2.7

Fig. 1 Distribution of (^{18}F)6-fluoropurine derivatives in mice



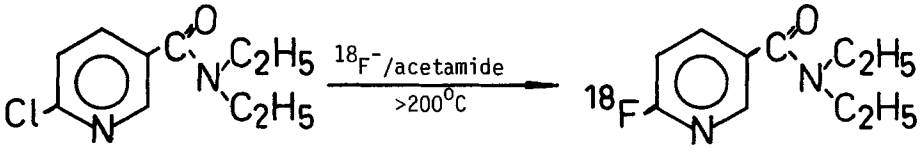
^{18}F-PRODUCTION IN A WATER TARGET WITH HIGH YIELDS FOR ^{18}F-LABELLING OF ORGANIC COMPOUNDS: SYNTHESIS OF 6-(^{18}F)-NICOTINIC ACID DIETHYLAMIDE

E.J. Knust, H.-J. Machulla, and M. Molls.
Institut für Med. Strahlenphysik und Strahlenbiologie, Universitätsklinikum, Hufelandstr. 55, D-4300 Essen, West Germany

In comparison to the available ^{18}F-production methods the ^{16}O(^{3}He,p)^{18}F reaction in the water target has the advantages of simplicity and easy handling. However, difficulties arise when i) producing ^{18}F with high yields due to the high energy deposit on the target and ii) transforming the aqueous solution of ^{18}F-fluoride into a form in which the halogen can easily undergo organic reactions such as nucleophilic substitution. Successful ^{18}F-labelling of aliphatic and aromatic compounds in high radiochemical yields using ^{18}F-fluoride produced by a water target was described by Robinson and Knust et al. (1,2,3).

With the aim of a high ^{18}F-production rate the water target was improved by an efficient cooling system, thus irradiations were performed with ^{3}He (36 MeV) at a beam current of 30 µA resulting in a reproducible ^{18}F-yield of 0.5 Ci ($1.85 \cdot 10^{10}$ Bq) at the end of bombardment.

The promising results of the animal experiments of 2-(^{18}F)-nicotinic acid diethylamide (3) prompted us to extend the investigation to the isomeric compound, i.e. 6-(^{18}F)-nicotinic acid diethylamide which was synthesized in a similar way:



After purification by high pressure liquid chromatography radiochemical yields up to 40% could be obtained in less than one half-life of ^{18}F.

Tissue distribution of 6-(^{18}F)-nicotinic acid diethylamide in various organs of mice showed a pattern similar to the 2-(^{18}F)-isomer, i.e. a very fast accumulation of activity in all well-perfused organs such as brain, lungs, heart, and kidneys. Within the first 30 seconds after intravenous injection a maximum of about 6%/g is accumulated in the brain decreasing to about 2.5%/g in the following 5 minutes and thereafter remaining practically constant for one hour.

A striking difference, however, is observed in the case of 6-(^{18}F)-nicotinic acid diethylamide when compared with the 2-(^{18}F)-isomer:
Separating the brain into cerebrum and cerebellum an activity ratio of about 30 is obtained at maximum uptake, indicating different pathways of metabolism of the two isomers.

(1) Robinson G.D., Proc. Symp. New Develop. Radiopharm. Lab. Comp., Copenhagen, Vol.I, IAEA Vienna, p.423 (1973).
(2) Knust E.J., Schüller M. and Stöcklin G., J. Lab. Comp. Radiopharm., 17, 353 (1980).
(3) Knust E.J., Müller-Platz C. and Schüller M., J. Radioanal. Chem., in press 73(1), (1982).

^{18}F-FLUOROACETATE: An AGENT FOR INTRODUCING NO-CARRIER-ADDED FLUORINE-18 INTO UROKINASE WITHOUT LOSS OF BIOLOGICAL ACTIVITY

C.M. Müller-Platz, G. Kloster, G. Legler[+], and G. Stöcklin.
Institut für Chemie 1 (Nuklearchemie), Kernforschungsanlage Jülich GmbH, D-5170 Jülich, FRG,
[+]Institut für Biochemie der Universität Köln, FRG.

Deep-vein thrombosis is by far the most frequent origin of embolism. Its localisation using radioiodine- or technetium-labelled proteins such as urokinase, which participate in clot formation and fibrinolysis, is hampered by a high blood background. In addition, high activity in the kidneys and bladder diminishes the accuracy of the method in the pelvic region (1). We therefore chose fluorine-18 as a label because it allows the application of positron emission tomography. Since direct labelling of urokinase (UK) with fluorine-18 in aqueous solution is not possible, 2-^{18}F-fluoroacetate (^{18}FA) was used as an intermediate. ^{18}FA activated with the water-soluble (N-ethyl-N'-(dimethylamino)propyl)carbodiimide (WSC) can be coupled covalently to a free amino group of the protein. This label is more stable compared to direct labelling with radioiodine or chelating with suitable metal isotopes, and should therefore prevent additional increase of the blood background due to in vivo released label.

^{18}F was produced by the ^{16}O(^{3}He,p)^{18}F reaction at the Jülich CV 28 compact cyclotron using ^{3}He particles with an incident energy of 36 MeV on a water target provided with additional cooling. Yields of 2.9 GBq $^{18}F^-_{aq}$ were obtained after 1 hr bombardment with a 35 µA beam current.

^{18}F-fluoroacetyl urokinase (^{18}FA-UK) was synthetized in a two-step reaction (eq. 1 to 3, see below). ^{18}FA was prepared via nucleophilic exchange from redistilled ethylbromoacetate in an acetamide melt (previously recrystallized from toluene). This was followed by hydrolysis of the ether-extracted esters using aqueous 5N KOH under reflux, a procedure used before in our laboratory for the preparation of ω-^{18}F-fatty acids (2). Unlike the ^{18}F-fatty acids, however, the ^{18}FA was prepared in high radiochemical yields without the addition of carrier. By adding about 1 mmol KOH to the irradiated water before evaporation to dryness, and by using special teflon containers, the radiochemical yield was increased to more than 50%. This is probably due to a competition of OH^- for cationic impurities and residual water, so that $^{18}F^-$ maintains its nucleophilicity. This additional step requires about 80 min. Starting from 2.2 GBq $^{18}F^-_{aq}$, 670 MBq ^{18}FA with a minimum specific activity of about 37 TBq/mMol were obtained following purification.

$$BrCH_2CO_2Et \xrightarrow[-Br^- \text{ (acetamide melt)}]{^{18}F^-} {}^{18}FCH_2CO_2Et \qquad (1)$$

$$^{18}FCH_2CO_2Et \xrightarrow[-EtOH]{H_2O/KOH} {}^{18}FCH_2CO_2K \qquad (2)$$

$$^{18}FCH_2CO_2^- + UK\text{-}NH_2 \xrightarrow[-R\text{-}NH\text{-}CO\text{-}NH\text{-}R']{R\text{-}N=C=N\text{-}R'} UK\text{-}NH\text{-}CO\text{-}CH_2{}^{18}F \qquad (3)$$

The next procedure involves the proton-catalyzed activation of WSC and formation of the isourea ester with the ^{18}FA anion. This is followed by aminolysis of the ester with the free amino groups of UK to yield ^{18}FA-UK (3). The formation of the activated ester was performed at

pH 5 and the solution was added dropwise to a UK-solution at pH 7. This step, including the purification of the labelled UK using 1x4 cm Dowex by an anion-exchange column (OH^--form), takes about 30 min. 166 MBq ^{18}FA-UK (30% radiochemical yield) with a specific activity $\geq$ 411 MBq/mg UK were obtained.

A crucial point to be tested is the biological activity of the ^{18}FA-UK, since the large excess of unlabelled UK cannot be separated from the ^{18}FA-UK within the time available. Unchanged total enzymatic activity of the bulk material as determined by batch tests with the peptide substrate S2444 (KABI) (4) does not provide sufficient proof for the retention of biological activity of ^{18}FA-UK. For this reason, affinity chromatography (5) was used as an additional method to prove that the binding activity of ^{18}FA-UK was unchanged. In view of the time requirements for affinity chromatography, UK was labelled with 1-^{14}C-fluoroacetate.

Despite its lower specific activity, 1-^{14}C-fluoroacetyl-UK exhibited an identical behavior as authentic UK, and both the chromatographic identification and specific enzymatic activity results obtained with affinity chromatography were identical to those obtained with ion exchange chromatography.

These results clearly indicate that the ^{18}FA-UK is not selectively altered during labelling. The labelling method described here should be applicable to other proteins as well, provided that the necessary tests for biological activity can be performed.

(1) Neidhart P., Hofer B., Duckert F. and Fridrich R., Schweiz. med. Wschr., 104, 141 (1974).
(2) Knust E.J., Schüller M. and Stöcklin G., J. Lab. Comp. Radiopharm., 17, 353 (1980).
(3) Sheehan J.C., Preston J. and Cruickshank P.A., J. Am. Chem. Soc., 87, 2492 (1965).
(4) Claeson G., Aurell L., Karlsson G., Gustavson S., Friberger P., Arielly S. and Simonsson R., "Chromogenic peptide substrates: Chemistry and clinical usage", Churchill Livingstone, Edinburgh, 1979, p. 20.
(5) Soberano M.E., Ong E.B., Johnson A.J., Levy M. and Schöller G., Biochim. Biophys. Acta, 445, 763 (1976).