A New, Well-Retained Myocardial Imaging Agent: Radioiodinated 15-(p-Iodophenyl)-6-Tellurapentadecanoic Acid

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A method involving the acid-catalyzed decomposition of a piperidyltriazene intermediate in the presence of radioiodide has been developed for the synthesis of radioiodinated 15-(p-iodophenyl)-6-tellurapentadecanoic acid. The iodine-125-labeled agent shows rapid, pronounced myocardial uptake in rats (5.30–6.45% injected dose/g after 5 min) and also exhibits the prolonged retention previously observed with 9-[^{123m}Te]telluraheptadecanoic acid (9-[^{123m}Te]HDA). After 6 hr, the heart uptake remained high (3.89–5.33% dose/g) and decreased only to 3.02–3.41% dose/g after 24 hr. Very low blood activity was detected (0.24–0.27% dose/g at 5 min; 0.29–0.32% dose/g at 6 hr) and the heart-to-blood ratios were high (22:1 at 5 min; 15:1 at 6 hr). Minimal deiodination was demonstrated by the low thyroid uptake (1.41–1.63% dose/g at 5 min; 5.33–7.08% dose/g at 6 hr). The rapid and pronounced uptake, prolonged myocardial retention, and low in vivo deiodination make this agent attractive for further evaluation.

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A variety of terminal radioiodinated long-chain fatty acids (*1–3*) have been developed and evaluated as alternatives to thallium-201 to detect changes in myocardial perfusion (*4–6*). Feinendegen and coworkers have reported the use of 17-[^{123}I]-iodoheptadecanoic acid in humans to measure differences in regional myocardial fatty-acid metabolism between normal and ischemic tissue and in several types of cardiomyopathy (*6,7*). The clinical use of these agents may be limited, however, because of the significant radioactivity remaining in the blood pool after administration and the relatively short residence time of the radiolabeled fatty acid in the myocardium. The loss of iodide from the molecule could result from either complete catabolism of the fatty acid or direct cleavage of the carbon-iodine bond by chemical or enzymic routes. The relative contributions from these two processes have not been resolved, however (*6,7*). Despite these problems, 17-[^{123}I]iodoheptadecanoic acid has apparently proven to be a useful probe of regional fatty-acid metabolism when special correction methods are applied to account for the activity in the blood pool (*6,7*). More recently the problem of facile iodide loss has evidently been overcome by the preparation of radioiodinated 15-(p-iodophenyl)pentadecanoic acid (*8,9*), and the iodine-123-labeled agent is now under clinical evaluation (*10*). Although the product does not suffer deiodination, the myocardial washout is relatively rapid (*8,9*).

We have incorporated the tellurium heteroatom into the fatty-acid chain as a means of inhibiting metabolism. The 9-[^{123m}Te]telluraheptadecanoic acid (9-THDA) shows rapid and pronounced myocardial uptake in rats (*11–13*) and dogs (*14,15*). The unique feature of this fatty-acid analog is the prolonged myocardial retention or "trapping" of the radiolabel. In dogs, only 13% of the rapid, peak concentration washes out of the myocardium after 1 day and only 36% is lost after 5 days (*14*). Excellent myocardial images have been obtained in normal and infarcted dogs (*14,15*), and studies with a miniature cadmium telluride probe have evaluated the rates of

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uptake and loss from normal and ischemic regions of the dog myocardium (*15*).

More recently we have directed our efforts toward preparing a radioiodinated fatty acid containing stable tellurium. This fatty acid was envisioned as an agent that would retain the unique "trapping" behavior observed with 9-THDA. The nonradioactive tellurium heteroatom would produce the prolonged myocardial retention, and the favorable radionuclidic properties of iodine-123 could then be used for imaging studies and regional measurements of fatty-acid metabolism. The terminal position was chosen for introduction of the radioiodine because studies with 9-tellura-[10-^{14}C]heptadecanoic acid indicated that this region of the molecule was retained in the myocardium (*16*). A synthesis of radioiodinated 17-iodo-9-telluraheptadecanoic acid was developed and the product showed myocardial specificity but suffered rapid in vivo deiodination (*17*).

Studies have shown the pronounced and prolonged myocardial uptake of 15-phenyl-6-[^{123m}Te]tellurapentadecanoic acid, demonstrating that the presence of the terminal phenyl moiety does not interfere with the unique properties observed with 9-THDA (*18*). The goals of the present studies were to develop a synthesis of a model radioiodinated tellurium fatty acid in which the iodine atom is stabilized by attachment to the para position of a phenyl ring, and to evaluate the distribution of this new agent in rats.

MATERIALS AND METHODS

General. The I-125 was obtained commercially. All solvents and chemicals were analytical grade and were used without further purification. Column chromatography was performed with silicic acid (60–200 mesh). Thin-layer chromatographic analyses (TLC) used precoated silica gel PF-254 plates.* The synthesis and chromatographic procedures were conducted under red light to minimize photochemically induced oxidation of the tellurium compounds.

Tissue distribution studies. The distribution of the radiolabeled fatty acid was evaluated in 160- to 180-g female Fischer 344 rats† that were allowed food and water *ad libitum*. The fatty acid was dissolved in 200 μl of absolute ethanol and added dropwise to a stirred solution of delipidated 6% bovine serum albumin at 40°C. After filtration through 0.22-micron Millipore filters, the solution was administered to the rats by injection in a lateral tail vein. The injected solution was 0.5 ml in volume and contained 5–10 μCi of the radiolabeled fatty acid. Groups of four rats were killed after each time period; they were anesthetized with ether, killed by cervical dislocation, and blood samples were obtained by cardiac puncture. Organs were removed, rinsed with 0.9% saline solution, blotted dry, weighed, and counted in an autogamma counter. Distribution data are presented as percent injected dose/g tissue (Table 1) and percent injected dose/organ (Table 2). For excretion studies, animals were housed in metabolism cages, and urine and feces were collected daily.

Synthesis of unlabeled compounds. The synthesis of 15-p-(iodophenyl)-6-tellurapentadecanoic acid is shown in Fig. 1. The unlabeled compounds (**1** to **7**) exhibited properties consistent with the proposed structures by chromatography, ir and uv spectroscopy, low- and high-resolution spectrometry, and proton nuclear magnetic resonance. The syntheses and physical and chemical properties of these compounds are described in detail in another report (*19*).

TABLE 1. DISTRIBUTION OF RADIOACTIVITY (% INJECTED DOSE/G OF TISSUE) IN RAT AT VARIOUS TIMES AFTER INTRAVENOUS ADMINISTRATION OF 15-(p-[^{125}I]IODOPHENYL)-6-TELLURAPENTADECANOIC ACID*

Tissue	Time after injection; Percent injected dose/g (range)						
	5 min	30 min	60 min	2 h	6 h	1 d	5 d
Heart	5.87 (5.30–6.45)	5.43 (4.35–5.98)	5.55 (4.01–7.27)	5.85 (4.01–7.56)	4.78 (3.89–5.33)	3.25 (3.02–3.41)	0.94 (0.77–1.13)
Blood	0.26 (0.24–0.27)	0.45 (0.38–0.50)	0.34 (0.28–0.40)	0.42 (0.32–0.57)	0.31 (0.29–0.32)	0.20 (0.18–0.21)	0.03 (0.03–0.034)
Lungs	1.14 (0.86–1.52)	1.25 (1.14–1.40)	1.48 (1.10–1.95)	1.57 (1.29–2.05)	1.10 (0.97–1.17)	0.89 (0.77–1.05)	0.33 (0.30–0.36)
Liver	9.57 (8.97–10.28)	7.78 (6.73–8.86)	7.69 (6.68–8.28)	8.38 (7.32–9.90)	8.37 (7.86–9.08)	6.24 (5.53–6.73)	1.28 (1.23–1.36)
Kidneys	1.02 (0.79–1.15)	1.25 (1.09–1.39)	1.41 (1.23–1.55)	1.37 (1.20–1.69)	1.12 (0.95–1.20)	0.70 (0.64–0.80)	0.20 (0.18–0.22)
Thyroid	1.52 (1.41–1.63)	1.51 (0.83–2.21)	1.48 (0.95–1.94)	3.51 (1.91–4.42)	5.89 (5.33–7.08)	22.78 (18.15–27.13)	30.25 (25.36–34.81)

* Mean and range values for four female Fischer rats. Other tissues that were analyzed include spleen, brain and intestines.

TABLE 2. DISTRIBUTION OF RADIOACTIVITY (% INJECTED DOSE/ORGAN) OF RADIOACTIVITY IN RAT AT VARIOUS TIMES AFTER INTRAVENOUS ADMINISTRATION OF 15-(p-[^{125}I]IODOPHENYL)-6-TELLURAPENTADECANOIC ACID*

Tissue	Time after injection; Percent injected dose/organ (range)						
	5 min	30 min	60 min	2 h	6 h	1 d	5 d
Heart	2.98 (2.80–3.35)	2.61 (2.19–2.91)	2.87 (2.09–3.80)	2.87 (2.60–3.48)	2.32 (1.66–2.65)	1.60 (1.40–1.73)	0.45 (0.38–0.52)
Blood	2.00 (1.89–2.09)	3.41 (2.65–3.93)	2.66 (2.24–3.24)	3.01 (2.33–3.99)	2.26 (2.23–2.31)	1.50 (1.37–1.62)	0.23 (0.22–0.25)
Lungs	0.97 (0.74–1.30)	1.05 (0.97–1.04)	1.01 (0.85–1.16)	1.29 (1.12–1.67)	0.85 (0.73–0.94)	0.70 (0.58–0.85)	0.24 (0.22–0.26)
Liver	50.94 (46.13–55.20)	39.28 (34.11–43.16)	39.03 (34.62–41.81)	44.03 (41.59–47.61)	46.39 (43.86–49.26)	34.28 (32.02–37.42)	6.46 (6.00–6.84)
Kidneys	1.25 (0.95–1.44)	1.52 (1.42–1.63)	1.73 (1.47–1.97)	1.53 (1.37–1.80)	1.25 (1.09–1.38)	0.81 (0.74–0.87)	0.23 (0.21–0.27)
Thyroid	0.02 (0.02–0.02)	0.02 (0.01–0.02)	0.02 (0.01–0.02)	0.04 (0.02–0.05)	0.07 (0.06–0.09)	0.26 (0.22–0.30)	0.32 (0.28–0.35)

* Mean and range values for four female Fischer rats. Other tissues that were analyzed include spleen, brain and intestines.

Synthesis of radiolabeled compounds. The numbering system for the radiolabeled compounds described in this section (^{125}I-**2**, -**6**, and -**7**) refer to the structures shown in Fig. 1.

1-Chloro-9-(p-[^{125}I]iodophenyl)nonane (^{125}I-2). The triazene intermediate (compound **1**, 20 mg, 0.06 millimole) was dissolved in acetone (2 ml) and added dropwise to a mixture of trifluoroacetic acid (70 mg, 0.6 millimole) and sodium [^{125}I]iodide (4.68 mCi, 7.5 mg, 0.05 millimole) stirred at 0–5°C. The mixture was stirred for 5 min at 0–5°C, diluted with H_2O, and extracted three times with ether. The combined ether extracts were washed thoroughly with water, dried over anhydrous Na_2SO_4, and the solvent removed under a stream of argon. The resulting oily residue was dissolved in 2 ml of petroleum ether and applied to a silicic acid column (acidic grade, 2 cm i.d.) slurried in petroleum ether. Fractions 20 ml in volume were eluted with petroleum ether and aliquots were taken for counting and TLC analysis. Fractions 14–20 were combined to give 1.17 mCi (25%) of 1-chloro-9-(p-[^{125}I]iodophenyl)-nonane (^{125}I-**2**). The product showed a single radioactive component that co-chromatographed with unlabeled 1-chloro-9-(p-iodophenyl)nonane, R_f 0.70 (2% ether petroleum ether).

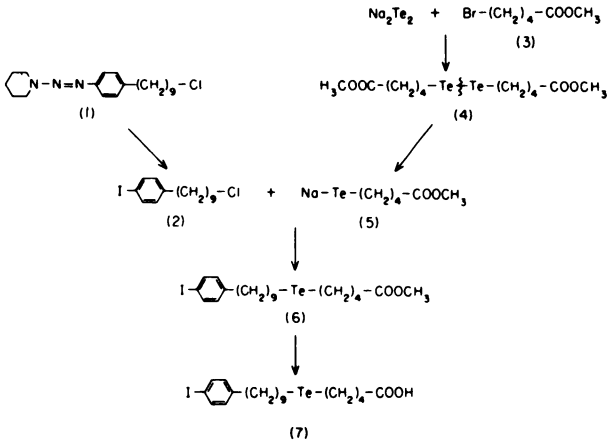

FIG. 1. Synthesis of 15-(p-iodophenyl)-6-tellurapentadecanoic acid.

Methyl-15-(p-[^{125}I]iodophenyl)-6-tellurapentadecanoate (^{125}I-6). The orange-colored *bis*-(methylvaleryl)-ditelluride (compound **4**, 48 mg, 0.10 millimole) was reduced under argon at room temperature with excess $NaBH_4$ in ethanol (10 ml) to a colorless solution of sodium (methylvaleryl)tellurol (compound **5**). A solution of 1-chloro-9-(p-[^{125}I]-iodophenyl)nonane (^{125}I-**2**; 1.17 mCi, 0.0125 millimole) in ethanol (5 ml) was added dropwise. The mixture was refluxed under argon for 1 hr, cooled in an ice bath, diluted with H_2O, and extracted three times with ether. The ether extracts were washed with H_2O, dried, and the solvent removed in the usual manner. The product was chromatographed on a silicic acid column (basic grade, 2 cm i.d.). Fractions 1 to 20 (20 ml each) were eluted with petroleum ether, and fractions 21 to 30 with C_6H_6. Fractions 24–27 were combined to give 1.1 mCi (80%) of methyl-15-(p-[^{125}I]iodophenyl)-6-tellurapentadecanoate (^{125}I-**6**). Upon TLC analysis, the product showed a single radioactive component that co-chromatographed in C_6H_6 with unlabeled **6** (R_f 0.45).

15-(p-[^{125}I]iodophenyl)-6-tellurapentadecanoic acid (^{125}I-7). The methyl ester (^{125}I-**6**; 1.1 mCi, 0.04 millimole) was refluxed under argon for 30 min in ethanol (6 ml) containing 2 ml of 1 *N* NaOH (2 millimole). After cooling, the solution was diluted with H_2O, the pH ad-

justed to 2-3 with 10% H_2SO_4, and the resulting cloudy solution extracted several times with ether. After washing with H_2O and drying over anhydrous Na_2SO_4, the solvent was evaporated under argon to give 715 μCi (65%) of 15-(p-[^{125}I]-iodophenyl)-6-tellurapentadecanoic acid, which showed a single radioactive component (R_f 0.50) on TLC analysis (8% MeOH/$CHCl_3$). The specific activity of ^{125}I-7 was 94 mCi/millimole.

RESULTS AND DISCUSSION

The synthesis of 15-(p-iodophenyl)-6-tellurapentadecanoic acid (Fig. 1) involves the preparation of the intermediate piperidyltriazene derivative of 1-chloro-9-(p-aminophenyl)nonane (**1**). Following a modification of a recently published procedure (*20*), we have treated the triazene (5-10 millimole) with HI-trifluoroacetic acid in acetone, with the rapid (<5 min) high-yield (>60%) formation of the 1-chloro-9-(p-iodophenyl)-nonane (**2**). The p-iodophenyl-substituted fatty acid (**7**) is then prepared by the steps outlined in Fig. 1. Since triazene derivatives such as **1** are generally stable substances that can be stored indefinitely, this regiospecific route for the introduction of the iodine in the para position may be more useful than methods that give isomeric mixtures requiring subsequent separation (*8-10*). Although the yield of the H[^{125}I]-triazene decomposition reaction has not yet been optimized in these preliminary studies, use of this approach allows the preparation of appreciably higher specific activities, which will place the expected human dose (250-500 μCi) well below toxic levels. An additional advantage of this route is the potential consumption of all of the radioiodide during this step. The excess triazene is easily removed from the iodophenyl product by simple chromatography. We are also using the triazene route for the preparation of 15-(p-iodophenyl)pentadecanoic acid (*8,9*) and other radioiodinated new methyl-branched fatty acids so that these agents will be available for comparative biological evaluation with the radioiodinated 15-(p-iodophenyl)-6-tellurapentadecanoic acid. An additional goal of future studies will be to determine the feasibility of preparing the piperidyltriazene derivative of 15-(p-aminophenyl)-6-tellurapentadecanoic acid, since its treatment with radioiodide would give the 15-(p-iodophenyl)-6-tellurapentadecanoic acid directly.

In Table 1 the tissue distribution of radioactivity in rats after intravenous administration of a bovine serum-albumin complex of the ^{125}I-7 is summarized for various time periods from 5 min to 5 days. For comparison, the data for % injected dose/organ are shown in Table 2. The rapid and pronounced myocardial uptake observed with this agent is analogous to that reported earlier for 9-[^{123m}Te]-HDA (*11-15*). The prolonged retention of radioactivity is a unique feature exhibited by 15-(p-iodophenyl)-6-tellurapentadecanoic acid and several other tellurium fatty acids (*12,13*). These studies illustrate that the terminal p-iodophenyl moiety does not interfere with the myocardial specificity observed with similar agents. After 6 hr the heart retained 80% of the maximum uptake observed after 5 min (Table 1). The radioactivity in the heart retained 55% of the maximal value after 24 hr. The mean heart-to-blood ratios were 22:1 at 5 min and 17:1 at 2 hr. Only marginal radioactivity accumulated in the thyroid tissue; 1.52% dose/g after 5 min and 5.84% dose/g after 6 hr. The minimal thyroid radioactivity (Table 1) and low blood levels demonstrate that the attachment of the radioiodine to the phenyl ring is an effective means of stabilizing the iodine and overcoming facile in vivo cleavage.

The rats used in the study were housed in metabolism cages and liquid and solid wastes were collected daily after injection of 15-(p-[^{125}I]-iodophenyl)-6-tellurapentadecanoic acid to determine the biological half-life of this new agent and its relative excretion in urine and feces. These studies were conducted over a 5-day period, which represents a decay period of 9 half-lives for 13.2-hr iodine-123. The cumulative excretion levels in urine and feces were 43.2 ± 1.8% injected dose after 2 days and 71.6 ± 8.3% after 5 days. The radioactive content of the urine (20.8 ± 2.1%) and feces (22.4 ± 1.1%) were similar after 2 days, but after five days the cumulative fecal activity (41.4 ± 4.3%) was greater than that for urine (30.5 ± 4.0%).

The pronounced heart uptake, minimal deiodination, rapid blood clearance, and prolonged myocardial retention of 15-(p-[^{125}I]iodophenyl)-6-tellurapentadecanoic acid suggest that the ^{123}I-labeled analog is an attractive agent with which to evaluate myocardial perfusion. Radioiodinated 15-(p-iodophenyl)-6-tellurapentadecanoic acid is currently being evaluated in normal dogs and dogs with partial coronary artery ligation to determine the extraction values and relative washout properties in normal and ischemic tissue. In these studies the radioiodinated fatty acid will be compared with thallium-201 as a myocardial perfusion agent. In addition, the correlation of uptake and retention of the fatty acid with regional fatty-acid metabolism will be assessed.

FOOTNOTES

* Analtech, Inc.
† Fischer rats, ORNL Biology Division Breeding Colony.

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REFERENCES

1. ROBINSON GD, JR: Synthesis of ^{123}I-16-iodo-9-hexadecenoic

acid and derivatives for use as myocardial perfusion imaging agents. *Int J Appl Radiat Isot* 28:149-156, 1977
2. MACHULLA H-J, STOCKLIN G, KUPFERNAGEL C, et al: Comparative evaluation of fatty acids labeled with C-11, Cl-34m, Br-77, and I-123 for metabolic studies of the myocardium: Concise communication. *J Nucl Med* 19:298-302, 1978
3. OTTO CA, BROWN LE, WIELAND DM, et al: Radioiodinated fatty acids for myocardial imaging: Effects of chain length. *J Nucl Med* 22:613-618, 1981
4. POE ND, ROBINSON GD, JR, GRAHAM LS, et al: Experimental basis for myocardial imaging with ^{123}I-labeled hexadecenoic acid. *J Nucl Med* 17:1077-1082, 1976
5. POE ND, ROBINSON GD, JR, ZIELINSKI FW, et al: Myocardial imaging with ^{123}I-hexadecenoic acid. *Radiology* 124:419-424, 1977
6. FREUNDLIEB CH, HOCK A, VYSKA K, et al: Myocardial imaging and metabolic studies with [17-^{123}I]iodoheptadecanoic acid. *J Nucl Med* 21:1043-1050, 1980
7. FEINENDEGEN LE, VYSKA K, FREUNDLIEB CH, et al: Non-invasive analysis of metabolic reactions in body tissues, the case of myocardial fatty acids. *Eur J Nucl Med* 6:191-200, 1981
8. MACHULLA H-J, MARSMANN M, DUTSCHKA K: Biochemical concept and synthesis of a radioiodinated phenyl fatty acid for in vivo metabolic studies of the myocardium. *Eur J Nucl Med* 5:171-173, 1980
9. MACHULLA H-J, MARSMANN M, DUTSCHKA K, et al: Radiopharmaceuticals. II. Radiobromination of phenylpentadecanoic acid and biodistribution in mice. *Radiochem Radioanal Lett* 42:243-250, 1980
10. DUDCZAK R, ANGELBERGER P, WAGNER-LÖFFLER M, et al: Radioiodinated phenylfatty acid for myocardial imaging. *J Nucl Med* 22:P81, 1981 (abst)
11. KNAPP FF, JR, AMBROSE KR, CALLAHAN AP, et al: Tellurium-123m-labeled isosteres of palmitoleic and oleic acids show high myocardial uptake. Proceedings, 2nd International Symposium on Radiopharmaceuticals II, Society of Nuclear Medicine, New York, 1979, pp 101-108
12. KNAPP FF, JR: Selenium and tellurium as carbon substitutes in *Radiopharmaceuticals: Structure-Activity Relationships*, Proceedings of the International Symposium, Chapter 16, Grune & Stratton, New York, 1980, pp 345-391
13. KNAPP FF, AMBROSE KR, CALLAHAN AP, et al: Effects of chain length and tellurium position on the myocardial uptake of Te-123m fatty acids. *J Nucl Med* 22:988-993, 1981
14. ELMALEH DR, KNAPP FF, YASUDA T, et al: Myocardial imaging with 9-[Te-123m]telluraheptadecanoic acid. *J Nucl Med* 22:994-999, 1981
15. OKADA RD, KNAPP FF, JR, ELMALEH DR, et al: Tellurium-123m-labeled-9-telluraheptadecanoic acid: A possible cardiac imaging agent. *Circulation* 65:305-310, 1982
16. KNAPP FF, JR, VEST M, ELMALEH DR, et al: Myocardial uptake of 10-[^{14}C]-9-telluraheptadecanoic acid (10-[^{14}C]-9-THDA): Evidence for retention of the alkyl region of 9-THDA. *J Nucl Med* 22:P74, 1981 (abst)
17. GOODMAN MM, KNAPP FF, JR, CALLAHAN AP, et al: Synthesis and biological evaluation of 17-[^{131}I]-iodo-9-telluraheptadecanoic acid—A potential myocardial imaging agent. *J Med Chem* 25:613-618, 1982
18. GOODMAN MM, KNAPP FF, JR, STRAUSS HW, et al: Prolonged myocardial retention of 15-phenyl-6-tellurapentadecanoic acid: A new agent for potential labeling with Iodine-123. *Clin Nucl Med* 6:P157, 1981 (abst)
19. GOODMAN MM, KNAPP FF JR: Synthesis of 15-(p-Iodophenyl)-6-tellurapentadecanoic acid. 47:3004-3006, 1982—A new myocardial imaging agent. *J Org Chem* (in press)
20. FOSTER NI, HEINDEL ND, BURNS HD, et al: Aryl iodides from anilines via triazene intermediates. *Synthesis* 572, 1980