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Synthesis of 6-n-Propyl-2-Thiouracil-6-^{14}C

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SUMMARY

6-n-Propyl-2-thiouracil-6-^{14}C was prepared from sodium butyrate-1-^{14}C, with a radiochemical yield of 34 %. Butyryl chloride-1-^{14}C was prepared from butyric acid-1-^{14}C and condensed with sodio ethylacetoacetate to yield ethyl β-oxohexanoate-β-^{14}C. The β-keto ester was condensed with thiourea in sodium ethoxide to yield the 2 substituted thiouracil.

INTRODUCTION.

The antithyroid drug 6-n-propyl-thiouracil is used in the management of hyperthyroid states. Pharmacological activity of the compound depends on an intact ring structure which contains sulphur and resembles thiocarbamide. Relative rates of metabolism of ^{35}S labelled drug have been estimated in man by measuring the rate of decrease of plasma ^{35}S [1]. However, an accurate measure of the rate of metabolism of a drug requires a knowledge of the mode of detoxication, if any. Thus a drug may be transformed, without loss of radioactive label, to an inactive metabolite, and a study of the time course of disappearance of drug, based on radioactivity measurements, would not give a realistic measure of rate of metabolism. The mode of detoxication, or metabolism, of propylthiouracil has not been determined. The metabolites of this drug would be difficult to identify and estimate in biological fluids. By analogy to thiouracil [2] it might be predicted that sulphur could be cleaved from the molecule and leave a residue that could ultimately be metabolised to β-norleucine. In a study planned to elucidate the pattern of biotransformation, a ^{14}C label was incorporated at a position that we consider to be the most stable, metabolically. It is anticipated therefore that the label will remain in many of the possible metabolites.

The synthetic route used (Fig. 1) was adapted from a variety of reported methods that were considered to be most suitable for conserving radioactive

label. The original synthetic route [3] was unsuitable, as we could not reproduce the reported yield (39 %) of the β-keto ester. Our yield by this method was 16 %.

Butyryl chloride was made in good yield (76 %) by the method of H. C. Brown [4]. Ethyl β-oxohexanoate was prepared in poor yield (20 %) by our modification of the methods reported for this ester [3] and for the methyl ester of β-oxocaproic acid [5]. Condensation of the ester with thiourea proceeded with good yield (71 %) according to the method of Anderson *et al.* [3]. This scheme is summarised in Figure 1.

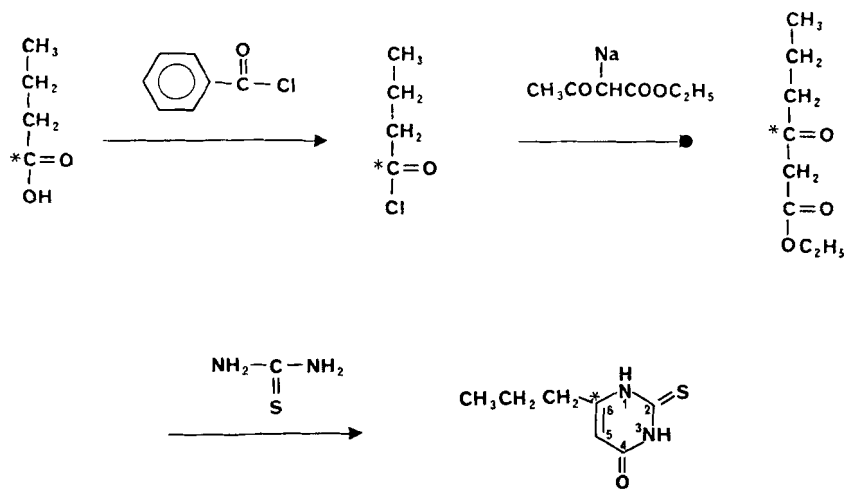

FIG. 1. Outline of synthesis of 6-n-Propyl-2-Thiouracil-6-^{14}C.

EXPERIMENTAL PROCEDURE.

Butyryl chloride-1-^{14}C.

Sodium butyrate-1-^{14}C (40 mg, 0.8 mCi obtained from Amersham-Searle, Toronto, Canada) was mixed with 8.8 g (0.1 mole) butyric acid and HCl gas passed into the solution for 10 min to ensure equilibration of the label. To this solution, in a 50 ml round bottom flask equipped with a short distillation column, was added in one portion 28.2 g (0.20 mole) benzoyl chloride, and butyryl chloride was distilled over as rapidly as possible. When no more distillate formed, 0.96 g (1 ml = 0.01 mole) butyric acid carrier and 2.8 g (0.02 mole) benzoyl chloride were added to the distillation flask and distillation recontinued until no more product could be collected. The yield of butyryl chloride was 9.0 g (76 %) b. range 100-104° C; reported, 101-102.5° C [4].

Ethyl β-oxohexanoate-β-^{14}C.

Sodio ethylacetoacetate was prepared by adding an ethereal solution of acetoacetic ester to small chips of sodium metal stirred in anhydrous ether [6]. The ether had been dried by standing reagent grade material over Linde 3A molecular sieve until it no longer reacted with sodium. The reaction was completed by refluxing for 2 hours and allowed to stand overnight. Sodio ethylacetoacetate was filtered, washed with anhydrous ether, and stored in a desiccator. Butyryl chloride-1-^{14}C, 9.0 g (0.08 mole), was added dropwise over a period of 18 min to a slurry of 12.8 g (0.08 mole) sodio ethylacetoacetate, stirred in 50 ml anhydrous ether. After standing overnight NaCl was removed by filtration and dry ammonia gas (20 g, 1.2 mole) was passed for 6 hours, into the ether solution cooled in ice. The reaction mixture was then allowed to stand overnight at room temperature. After washing with water this mixture was stirred with 30 ml dil HCl for 2 hours. The ether layer was then rewashed with water followed by 5 % sodium bicarbonate. The ether layer was evaporated and the residue extracted 5 times with 5 ml portions of saturated sodium bisulphite solution. After rewashing with water the residue was dissolved in ether and dried over anhydrous sodium sulphate. After decantation excess ether was removed by distillation and the resulting oil distilled under vacuum to yield 2.6 g (0.02 mole) ethyl β-oxohexanoate-β-^{14}C b. range 94-105° C at 15 mm Hg; reported, 93-94° C at 15 mm Hg [3]. Carrier ester (1 ml = 0.97 g) was added to the boiler flask and the distillation recontinued. The infrared spectrum of the distillate was identical to that of authentic material (Aldrich Chemical Co., Milwaukee, U. S. A.).

6-n-propyl-2-thiouracil-6-^{14}C.

Ethyl β-oxohexanoate-β-^{14}C (3.51 g, 0.02 mole) dissolved in 5 ml anhydrous ethanol (Linde 3A sieve) was added to a solution of sodium metal

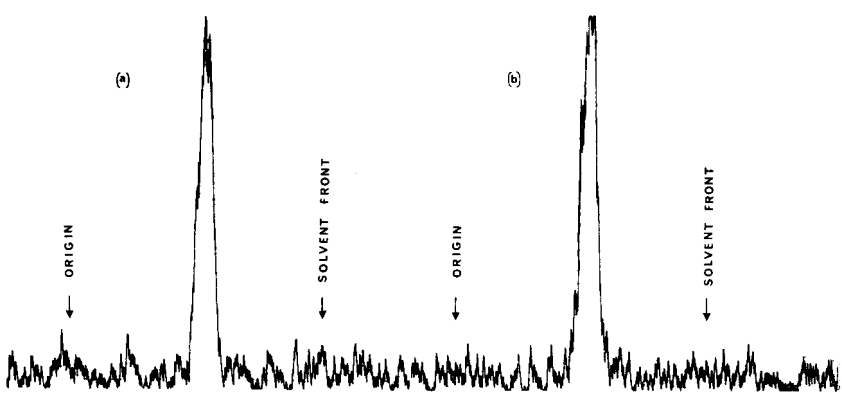

FIG. 2. Radiochromatographs of 6-n-Propyl-2-Thiouracil-6-^{14}C in (*a*) Isopropanol/Benzene 15/85 and (*b*) Benzene/Methanol/Acetic Acid 79/14/7.

(1.1 g, 0.05 mole) and thiourea (2.35 g, 0.03 mole) in anhydrous ethanol (25 ml). The mixture was heated on a steam bath for six hours and allowed to stand overnight at room temperature. The flask was then connected to a distillation assembly and most of the ethanol distilled over. The distillation of the ethanol was completed at reduced pressure. The residue was then dissolved in 20 ml H_2O and the desired product precipitated by adding 3 ml conc HCl. The slurry was then adjusted to pH 4.0 with glacial acetic acid, the product filtered, washed with cold water, and dried under vacuum to yield 2.69 g (0.016 mole = 71 %) 6-n-propyl-2-thiouracil-6-^{14}C. The product was recrystallised from hot water, to constant specific activity, to give material of m.p. 217-218° C; reported 218-219° C [3]. Counting was carried out in a Philips Liquid Scintillation Analyser after dissolution in a Toluene/PPO/POPOP system. The material had a specific activity of 2.29×10^5 dpm/mg or 17.5 μCi/mmole.

The overall chemical yield based on butyryl chloride was 14 % and the radiochemical yield based on sodium butyrate-^{14}C was 34 %. Radiochemical purity was confirmed with TLC on fluorescent silica gel (Brinkman). In two solvent systems the product migrated to give a discrete spot easily seen by quenching of background fluorescence under UV illumination, with an R_f value identical to authentic material. The developed TLC plates were scanned with an Actigraph III Thin Layer Scanner (Nuclear Chicago Corp., U. S. A.) and radioactivity was located at only one region, corresponding to the quenching of fluorescence, in each developer system (Fig. 2). The developers used were Isopropanol/Benzene 15/85; $R_f = 0.53$, and Benzene/Methanol/Acetic acid 79/14/7; $R_f = 0.54$.

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