SYNTHESIS OF CARBON-14 LABELED DOXYLAMINE SUCCINATE*

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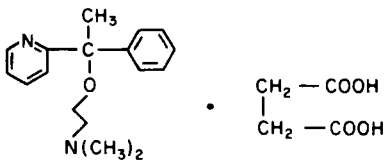
SUMMARY

Doxylamine succinate, N,N-dimethyl-2-[1-phenyl-1-(2-pyridinyl)-ethoxy]ethanamine succinate is an antihistamine used primarily as a sedative. Carbon-14 labeled doxylamine succinate, required for toxicological studies, was synthesized in two steps starting from 2-benzoyl pyridine.

Key words: [^{14}C]-Doxylamine succinate, 2-benzoyl pyridine

INTRODUCTION

Doxylamine succinate, N,N-dimethyl-2[1-phenyl-2-(2-pyridinyl)-ethoxy]ethanamine succinate, is an antihistamine of the ethanolamine class, used primarily as a sleep inducing drug and in the therapeutic formulation of Bendectine® as an anti-nausea agent taken by pregnant women.



DOXYLAMINE SUCCINATE

Commercial production of doxylamine succinate for therapeutic formulations has steadily increased over the years, and its current production in the United States is in excess of 160 metric

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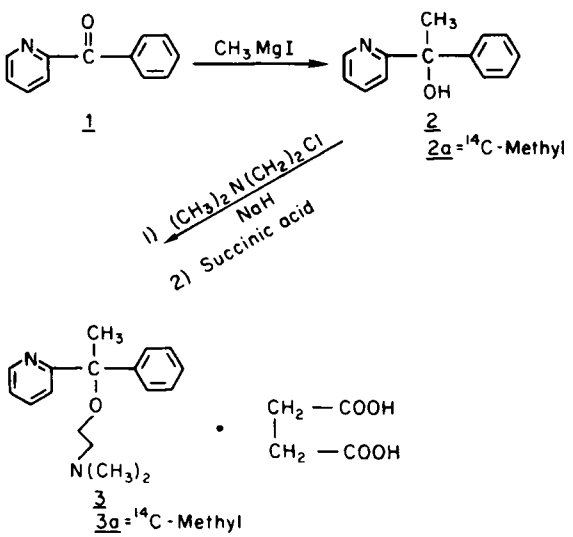
tons per year. Teratogenic studies with Bendectine® in rats and rabbits (1), as well as in pregnant women (2,3), gave no evidence of fetal malformations. However, in a recent report it was mentioned that Bendectine®, when administered (10 mg/kg) to cynomolgus monkeys on days 20-50 of pregnancy, produced abnormalities of the heart in the fetuses (4). Doxylamine contains a dimethylaminoethoxy side chain and there is some concern about the *in vivo* formation of a nitroso derivative which might result in a potential carcinogen. In order to evaluate its role in teratogenesis and carcinogenesis, [^{14}C]-doxylamine succinate was synthesized.

DISCUSSION

Published procedures (5,6) for the synthesis of doxylamine are not ideally suitable for radiochemical synthesis. It is relatively cost-effective to incorporate carbon-14 in the methyl group of doxylamine, since [^{14}C]-methyl iodide is readily available. The synthetic method we developed for [^{14}C]-doxylamine succinate is described below. Initially, the reactions were carried out with unlabeled methyl iodide to optimize the reaction conditions.



Grignard reaction of 2-benzoyl pyridine (1) with methyl magnesium iodide gave the carbinol 2 in excellent yield. The carbinol 2 was heated under reflux with an excess of 2-N,N-dimethylaminoethyl chloride in the presence of sodium hydride in toluene solution to give doxylamine (3), which was mixed with an equivalent amount of succinic acid to yield crystalline succinate salt. For the synthesis of the labeled compound 3a, 2-benzoyl pyridine was subjected to Grignard reaction with [^{14}C]-methyl magnesium iodide to give the carbinol 2a, which was then reacted with 2-N,N-dimethylaminoethyl chloride under the same conditions described for the unlabeled compound. The identity and

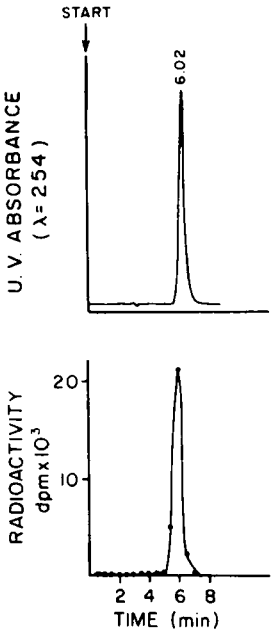


Figure 1. HPLC analysis of ^{14}C-labeled doxylamine succinate, to which carrier doxylamine succinate has been added, on a Whatman 5 μm Partisil PXS 5/25 ODS-3 column (25 cm, 4.60 mm I.D.) using methanol:potassium phosphate buffer (0.01 M, pH 7), 90:10, at a flow rate of 1 ml/min.

radiochemical purity of 3a were established by high performance liquid chromatography as shown in Figure 1.

EXPERIMENTAL

Most chemicals and solvents were analytical grade and were used without further purification. [^{14}C]methyl iodide (25 mCi; 25 mCi/mmol) was purchased from New England Nuclear, Boston, Massachusetts. Purity and identity of new compounds were established by normal spectral (IR, UV, NMR, MS) and analytical (TLC, HPLC, elemental analysis) techniques. Melting points were determined on a Thomas-Hoover capillary melting point apparatus and were uncorrected. IR spectra were obtained in a potassium bromide disc using a Perkin-Elmer Model 467 grating spectrophotometer. UV spectra were measured in methanol solution using a Varian Cary 210 spectrophotometer. Proton NMR spectra were obtained with a Varian EM-390 spectrometer. Mass spectra were determined on a Finnigan quadrupole mass spectrometer. Flash chromatography was carried out on Merck grade 60 silica in J. T. Baker column as described by W. C. Still (7). TLC analyses of unlabeled compounds were done on silica gel GF (Analtech) glass plates (2.5 x 10 cm, with 25 μm layer and prescored). HPLC

analysis of [^{14}C]-labeled doxylamine succinate was carried out on Waters Associates, Inc. HPLC equipment (Model 6000A pump) employing a Whatman 5 μm Partisil PXS 5/25 ODS-3 column (25 cm, 4.60 mm I.D.) and monitored by Laboratory Data Control Spectromonitor III. Radioactivity was determined with a Beckman LS 7500 scintillation counter. Microanalyses were performed by Midwest Microlab, Ltd., Indianapolis, Indiana.

1-Phenyl-1-(2-pyridinyl)ethanol (2)

Magnesium (24.3 mg) was placed in a 50 ml 2-necked flask and the flask flame-dried in a stream of dry nitrogen. Dry ether (0.5 ml) was added at room temperature and the mixture stirred while ice-cold water was circulated through a spiral condenser. A solution of methyl iodide (0.062 ml) in ether (1.5 ml) was added and the mixture stirred at room temperature for 30 min. After most of the magnesium had dissolved, the mixture was refluxed gently for 30 min and cooled to room temperature. A solution of 2-benzoyl pyridine (0.22 g) in dry toluene (2 ml) was added dropwise through a septum. The thick yellow precipitate formed was stirred for 2 hr at room temperature, and for another 2 hr at gentle reflux. The reaction mixture was cooled to room temperature and let stir overnight for 15 hr. It was then cooled in an ice-bath and carefully decomposed with cold 1N hydrochloric acid (3 ml). Enough saturated sodium bicarbonate was added to neutralize excess acid. Ammonium hydroxide (1 ml) was also added to render the mixture basic. The organic product was isolated by extraction with ether. The ether extract was washed with water and brine, then dried over anhydrous sodium sulfate. Removal of the solvents *in vacuo* gave an oil. The crude product so obtained was chromatographed on a flash chromatography column (silica gel, 27 mm x 15.25 cm) using hexanes:ether (90:10) containing 0.1% n-butylamine as solvent system to give pure carbinol 2, (0.19 g). IR:ν_{max} 3420, 1590, 1570 cm^{-1}; UV: λ_{max} 260 nm (ε = 3,990); NMR: ($CDCl_3$): 1.93(s, 3H, -CH_3), 5.80(br.s., 1H, -OH), 7.10-7.80(m, 8H, aromatic protons), and 8.54(d, J = 5.1 Hz, 1H, pyridinyl α-proton)ppm; MS: m/e = 199(M^+); Analysis: Calc'd for $C_{13}H_{13}NO$: C, 78.36; H, 6.58; N, 7.03; Found: C, 76.39; H, 6.73; N, 6.55. Repeated analysis gave unexplainable low value for carbon.

N,N-Dimethyl-2-[1-phenyl-1-(2-pyridinyl)ethoxy]ethanamine (3)

Sodium hydride (60 mg of 60% suspension in mineral oil) was washed free of mineral oil with dry toluene under dry nitrogen and stirred with toluene (2 ml). A solution of carbinol 2, (188 mg) in toluene (4 ml) was carefully added by means of a pipet and a vigorous reaction ensued. After the reaction subsided, the mixture was heated under reflux for 1 hr. A large excess of 2-N,N-dimethylaminoethyl chloride (0.540 g) (5) in toluene (2 ml) was added dropwise to the refluxing alcoholate solution, and the refluxing continued for 15 hr. The reaction mixture was cooled to room temperature, water added and

all the organic solvent evaporated *in vacuo* in a stream of nitrogen. More water was added to the residue and the organic material extracted with petroleum ether. The extract was washed several times with water and dried over anhydrous sodium sulfate. Removal of the solvent *in vacuo* gave an oil. The crude product was subjected to flash chromatography (silica gel, 27 mm x 15.25 cm) using methanol containing 0.1% n-butylamine as solvent system to afford a colorless oil (0.110 g). To this material succinic acid (0.048 g) was added and crystallized from a mixture of ethyl acetate:isopropanol (4:1) to give doxylamine succinate; m.p. 94-96°C [Lit. (5) 100-104°C]; IR:ν_{max}: 3420, 3050, 1725, 1690, 1640, 1585, 1570 cm^{-1}; UV:λ_{max}: 260 nm (ε = 3,810); NMR ($CDCl_3$): 2.00(s, 3H, -C-CH_3), 2.56(s, 4H, HOOC-$\underline{CH_2}$-$\underline{CH_2}$-COOH), 2.79(s, 6H, -NMe_2), 3.19(t, J = 6 Hz, 2H, -CH_2-$\underline{CH_2}$-NMe_2), 3.65(t, J = 6 Hz, 2H, -O$\underline{CH_2}$-CH_2-), 7.10-7.85(m, 8H, aromatic protons), and 8.60(d, J = 5.1 Hz, 1H, pyridinyl α-proton) ppm; MS: me/ = 200[M^+ -(117 + 71)] and m/e = 71[M^+- (200 + 117)]. The spectral data is in agreement with that of an authentic sample of doxylamine succinate.

1-Phenyl-1-(2-pyridinyl)ethanol-2-^{14}C (2a)

The reaction was carried out exactly as described for the unlabeled compound employing magnesium (24.5 mg), [^{14}C]-methyl iodide (25.0 mCi, 25 mCi/mmol) and 2-benzoyl pyridine (0.22 g) to give the [^{14}C]-labeled carbinol 2a. Total radioactivity of the crude ^{14}C-carbinol was determined to be 20.3 mCi. It was purified by flash chromatography on a column (27 mm x 15.25 cm) of silica gel (50 g) using hexanes:ether (90:10) containing 0.1% n-butylamine as solvent. The purified ^{14}C-carbinol 2a, (0.173 g) when examined by TLC employing hexanes:ether (8:2) as solvent system exhibited identical R_f value (0.135) with that of the unlabeled 2.

N,N-Dimethyl-2-[1-phenyl-1-(2-pyridinyl)ethoxy-2-^{14}C]ethanamine (3a) and its succinate

The above ^{14}C-carbinol 2a, (0.173g) was reacted under reflux with an excess of 2-N,N-dimethylaminoethyl chloride in the presence of sodium hydride in toluene solution exactly as described in the experimental for unlabeled 3. After the addition of requisite amounts of succinic acid, the succinate salt of [^{14}C]doxylamine crystallized. Additional crystallization from ethyl acetate:isopropanol (4:1) gave the pure product. The total radioactivity of [^{14}C]-doxylamine succinate so obtained was determined to be 10.8 mCi. Its identity and radiochemical purity were established by HPLC. Carrier doxylamine succinate was added to the [^{14}C]-labeled compound and then subjected to HPLC employing a Whatman 5 μm Partisil PXS 5/25 ODS-3 column (25 cm, 4.60 mm I.D.) using methanol:potassium phosphate buffer (0.01 M, pH 7), 90:10, and UV detector (254 nm), at a flow rate of 1 ml/min.

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REFERENCES

1. Gibson J.P., Staples R.E., Larson E.J., Kuhn W.L., Holtkamp D.E., and Newberne J.W.—Toxicol. Appl. Pharmacol. 3:439 (1968)
2. Smithells R.W. and Sheppard S.—Teratology 17:31 (1978).
3. Shapiro S., Heinonen O.P., Siskind V., Kaufman D.W., Monson R.R., and Slone D.—Am. J. Obstet. Gynecol. 128:480 (1977).
4. Hendrickx A.G., Prahalada S., and Rowland J.M.—Teratology 25:47A (1982).
5. Tilford C.H., Shelton R.S., and Van Campen M.G., Jr.—J. Am. Chem. Soc. 70:4001 (1948).
6. Sperber N., Papa D., Schwenk E., and Sherlock M.—J. Am. Chem. Soc. 71:887 (1949).
7. Still W.C., Kahn M., and Mitra A.—J. Org. Chem. 43:2923 (1978).