^{125}I-LABELED 1-(2'-ACETYL-4'-[ORTHO-IODOBENZOYL]-AMINOPHENOXY)-3-ISOPROPYL-AMINOPROPAN-2-OL

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SUMMARY

The preparation of the iodine-125 labeled beta adrenergic antagonist, 1-(2'-acetyl-4'-[ortho-iodobenzoyl]-aminophenoxy)-3-isopropylamino-propan-2-ol, suitable for biodistribution studies is described. Synthesis of the unlabeled precursor proceeds in a 44% yield and the iodine-125 exchange results in 89% incorporation of label. The final product is radiochemically pure (98-99%) with a specific activity of 3.2 Ci/mmol.

Key Words: Beta Adrenergic Antagonist, Iodine-125

INTRODUCTION

The recent availability of iodine-125 labeled beta adrenergic agonists and antagonists has been of great benefit in the identification and characterization of the beta adrenergic receptor (1,2,3). Investigators have been primarily concerned with the receptors present in isolated tissue preparations (4,5,6) and cell lines (7) and therefore they have emphasized the specific activity and receptor affinity of the agents as opposed to the *in vivo* radiochemical stability or organ selectivity. Radioiodination of hydroxyphenyl or hydroxybenzyl analogs of propranolol, isoproterenol, pindolol and alprenolol has resulted in virtually carrier-free agents which retain high receptor affinity and possess reasonable *in vitro* stability (2-3 weeks). However, the placement of the label ortho to

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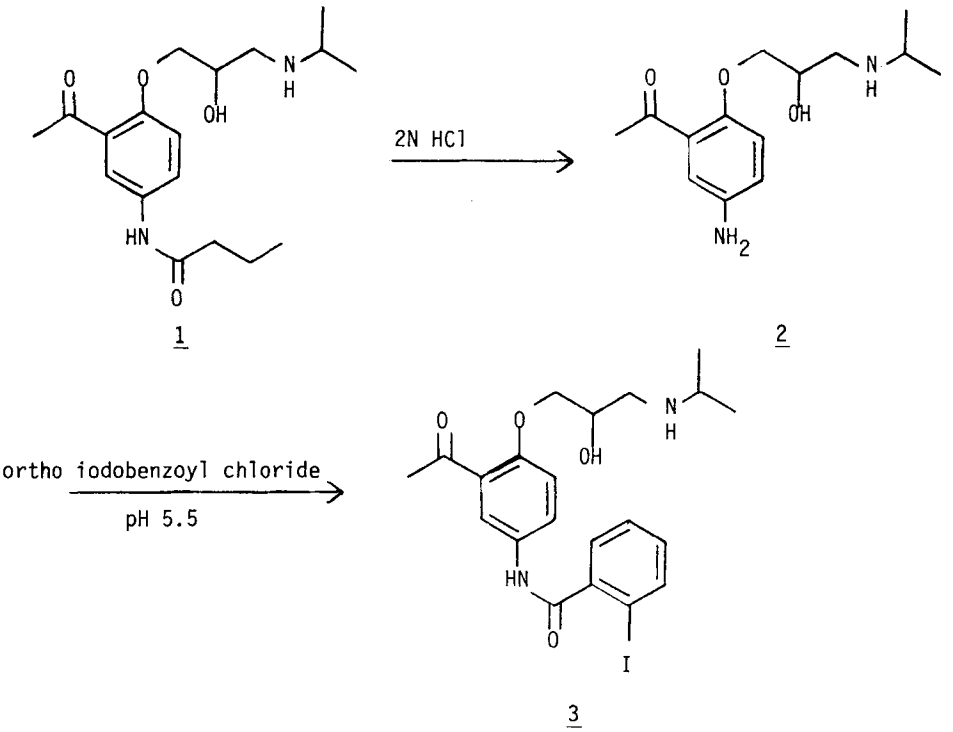
the phenolic hydroxyl leads to significant deiodination in vivo and would be expected to result in high accumulation of the radioisotope in the thyroid (8). Because our interest is the preparation of organ specific radiodiagnostic agents, myocardial selectivity (beta-1 adrenergic receptors) and resistance to in vivo deiodination must be the primary attributes of our radiolabeled iodinated beta adrenergic antagonists.

We wish to describe the synthesis and radioisotopic labeling of an iodobenzoyl analog of the cardioselective beta adrenergic antagonist acebutolol (9). The nonlabeled compound was prepared by hydrolysis of acebutolol, 1, in 2N hydrochloric acid. Reacylation of the aminophenoxy intermediate 2 with ortho-iodobenzoyl chloride followed by column chromatographic separation gave the pure 1-(2'-acetyl-4'-[iodobenzoyl]-aminophenoxy)-3-isopropylaminopropan-2-ol, 3, in 44% yield. Conversion to the hydrochloride salt provided the precursor for the radioisotope exchange procedure, and isotopic exchange was achieved in 89% yield by heating a sealed serum vial containing a mixture of the hydrochloride salt, carrier free sodium iodide-^{125}I and pH 6.0-6.5 buffer at 130° C for 90 minutes (10).



1 2 3

Extraction of the product from the mixture gave a radiopharmaceutical with 98-99% radiochemical purity and a specific activity of 3.2 Ci/mmol. Adjustment of the reactant ratios in the exchange step would readily permit increases in the specific activity by a factor of 10 or more. In addition, this scheme could be easily adapted to the use of the iodine-123 or -131 nuclide.

MATERIALS AND METHODS

The following materials were obtained from the sources indicated: sodium iodide-^{125}I (New England Nuclear Corporation, carrier-free, 10 mCi in 80 µl 0.08 N sodium hydroxide); acebutolol hydrochloride (provided by May and Baker, Ltd.); ortho-iodobenzoyl chloride (Eastman Chemicals); silica gel and alumina tlc sheets (Eastman Chromagram with UV indicator 13181 and 13252); silica gel tlc plates (Analtech silica GF60 plates with indicator).

Radioactive counting was carried out using a NaI (Tl) gamma well scintillation counter. Melting points were determined using a Thomas-Hoover melting point apparatus and are uncorrected. NMR spectra were recorded on a Varian T-60 spectrometer in d_6-dimethyl sulfoxide and using tetramethylsilane as an internal standard. IR and UV spectra were recorded using Perkin-Elmer Model 711 and Cary 17 spectrophotometers, respectively. Elemental analyses (C, H, N, I) were performed by Schwarzkopf Microanalytical Laboratory, Woodside, N.Y. and agreed to within ± 0.4% of theoretical values unless otherwise stated. Radiochemical purity determinations were carried out with three tlc systems: system A - silica gel sheets and chloroform-methanol (85:15); system B - alumina sheets and chloroform-methanol (95:5); system C - silica gel plates and butanol: acetic acid: water (4:1:2). For the determination of radiochemical purity the radiolabeled sample was applied adjacent to the nonlabeled compound. After the chromatogram had been developed for 6 cm, the strips were visualized under UV light (254 nm), cut into 0.5 cm wide pieces and counted.

1-(2'-Acetyl-4'-[ortho-iodobenzoyl]-aminophenoxy)-3-isopropylaminopropan-2-ol 3. A solution of acebutolol hydrochloride 1 (5.0 mmol) in 30 ml 2 N hydrochloric acid was refluxed for 2 hours, the solvent removed under reduced pressure and the resulting colorless residue was redissolved in 50 ml 0.1 M disodium phosphate.

The solution was adjusted to pH 5.5 by the addition of 0.2 N sodium hydroxide, cooled to 0° and ortho-iodobenzoyl chloride (6.0 mmol) in 3 ml benzene was added dropwise over 15 minutes. The solution was readjusted to pH 5.5 and stirred at 0° C for 2 hours, warmed to room temperature, brought to pH 10 and extracted with chloroform-isopropanol (3:1). The organic layer was dried over magnesium sulfate (anhydrous), filtered, and concentrated to an oil which was purified by silica gel chromatography. The desired product was eluted with chloroform-ethanol (7:3), and after removal of solvent, the free base was crystallized from ethyl acetate. Yield 44%, m.p. 152-156°C. The hydrochloride salt was prepared by the passage of hydrogen chloride into an ethyl acetate solution of the product, m.p. 209-212°C. R_f solvent system A = 0.30, B = 0.42, C = 0.48.

IR (KBr): Broad absorption of O-H and N-H centered at 3250 cm^{-1}, acetyl and benzoyl carbonyl absorptions at 1680 and 1660 cm^{-1}. NMR (D_6-DMSO): δ 0.98 (6H, doublet, J - 6 Hz); 2.62 (3H, singlet); 2.70 (1H, heptet, J - 6 Hz); 3.30 (2H, broad); 4.02 (3H, broad); 7.06-7.57 (4H, overlapping multiplets); 7.72-8.02 (3H, overlapping multiplets).

Elemental Analysis: $C_{21}H_{25}N_2O_4I$ (C, H, N, I), Calc'd for I 25.57; Found 25.03.

Radioisotope exchange. To sodium iodide-^{125}I (7.0 mCi) in 60 µl 0.08 N sodium hydroxide in a serum vial were added 100 µl of an aqueous solution containing 1.0 mg (2 µmol) of 3 as the hydrochloride salt. To the mixture was added 0.1 N hydrochloric acid until a clear solution was formed. The solution was back titrated with 0.1 N sodium hydroxide until the iodinated beta antagonist began to precipitate (pH 6.0-6.5). The vial was flushed with nitrogen, sealed and then heated at 130°C for 90 minutes. The vial was cooled and the reaction mixture was brought to pH 10 by the addition of 0.1 N sodium hydroxide. The mixture was extracted (3 x 2 ml) with chloroform-isopropanol (3:1), dried over magnesium sulfate (anhydrous), filtered and evaporated to dryness. The residue was redissolved in a small volume of 0.1 N hydrochloric acid, adjusted to pH 6.0 with phosphate buffer and diluted to the desired activity with physiologic saline. This solution was stable for 3-4 weeks at -4°C.

The yield of iodine-125 labeled 3 was 6.4 mCi (3.2 mCi/mmol) with 89% incorporation and 98+% radiochemical purity.

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