Synthesis and Biodistribution of Radioiodinated Selenonium Salts: Potential Myocardial Imaging Agents

Parikh A. S., Basmadjian G. P., Gilliland D. L., Greenwood R. B., Rieger J. A. and Weaver A.

College of Pharmacy, University of Oklahoma, Health Sciences Center, Oklahoma City, Oklahoma 73190 USA.

SUMMARY

(o-[^{125}I]iodobenzyl)benzylmethylselenonium tetrafluoroborate (1) and (o-[^{125}I]iodobenzyl)dibenzylselenonium tetrafluoroborate (2) were synthesized by methylation and benzylation of o-[^{125}I]iodobenzyl benzyl selenide respectively. The starting compound o-iodobenzyl benzyl selenide was radioiodinated with sodium [^{125}I]iodide by melt-exchange in a sealed ampoule maintained at 75°C for 12 hours, with subsequent chromatographic purification yielding the pure compound in a radiochemical yield of 33.0% and specific activity of at least 68.0 mCi/mmol. Tissue distribution studies of compounds 1 and 2, performed in rats, showed that the labelled compounds accumulated rapidly in the heart. The maximum heart-to-blood ratios for compounds 1 and 2 were 4.2:1 and 4.4:1 compared to that of 2.0:1 for previously synthesized [^{75}Se]Trimethylselenonium Iodide (3).

Keywords: Radioiodinated Selenonium, Salts, Myocardial Imaging Agents

INTRODUCTION

Thallium-201 is the most widely used myocardial perfusion agent but has the disadvantages of high cost and inefficient detection of its low-energy photons. The search for a better myocardial imaging agent to supersede the cyclotron-produced [^{201}Tl]thallous chloride has centered upon a number of lipophilic cationic complexes labeled with isotopes having better physical properties.

Investigations with radiolabelled phosphonium monocations such as [^{3}H]-tetraphenyl phosphonium bromide, [^{3}H]methyl triphenyl phosphonium bromide, [^{11}C]methyl triphenyl phosphonium bromide (1,2), and a series of radioiodinated iodomethyl ammoniums have shown promising myocardial uptake (3).

Previous studies in our laboratory with the cation [^{75}Se]trimethylselenonium iodide (3) showed relatively low myocardial uptake with the maximum uptake

0362-4803/86/080815-10$05.00

Received November 21, 1985
Revised February 3, 1986

in kidneys, as predicted, due to the hydrophilic nature of the compound (4). The purpose of this study was to substitute one or more methyl group(s) from [^{75}Se]trimethylselenonium iodide with benzyl group(s), radioiodinate and evaluate the resulting lipophilic compounds (o-[^{125}I]iodobenzyl)benzylmethylselenonium tetrafluoroborate (1) and (o-[^{125}I]iodobenzyl)dibenzylselenonium tetrafluoroborate (2) for use as potential myocardial perfusion agents (Fig. 1).

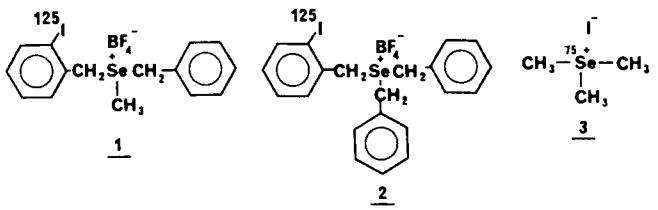


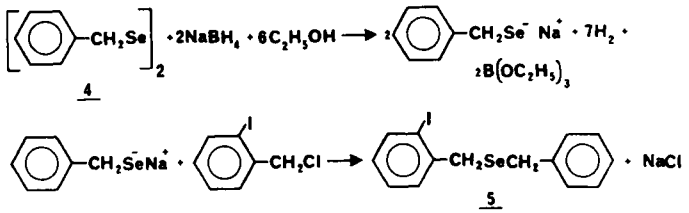
Figure 1. Selenonium Salts 1, 2, 3.

RESULTS AND DISCUSSION

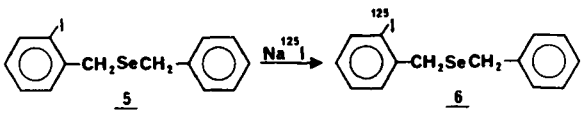
The compound o-iodobenzyl benzyl selenide (5) was successfully synthesized from dibenzyl diselenide (4) according to Scheme 1. The synthesis of o-iodobenzyl benzylmethylselenonium tetrafluoroborate (7) and o-iodobenzyl dibenzylselenonium tosylate (8) shared the common intermediate (5) which was methylated or benzylated to form compound 7 or compound 8 respectively (cf Scheme 3).

Radioiodination by the exchange-reaction could not be carried out on the final cold selenonium compounds VII and VIII, since at that stage, radioiodine would exchange with the anion BF_4^- in preference to the ortho-iodine atom on the benzyl ring of the selenonium salts. Thus radioiodination by exchange-reaction was carried out on o-iodobenzyl benzyl selenide (Scheme 2), before it was methylated or benzylated to form the selenonium salts.

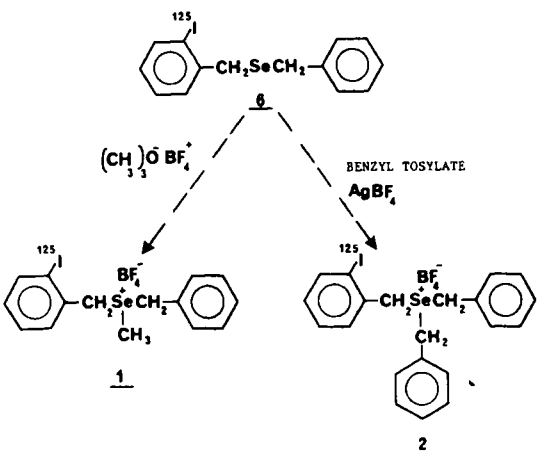
The synthesis of 1 and 2 shared a common intermediate (o-[^{125}I]iodobenzyl) benzyl selenide (6) which was methylated or benzylated to form compound 1 or compound 2 respectively (Scheme 3). Compounds 1, 2, and 6 were identified by TLC using their corresponding non-radioactive analogs as references. In each case, radiochromatograms showed a single peak corresponding to a radioactive



Scheme 1. Synthesis of o-Iodobenzyl Benzyl Selenide (5).



Scheme 2. Preparation of (o-[^{125}I]Iodobenzyl) Benzyl Selenide (6).



Scheme 3. Preparation of (o-[^{125}I]Iodobenzyl)benzylmethylselenonium Tetrafluoroborate (1) and (o-[^{125}I]Iodobenzyl)dibenzylselenonium Tetrafluoroborate (2).

compound that co-chromatographed with the reference compound.

Compound 2, being a triarylselenonium salt, was found to be less stable than compound 1, which is a diarylakylselenonium salt, and the former slowly decomposed to o-[^{125}I]iodobenzyl benzyl selenide, *in vitro*, as seen on TLC. This phenomenon was in agreement with the known thermal instability of selenonium salts in general (5), and photolysis of triarylselenonium salts in

TABLE 1

Tissue distribution of 1 and 2 in rats*

Tissue	Minutes after administration						
	1	2	5	15	30	60	120
	Compound 1						
Blood	0.12±0.00	0.11±0.00	0.12±0.00	0.11±0.01	0.07±0.01	0.07±0.00	0.06±0.00
Heart	0.43±0.01	0.46±0.01	0.21±0.00	0.08±0.00	0.05±0.00	0.05±0.00	0.05±0.00
Lung	0.13±0.01	0.83±0.03	0.10±0.00	0.06±0.01	0.05±0.00	0.04±0.00	0.03±0.00
Liver	0.43±0.01	0.78±0.03	0.45±0.01	0.42±0.03	0.37±0.02	0.17±0.02	0.11±0.01
Kidney	1.25±0.04	0.56±0.01	1.35±0.07	0.99±0.06	0.75±0.03	0.69±0.10	0.46±0.02
Urine	0.01±0.01	0.04±0.01	0.96±0.26	3.38±1.98	14.76±5.60	15.28±7.02	17.12±1.12
	Compound 2						
Blood	0.15±0.02	0.11±0.01	0.08±0.01	0.07±0.00	0.06±0.00	0.05±0.00	0.03±0.00
Heart	0.63±0.07	0.48±0.02	0.34±0.02	0.16±0.00	0.08±0.01	0.05±0.01	0.02±0.00
Lung	1.08±0.05	1.02±0.08	0.51±0.03	0.48±0.02	0.37±0.01	0.23±0.01	0.12±0.01
Liver	0.64±0.03	0.74±0.03	0.64±0.02	0.48±0.03	0.34±0.02	0.20±0.04	0.11±0.01
Kidney	0.53±0.03	0.52±0.03	0.39±0.02	0.36±0.01	0.28±0.01	0.29±0.02	0.19±0.03
Urine	0.00±0.00	0.03±0.00	0.13±0.03	0.81±0.07	1.14±0.26	1.87±0.47	6.81±1.71

*Values represent mean %Kg-dose/gm ± Standard Error of Mean (S.E.M.) for 3 rats per time interval. Dose is 5 μCi. Other tissues studied but not listed are: brain, adrenal, spleen, small intestine, fat, muscle, femur and testes.

TABLE 2

Tissue-to-Blood Ratios of 1 and 2

Tissue	Minutes after administration						
	1	2	5	15	30	60	120
				Compound 1			
Heart	3.55	4.23	1.70	0.74	0.77	0.79	0.85
Lung	1.09	7.67	0.84	0.53	0.71	0.54	0.57
Liver	3.53	7.21	3.67	3.89	5.32	2.47	1.87
Kidney	10.18	5.18	11.08	9.23	10.74	10.24	7.91
Urine	0.09	0.35	7.87	31.40	210.26	225.74	292.19
				Compound 2			
Heart	4.23	4.41	4.20	2.32	1.26	0.94	0.55
Lung	7.25	9.39	6.29	7.12	6.02	4.57	3.32
Liver	4.31	6.82	7.89	7.01	5.51	3.90	3.21
Kidney	3.57	4.83	4.84	5.25	4.65	5.62	5.39
Urine	0.02	0.26	1.61	11.92	18.65	36.55	190.79

particular (6). Hence, the reactions of Scheme 3 were carried out in the dark and the selenoniums 1 and 2 were found to be stable after storage in the dark at refrigeration temperatures for one week. Thus, compounds 1 and 2 were used for biodistribution studies within one week following their preparation.

The results of the biodistribution studies of compounds 1 and 2 are summarized in Tables 1 and 2. The absolute uptake of compound 2 (0.63% Kg-dose/gm) in the myocardium was higher than that of compound 1 (0.46% Kg-dose/gm), as predicted, probably due to the increased lipophilicity of compound 2. The maximum heart-to-blood ratios for compounds (1) and (2) were 4.2:1 and 4.4:1 respectively. Both compounds were rapidly cleared from circulation by the kidneys. The higher rate of renal clearance of compound 1 (17% Kg-dose/gm in urine) compared to that of the compound 2 (7% Kg-dose/gm in urine) could be due to the fact that the latter has potential to undergo rapid *in vivo*

degradation to a neutral and more lipophilic molecule of a selenide, which could have slower renal clearance, and higher and prolonged uptake in tissues such as fat, adrenal, liver, lung and brain.

Thus, attempts to make the cationic compound 3 more lipophilic by replacing one or more of the methyl group(s) with benzyl group(s), did increase the absolute myocardial uptake and decrease the amount of this cationic compound being cleared through the renal system.

EXPERIMENTAL

MATERIALS AND METHODS:

All reagents were of analytical grade and purchased from Aldrich Chemical Company except for trimethyloxonium tetrafluoroborate, which was purchased from Fluka Chemical Corporation. Sodium [^{125}I] iodide (C.F., Carrier Free) was purchased from ICN Chemical Isotope Division. Melting points were determined on a Thomas Hoover melting point apparatus. ^{1}H-NMR spectra were recorded on a Varian EM 360A NMR spectrometer (60MHz). Thin layer chromatography (TLC) was done on silica gel plates containing a fluorescent indicator (Eastman, cat. #13181). Radiochromatograms were recorded on a Dunnschitt-Scanner II auto scanner.

o-Iodobenzyl Benzyl Selenide (5): A method modified from Gassman, Miura and Mossman (7) was used. The reaction was run under a nitrogen atmosphere to prevent oxidation of oxygen-sensitive selenide ions. To a suspension of dibenzyl diselenide (4) (60 mmol, 20.4 g) in 200 ml of absolute ethanol at room temperature was added sodium borohydride (130 mmol, 4.9 g) dissolved in 50 ml of absolute ethanol, with magnetic stirring and in dropwise fashion. To the resulting colorless mixture containing sodium benzyl selenide was added dropwise a solution of o-Iodobenzyl chloride (120 mmol, 30.3 g) in 30 ml of absolute ethanol at room temperature. The reaction mixture was stirred overnight, evaporated to dryness under reduced pressure and extracted with ether. The extract was dried over anhydrous $MgSO_4$, filtered and evaporated. The yellowish white solid was recrystallized twice from absolute ethanol. Yield 16.83 g (36.24%), mp 61-63°C. ^{1}H-NMR ($CDCl_3$): 7.90-6.73 (m, 9H, ArH), 3.8 (s, 4H, CH_2). TLC on silica gel

developed with solvent systems petroleum ether; chloroform:hexane (1:9); chloroform; ethylacetate:benzene (1:1) and methanol indicated the presence of only one compound with R_f value of 0.11, 0.32, 0.71, 0.72 and 0.77 respectively. Analysis calculated for $C_{14}H_{13}SeI$:C, 43.43; H, 3.39. Found: C, 43.36; H, 3.49.

o-[^{125}I]Iodobenzyl Benzyl Selenide (6): The synthesis of 6 was carried out by a modification of the melt method, first developed by Elias, Arnold and Kloss (8). o-Iodobenzyl benzyl selenide (4 mg, 0.0103 mmol) was dissolved in 1.0 ml of absolute ethanol in a 5 cc glass ampoule. To this was added 4.0 mCi of sodium [^{125}I] iodide (Carrier Free), and the solvent was evaporated to dryness with a stream of nitrogen without heating. The ampoule was sealed, assayed in a dose calibrator (2.13 mCi), and kept in an oven at 75°C for 12 h. The ampoule was allowed to cool to room temperature, broken open and 0.5 ml of chloroform added to dissolve the contents. The crude product was purified by preparative TLC utilizing chloroform:hexane (1:9) as the solvent system. The pure product was dissolved in 2.0 ml of dry methylene chloride. Radiochemical yield was 0.703 mCi (33.0%), specific activity of at least 68.0 mCi/mmol. Radiochromatograms of TLC strips developed with the solvent systems of petroleum ether; chloroform:hexane (1:9); chloroform; ethylacetate:benzene (1:1) and methanol showed a single peak (R_f 0.11, 0.32, 0.71, 0.72 and 0.77 respectively) corresponding to the radioactive compound 6 that co-chromatographed with the reference nonradioactive compound 5.

o-Iodobenzyl Benzylmethylselenonium Tetrafluoroborate (7): A method modified from Gassman, Miura and Mossman (6) was used. To a suspension of trimethyloxonium tetrafluoroborate (100 mg, 0.67 mmol) in 5 ml of methylene chloride at -15°C was added dropwise a solution of o-iodobenzyl benzyl selenide (174 mg 0.5 mmol) in 5 ml of methylene chloride. The reaction mixture was stirred for 12 h under nitrogen atmosphere, while it gradually attained room temperature. The reaction mixture was filtered, and the solvent was evaporated under reduced pressure. The solid residue was recrystallized twice from methylene chloride-ether to give a white solid. Yield 116 mg (51.6%), mp 82-85°C. ^{1}H NMR ($CDCl_3$): 7.97-6.97 (m, 9H, ArH), 4.97-4.7 (m, 4H, CH_2), 2.7 (s, 3H, CH_3). TLC on silica gel developed with solvent systems of chlorform:hexane (1:9); chloroform:methanol

(9:1) and methanol indicated the presence of one compound with R_f values of 0, 0.15 and 0.18 respectively.

(o-[^{125}I]Iodobenzyl)benzylmethylselenonium tetrafluoroborate (1): To a suspension of trimethyloxonium tetrafluoroborate (5.0 mg, 0.03 mmol) in 5 ml of methylene chloride at -15°C was added dropwise a solution of 250 μCi of (o-[^{125}I]iodobenzyl) benzyl selenide (1.42 mg, 0.0037 mmol, 68.0 mCi/mmol) in 2 ml of dry methylene chloride. The reaction mixture was stirred for 12 h nitrogen atmosphere, while it gradually attained room temperature. The reaction mixture was filtered, and the filtrate was evaporated with a stream of nitrogen. Radiochemical yield was 204 μCi (81.6%), specific activity at least 68.0 mCi/mmol. Radiochromatograms of TLC strips developed with solvent systems of chloroform:hexane (1:9); chloroform:methanol (9:1) and methanol showed a single peak (R_f 0, 0.15, 0.18 respectively) corresponding to 1 that co-chromatographed with the reference nonradioactive compound 7.

(o-Iodobenzyl)dibenzylselenonium Tosylate (8): To a solution of o-iodobenzyl benzyl selenide (193.5 mg, 0.5 mmol) in 5 ml of methylene chloride at 0°C was added dropwise a freshly prepared solution of benzyl tosylate (262 mg, 1 mmol), as described by Kochi and Hammond (9). The reaction mixture was stirred for 8 h under nitrogen atmosphere, while it gradually attained room temperature. The reaction mixture was filtered, and the solvent was evaporated under reduced pressure. The solid residue was recrystallized twice from methylene chloride-ether to give a white solid. Yield 168 mg (51.8%), mp 122-124°C. ^{1}H NMR ($CDCl_3$). 7.97-7.0 (m, 18H, ArH), 5.3-4.9 (m, 6H, CH_2), 2.4 (s, 3H, CH_3). TLC on silica gel developed with solvent systems of chloroform: hexane (1:9) and chloroform:methanol (9:1) indicated the presence of one compound with R_f values of 0 and 0.44.

(o-[^{125}I]Iodobenzyl)dibenzylselenonium Tetrafluoroborate (2): To a solution of 200 μCi of o-[^{125}I]iodobenzyl benzyl selenide (1.14 mg, 0.003 mmol, 68.0 mCi/mmol) in 2 ml of dry methylene chloride at 0°C was added 5 ml of saturated solution of $AgBF_4$ in methylene chloride (about 50 mg $AgBF_4$/5 ml). To the above mixture was added dropwise a freshly prepared solution of benzyl tosylate (8 mg, 0.03 mmol) in methylene chloride. The reaction mixture was

stirred for 8 h under a nitrogen atmosphere, while it gradually attained room temperature. The solvent was evaporated with a stream of nitrogen. Radiochemical yield was 152 μCi (78%) specific activity at least 68.0 mCi/mmol. Radiochromatograms of TLC strips developed with solvent systems of chloroform: hexane (1:9) and chloroform:methanol (9:1) showed a single peak with R_f values of 0 and 0.44 respectively.

Formulation of Radioactive Compounds

Aseptic techniques were followed for the formulation of radioactive compounds. 204 μCi of (o-[^{125}I]iodobenzyl)benzylmethylselenonium tetrafluoroborate (1) was dissolved in 6 ml of sterile physiological saline and the solution was filtered through a 0.2 μm filter into a sterile, pyrogen-free vial. The formulation 'ready for injection' had a concentration of about 3.3 μCi/0.1 ml. 152 μCi of (o-[^{125}I]iodobenzyl)dibenzylselenonium tetrafluoroborate (2) was dissolved in 5 ml of 1:1:8 ethanol:tween 80:normal saline. The solution was filtered through a 0.2 μm filter into a sterile, pyrogen-free vial. The formulation 'ready for injection' had a concentration of about 2.9 μCi/0.1 ml.

Biodistribution of the Radipactive Compounds 1 and 2

Biodistribtuion studies were performed in mature male Sprague-Dawley rats. All animals were maintained on a chow diet, and provided with tap water until used. The mature rats weighed between 150 and 300 grams.

The animals were anesthetized with sodium pentobarbital, 30 mg/kg, intraperitoneally. After assuring tolerance to the applied pressure on the foot, the right ascending femoral vein was exposed, and approximately 0.2 ml of the 'ready for injection' formulation of 1 or 2 was injected via the ascending path of the femoral vein. At various time periods post injection, the heart was exposed (1-2 seconds prior to the desired time period) and 1-1.5 ml of blood was aspirated into a heparinized syringe by cardiac puncture. The heart was then quickly excised, a urine sample withdrawn from the bladder, and other tissues and/or organs including liver, lungs, kidneys, spleen, small intestine, testes, muscle, fat from around the heart muscle, brain and adrenals were removed. Each excised tissue was stripped of adipose tissue, washed with normal saline, blotted dry, weighed on an electronic digital balance, placed in 12 x 75 mm counting

tubes and counted in a Packard Multi-Prias gamma counter. The amount of radioactivity in the tissue samples was expressed as percent of injected dose per gram of the tissue sample normalized to an animal body weight of one Kg, using the following equation:

$$\%\text{Kg-Dose/Gm} = \frac{\text{cpm sample-cpm BKG}}{\text{sample weight (gm)}} \times \frac{\text{Activity in Standard (in } \mu\text{Ci)}}{\text{Activity injected (in } \mu\text{Ci)}} \times \frac{100}{\text{Standard (cpm) X Dil. Factor}} \times \text{Body wt. (in Kg)}$$

REFERENCES

1. Woo D.V. - Circulation 66: II-124 (1982)

2. Srivastava P.C., Callahan A.P. and Knapp F.F. - J. Nucl. Med. 24: P43 (1983)

3. Huang C.C., Friedman A.M. and Rayudu G.V.S. - J. Label. Cmpd. Radiopharm. 18: 270 (1981)

4. Weaver A., Basmadjian, G. and Sadek S. - Clinical Nucl. Med. 8: P9 (1983)

5. Irgolic K.J. and Kudchakar M.V. - Selenium, (Zingaro R.A. and Cooper W.C., eds.), Van Nostrand Reinhold Company, New York. 408 (1974)

6. Crivello J.V. and Lam J.H.W. - J. Polym. Sci. Polym. Chem. Ed. 17: 1047 (1979)

7. Gassman P.G., Miura T. and Mossman A. - J. Org. Chem. 47: 954 (1982)

8. Elias H., Arnold C. and Kloss G. - Int. J. Appl. Radiat. Isot. 24: 463 (1975)

9. Kochi J.K. and Hammond G.S. - J. Am. Chem. Soc. 75: 3443 (1953)