Tumor-Localizing Agents X: Radioiodinated Hypoglycemic Agents

G. N. HOLCOMB*‡, C. M. BOYD*, R. E. COUNSELL*, W. H. BEIERWALTES*, R. A. SZCZESNIAK†, D. R. K. MURTY†, and G. A. BRUNO†

Abstract □ This study was undertaken to find a pancreas-specific photoscanning agent. 1-(*p*-Iodobenzenesulfonyl)-3-*n*-propylurea-^{125}I was synthesized for this purpose, since it contains a gamma-emitting radionuclide and there seemed to be a reasonable possibility that a sulfonylurea such as this might concentrate in the pancreas to a greater extent than in other tissues and organs. Tissue-distribution studies with this compound showed that it has no particular predilection for the pancreas of dogs.

Keyphrases □ Tumor-localizing agents—radioiodinated hypoglycemic agents □ 1-(*p*-Iodobenzenesulfonyl)-3-*n*-propylurea-^{125}I—synthesis □ Tissue distribution—radioiodinated hypoglycemic agent □ TLC—separation, identification □ IR spectrophotometry—identification □ NMR spectroscopy—identification □ Scintillometry—analysis

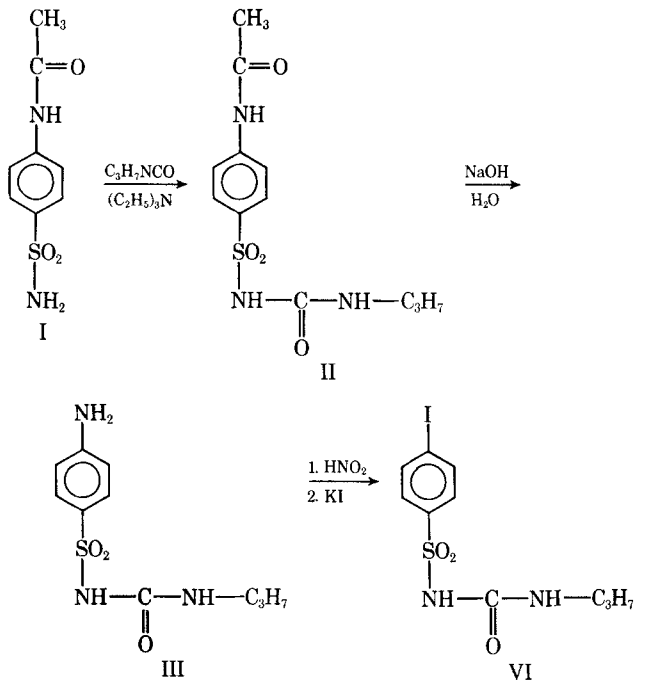
This research is part of a continuing program to develop new radioscanning agents for selected tumors. The present study was undertaken to find an agent that would selectively concentrate in the pancreas. Such a compound, when labeled with a gamma-emitting radionuclide, could be used in conjunction with external scintillation scanners and cameras for the diagnosis of pancreatic carcinoma. This type of carcinoma is particularly difficult to diagnose by existing methods (1).

The task of designing an agent to accomplish this purpose was somewhat difficult because very few substances are known to concentrate specifically in pancreatic tissue. A number of ^{14}C- and ^{35}S-labeled amino acids have been shown to concentrate in the pancreas following their intravenous administration (2–4). These substances do not, however, contain gamma-emitting radionuclides and cannot be used for external scanning purposes. Further studies along these lines, however, indicated that replacement of the sulfur of methionine with gamma-emitting ^{75}Se could be accomplished without destroying the amino acid's predilection for the pancreas (5). Subsequent clinical evaluations of 1-selenomethionine-^{75}Se resulted in mixed conclusions regarding its value as a pancreatic scanning agent (6, 7). More recently, it was found that certain radioiodinated phenylalanine analogs also display a high degree of specificity for concentrating in the pancreas of mice (8–10), but no selective localization could be achieved in dogs or monkeys (11). Consequently, studies with radioiodinated phenylalanine derivatives were discontinued.

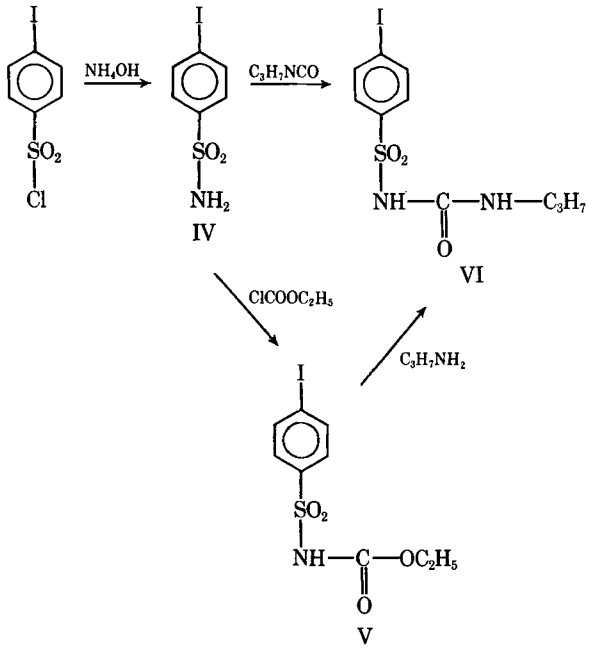
As a continuation of studies in this area, 1-(*p*-iodobenzenesulfonyl)-3-*n*-propylurea-^{125}I was selected as a compound that might prove useful as a pancreatic scanning agent. This compound is structurally similar to chlorpropamide [1-(*p*-chlorobenzenesulfonyl)-3-*n*-propylurea] and other sulfonylurea hypoglycemic agents. Although the sulfonylureas are known to have extrapancreatic effects, it is generally agreed that they exert their effect primarily by stimulating the release of insulin from the pancreatic beta cells (12). Accordingly, it seemed quite possible that these agents might concentrate in pancreatic tissue to a greater extent than in other tissues and organs. Relatively little research has been carried out on the tissue distribution of sulfonylureas. It has been reported that ^{35}S-carbutamide [1-(*p*-aminobenzenesulfonyl)-3-*n*-butylurea-^{35}S] concentrates to a greater extent in the liver than in other organs (13). However, some evidence indicates that carbutamide may differ from other sulfonylureas in this regard (14).

CHEMISTRY

1-(*p*-Iodobenzenesulfonyl)-3-*n*-propylurea (VI) was initially synthesized by a series of reactions which terminated in a diazotization step to introduce the iodine. This approach was taken because it was felt that the diazotization reaction would provide a convenient means of introducing the ^{125}I into the molecule. The starting material (I) was obtained by the chlorosulfonation of acetanilide (15), followed by reaction of the product with ammonia (16). Compound I was converted to II by reaction with *n*-propyl isocyanate. The *N*-acetyl group was removed by base-catalyzed hydrolysis. Iodine was introduced by diazotizing III and reacting the diazonium salt with potassium iodide to yield VI (Scheme I). The yield for the diazotization reaction was poor, thus making it seem impractical to introduce the ^{125}I in this manner. Accordingly, other methods for the synthesis of VI were explored. The most direct method involved treatment of *p*-iodobenzenesulfonyl chloride with ammonia to form the sul-



Scheme I



Scheme II

fonamide (IV), which was converted to VI by reaction with *n*-propyl isocyanate (Scheme II). An alternate method utilizes the formation of the intermediate sulfonylcarbamate (V) which was prepared from IV by refluxing with ethyl chloroformate. Conversion of V to VI proceeded smoothly by warming V with an excess of *n*-propylamine. The final products (VI) prepared by all three methods were identical.

Attempts to effect an isotope exchange on VI were unsuccessful; it underwent decomposition to *p*-iodobenzenesulfonamide when heated to 170° in any of a wide variety of solvents. Therefore, the isotope exchange was carried out on IV to produce *p*-iodobenzenesulfonamide-^{125}I, which was then converted to 1-(*p*-iodobenzenesulfonyl)-3-*n*-propylurea-^{125}I by reaction with *n*-propyl isocyanate.

TISSUE-DISTRIBUTION STUDIES

1-(*p*-Iodobenzenesulfonyl)-3-*n*-propylurea-^{125}I (100 μc.) was injected intravenously into four adult beagle dogs. The dogs were sacrificed at the end of 30 min. or 4, 24, and 72 hr., respectively. Other details pertaining to this phase of the study have been reported (17).

Data presented in Table I show that the desired uptake by the pancreas was not obtained. However, autoradiographic studies revealed that most of the radioactivity present in the pancreas was associated with the islet cells (17). Since the islet cells comprise only 1–4% of the total pancreatic weight (18), the concentration of drug in the islet cells could actually be quite high. Accordingly, this compound may have potential as a scanning agent for certain pancreatic islet cell tumors. Studies are currently in progress to assess its value for this purpose.

EXPERIMENTAL[1]

Melting points were taken on a Fisher-Johns melting-point apparatus and are corrected. IR spectra were taken on a Perkin-Elmer 337 spectrophotometer. The NMR spectra were obtained with a Varian A-60 spectrometer in $CDCl_3$ at a concentration of 10% with trimethylsilane as an internal reference. TLC was run with 2.54-cm. (1-in.) wide Eastman chromatograms, type K301R, with fluorescence indicator, and spots were detected with UV light. Chromatograms of radioiodinated compounds were scanned with an Atomic Associates RCS-363 radiochromatogram scanner. Sodium iodide-^{125}I in 0.05 *M* sodium bisulfite was purchased[2]; the specific activity was 0.17 mc./mg. of iodine. The specific activity of the final compound was determined with a Picker isotope calibrator.

[1] Elemental analyses were performed by Spang Microanalytical Laboratories, Ann Arbor, Mich.

[2] New England Nuclear Corp.

Table I—Tissue Distribution of 1-(*p*-Iodobenzenesulfonyl)-3-*n*-propylurea-^{125}I in the Dog[a]

Tissue	30 min.	4 hr.	24 hr.	72 hr.
Pancreas	0.008	0.009	0.019	0.017
Blood	0.079	0.044	0.056	0.060
Liver	0.018	0.026	0.031	0.021
Spleen	0.043	0.045	0.081	0.088
Kidney	0.024	0.027	0.051	0.031
Thyroid	0.006	0.006	0.016	0.171
Muscle	0.005	0.008	0.014	0.012
Fat	0.002	0.005	0.010	0.003

[a] Values are expressed as percent of dose per gram of tissue.

1-(*p*-Acetamidobenzenesulfonyl)-3-*n*-propylurea (II)—A solution of I (20 g., 0 1 mole) and triethylamine (3 ml.) in *n*-propylisocyanate (50 ml., 0.5 mole) was heated on a steam bath for 14 hr. and was then poured into 10% AcOH (250 ml.). The product precipitated as a yellow oil, which was dissolved in 10% NaOH (50 ml.). The pH of the solution was adjusted to 5 by the addition of HCl, and II precipitated as a solid. Recrystallization (EtOH–H_2O) afforded pure II (14 g., 50%), m.p. 197–198° [lit. (19) m.p. 198°]. The IR spectrum was as expected.

1-(*p*-Aminobenzenesulfonyl)-3-*n*-propylurea (III)—1-(*p*-Acetamidobenzenesulfonyl)-3-*n*-propylurea (II) (12 g., 0 04 mole) was dissolved in 15% NaOH (75 ml.), and the solution was refluxed for 3 hr. The product precipitated as a yellowish solid when the pH was adjusted to 5 with HCl. It was recrystallized (H_2O) to give 9 g. (88%) of III, m.p. 140–141° [lit. (20) m.p. 141°]. The IR spectrum was as expected.

***p*-Iodobenzenesulfonamide (IV)**—A solution of *p*-iodobenzenesulfonyl chloride (20 g., 0.065 mole) in NH_4OH (800 ml.) was refluxed with stirring for 3 hr. The hot reaction mixture was filtered, and the filtrate was placed in a refrigerator overnight. The product precipitated and was isolated by filtration. Recrystallization (EtOH–H_2O) produced pure IV (17 g., 91%), m.p. 190–191° [lit. (21) m.p. 189°]. The IR spectrum was as expected.

Ethyl *p*-Iodobenzenesulfonylcarbamate (V)—A mixture of IV (2.8 g., 0.01 mole) and anhydrous K_2CO_3 (3.6 g., 0.025 mole) in acetone (12 ml.) was stirred while ethyl chloroformate (1.25 ml., 0.013 mole) was added dropwise. The mixture was refluxed with stirring for 2 hr. A second portion of acetone (20 ml.) was added, and heating and stirring were continued for an additional 30 hr. The reaction was cooled to room temperature and filtered; the filtrate was acidified with 4.2 ml. of HCl, and an oil separated. The supernatant was decanted and the oil rubbed under water. The oil solidified on standing. Recrystallization (EtOAc–petroleum ether) produced pure V (1.54 g., 43%), m.p. 81–82°. The IR and NMR spectra were as expected.

Anal.—Calcd. for $C_9H_{10}INO_4S$: C, 30.43; H, 2.84; N, 3.94. Found: C, 30.60; H, 2.86; N, 3.83.

1-(*p*-Iodobenzenesulfonyl)-3-*n*-propylurea (VI)—*Procedure A*—*p*-Iodobenzenesulfonamide (IV) (300 mg., 0.001 mole), triethylamine (0.05 ml.), and *n*-propyl isocyanate (0.1 ml., 0.001 mole) were dissolved in anhydrous dimethylformamide (0.5 ml.) and sealed in a thick-walled test tube. The mixture was heated at 90° for 12 hr. The tube was opened and the reaction mixture was diluted with 10% AcOH (10 ml.). The product was collected by filtration and dissolved in 10% NaOH (10 ml.). The solution was filtered and the pH was adjusted to 5 with HCl. The resulting white solid was recrystallized (EtOH–H_2O) to furnish 230 mg. (60%) of VI, m.p. 155–156°. The IR and NMR spectra were as expected.

Anal.—Calcd. for $C_{10}H_{13}IN_2O_3S$: C, 32.62; H, 3.56. Found: C, 32.50, H, 3.57.

Procedure B—Ethyl *p*-iodobenzenesulfonylcarbamate (V) (400 mg., 0.001 mole) was dissolved in *n*-propylamine (0.4 ml., 0.05 mole) and shaken for 1.5 hr. The excess amine was removed under reduced pressure, and the residue was heated at 110° under reduced pressure (35 mm.) for 0.5 hr. An oil resulted. Recrystallization (EtOH–H_2O) furnished pure VI (195 mg., 47%), m.p. 155–156°. The IR spectrum was as expected.

Anal.—Calcd. for $C_{10}H_{13}IN_2O_3S$: C, 32.62; H, 3.56; I, 34.47; N, 7.61; Found: C, 32.62; H, 3.48; I, 34.77; N, 7.61.

Procedure C—1-(*p*-Aminobenzenesulfonyl)-3-*n*-propylurea (III) (7.7 g., 0.03 mole) was dissolved in HCl (15 ml.), and the solution was cooled to −5° with an ice–salt bath. A solution of $NaNO_2$ (2.4 g., 0.05 mole) in H_2O (10 ml.) was added dropwise at a rate that maintained the temperature of the reaction between 0 and 5°. Stirring was continued for an additional 15 min., and then the reaction mixture was filtered through glass wool into a solution of KI (18 g., 0.1 mole) in H_2O (60 ml.). A brown resin precipitated which was dissolved in 10% NaOH (100 ml.). The product precipitated as a yellowish solid when the pH of the solution was adjusted to 5 with HCl. Several recrystallizations (MeOH–H_2O) gave pure VI (1.5 g., 14%), m.p. 155–156°. A mixed melting point of this compound with the product from *Procedure A* showed no depression.

p-Iodobenzenesulfonamide-^{125}I—*p*-Iodobenzenesulfonamide (IV) (100 mg.) was treated with Na-^{125}I (3 mc.) in ethylene glycol (2 ml.) at 170° for 12 hr. as described previously (22). The radioiodinated product was recrystallized (EtOH–H_2O) to give a pure product (75 mg.), which was shown by TLC to be identical with the unlabeled material (IV): R_f = 0.91 in EtOAc–MeOH (4:1). A radiochromatogram of the strip showed that the radioactivity was present in the same spot.

1-(p-Iodobenzenesulfonyl)-3-n-propylurea-^{125}I—*p*-Iodobenzenesulfonamide-^{125}I (75 mg., 0.3 mmole), triethylamine (0.05 ml.), and *n*-propyl isocyanate (0.05 ml., 0.04 mmole) were dissolved in anhydrous dimethylformamide (0.5 ml.) and placed in a 10-ml. round-bottom flask equipped with a magnetic stirrer and a reflux condenser. The reaction was heated at 70° with stirring for 10 hr. and then diluted with H_2O (10 ml.). The product was collected by filtration and washed well with water. Recrystallization (EtOH–H_2O) gave a pure product (50 mg., 53%), which was identical with the unlabeled material (VI) on TLC in EtOAc–MeOH (4:1), R_f = 0.74. Radiochemical purity was established by TLC. The specific activity was 24 μc./mg., representing 80% exchange.

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ACKNOWLEDGMENTS AND ADDRESSES

Received August 31, 1970, from the * *Laboratory of Medicinal Chemistry, College of Pharmacy and Department of Internal Medicine (Nuclear Medicine), University of Michigan, Ann Arbor, Mich.*, and the † *Radiopharmaceutical Research Laboratories, E. R. Squibb and Sons, New Brunswick, N. J.*

Accepted for publication October 20, 1970.

After the completion of studies at the University of Michigan, it came to the attention of the Michigan researchers that similar investigations had been conducted in the Squibb Laboratories. This paper represents a joint publication from both laboratories.

This research was supported by grants from The Squibb Institute for Medical Research, the National Institutes of Health (CA-08349), and the American Cancer Society (PRA-18).

The authors thank Dr. L. M. Lieberman for his interest and suggestions.

‡ Present address: Department of Medicinal Chemistry, Ferris State College, Big Rapids, Mich.