SYNTHESES OF N-[1-^{2}H]- AND N-[1-^{3}H]-CYCLOPROPYLBENZYLAMINE AND [PHENYL-^{14}C]-N-CYCLOPROPYLBENZYLAMINE

Richard B. Silverman and Stephen J. Hoffman
Department of Chemistry, Northwestern University
Evanston, Illinois 60201

SUMMARY

N-[1-^{2}H]- (5a) and N-[1-^{3}H]-Cyclopropylbenzylamine (5b) were synthesized from cyclopropanecarbonitrile. The α-proton of cyclopropanecarbonitrile was exchanged in [O-^{2}H]- and [O-^{3}H]-methanol using sodium methoxide as the base. [α-^{2}H]- and [α-^{3}H]-Cyclopropanecarbonitrile were hydrolyzed in 4N HCl to the α-labeled cyclopropanecarboxylic acids which were converted to [1-^{2}H]- and [1-^{3}H]-cyclopropylamine·HCl by Curtius rearrangements. The labeled cyclopropylamines were benzylated by reaction with benzaldehyde followed by sodium cyanoborohydride reduction. [phenyl-^{14}C]-Cyclopropylbenzylamine (2b) was prepared by the reaction of cyclopropylamine with [phenyl-^{14}C] benzaldehyde followed by sodium cyanoborohydride reduction.

Key Words: N-Cyclopropylbenzylamine, cyclopropylamine, cyclopropanecarboxylic acid

INTRODUCTION

The use of selectively deuterated or radiolabeled molecules in determining enzyme mechanisms is well precedented (1). In our work on the elucidation of the mechanism of inactivation of mitochondrial monoamine oxidase (MAO) by N-cyclopropyl-N-arylalkyl amines (2), we required selectively labeled N-cyclopropylbenzylamine, a model for the N-cyclopropyl class of MAO inhibitors (3,4). In this paper we describe our syntheses of [1-^{2}H]- and [1-^{3}H] cyclopropylamine hydrochloride, N-[1-^{2}H]- and N-[1-^{3}H] cyclopropylbenzylamine hydrochloride and [phenyl-^{14}C]-N-cyclopropylbenzylamine hydrochloride.

0362-4803/81/060781-10$01.00

Received January 11, 1980
Revised February 22, 1980

EXPERIMENTAL

Melting points were obtained on a Fisher-Johns melting point apparatus and are uncorrected. NMR spectra were recorded on a Hitachi Perkin-Elmer R-20B spectrometer with an internal standard of tetramethylsilane (TMS). All chemical shifts are expressed as parts per million (δ) downfield from TMS. IR spectra were obtained on a Perkin-Elmer 283 spectrophotometer. High voltage paper electrophoresis was performed on a Gilson Model D high voltage electrophoreter using a water: 95% formic acid: acetic acid (45:1:4) pH 1.9 buffer on Whatman 3 MM paper. Radioactivity was counted in Aquasol (New England Nuclear) or Beckman EP scintillation fluid, on a Beckman LS-3133T liquid scintillation counter.

[phenyl-U-^{14}C] Benzaldehyde (0.05 mCi, 13.1 mCi/mmol) was purchased from Pathfinder Laboratories, Inc., St. Louis, MO. Tritiated water (1 Ci, 90 mCi/mmol) was purchased from Amersham Corp., Arlington Heights, IL. Cyclopropylamine, cyclopropanecarbonitrile, sodium cyanoborohydride, sodium methoxide and methanol-d (99.5%) were purchased from the Aldrich Chemical Co. Methylene chloride and chloroform were distilled from P_2O_5 under an argon atmosphere and were stored under argon over 4 Å molecular sieves. Toluene was distilled under an argon atmosphere from sodium metal, and stored under argon over sodium. Methanol was distilled from magnesium turnings and stored under argon over molecular sieves. Benzaldehyde was distilled under reduced pressure before use. All other chemicals are commercially available and were used without further purification.

N-Cyclopropylbenzylamine·HCl (2a):

This compound was synthesized by the method of Bumgardner et al. (5).

The hydrochloride was isolated after the addition of an excess of 10% HCl/i-PrOH and removal of the solvent under reduced pressure; mp 158-160° (lit. (5) 158-160°); NMR: ($CDCl_3$) δ: 0.7 (m, 2H), 1.2 (m, 2H), 2.3 (m, 1H), 4.05 (s, 2H), 7.4 (m, 5H), 9.8 (broad s, 2H).

[phenyl-U-^{14}C]-N-Cyclopropylbenzylamine·HCl (2b):

[phenyl-U-^{14}C] Benzaldehyde (0.05 mCi, 0.4 mg, 3.8 μmol) in 0.36 mL of CH_2Cl_2 was diluted with benzaldehyde (5.3 mg, 50 μmol) in 0.7 mL of dry CH_2Cl_2. Molecular sieves (4Å) were added to the solution and the flask was flushed with argon. Cyclopropylamine (5.2 μL, 75 μmol) was syringed into the solution, and after 20 h the solution was filtered and washed with 5 mL of CH_2Cl_2. Removal of the solvent by rotary evaporation yielded [phenyl-U-^{14}C]-N-benzylidenecyclopropylamine as a pale yellow oil. The product was dissolved in anhydrous methanol (0.5 mL), and 3 Å molecular sieves were added. A solution of sodium cyanoborohydride (2.2 mg. 35 μmol) in 0.3 mL of anhydrous methanol was added followed by 20 μL of 5N methanolic HCl. After 51 h, the solution was filtered and the sieves were washed with 8 mL of anhydrous methanol. Rotary evaporation of the solvent resulted in an off-white residue which was stirred in 20 mL of $CHCl_3$, filtered, and the solvent removed. The resulting slightly yellow solid was recrystallized from ethyl acetate, yielding 3.2 mg (33%) of 2b as white flakes; specific activity = 1.97×10^6 dpm/μmole. All of the radioactivity comigrated with carrier N-cyclopropylbenzylamine·HCl by high voltage paper electrophoresis.

[α-^{2}H]Cyclopropanecarboxylic acid (3a):

Cyclopropanecarbonitrile (4.4 mL, 60 mmol) was added to 25 mL of a 1 M solution of sodium methoxide in methanol-d and was refluxed for 16 h.

The methanol-d was removed by distillation and replaced with 25 mL of fresh methanol-d and the exchange reaction was continued for an additional 24 h (the exchange of the α proton was followed by a decrease in the NMR signal at 1.25 δ). Approximately 25 mL of methanol-d was removed by distillation to give a yellow solution that was cooled to 0°, then 35 mL of 4.7N HCl was added. The residual methanol was removed by boiling until the temperature of the vapor was > 95°. The acidic aqueous solution was refluxed for 16 h, cooled to room temperature, saturated with solid NaCl and extracted with 6 x 40 mL of ether. The ether was dried ($MgSO_4$), filtered, and the solvent was removed under reduced pressure to yield an almost colorless liquid. Vacuum distillation resulted in 946 mg (18%) of 3a; bp 64° (3.5 mm); NMR ($CDCl_3$) δ: 0.8 (d, 4H), 11.1 (s, 1H).

[1-^{2}H]Cyclopropylamine·HCl (4a):

Thionyl chloride (0.44 mL, 6.1 mmol) was added via syringe to 3a (559 mg, 6.4 mmol). The flask was swirled, allowing the gases to vent through a syringe needle, while the contents warmed to room temperature. After standing for 22 h, the flask was flushed with N_2 and 3 mL of dry toluene was added. The resultant solution was syringed into a mixture of NaN_3 (830 mg, 12.8 mmol) in 35 mL of dry refluxing toluene, and heated at 110-115° for 10.5 h. An off-white precipitate was filtered from the toluene solution, and 25 mL of 4 N HCl was added to the filtrate which was then refluxed with vigorous stirring for 10.5 h. After cooling, the aqueous and organic layers were separated and the organic layer was extracted with 5 mL of H_2O. The combined aqueous layers were rotary evaporated to give 373 mg (62%) of an off-white solid; NMR (D_2O) δ 0.65 (s).

N-[1-^{2}H] Cyclopropylbenzylamine·HCl (5a)

4a (186 mg, 2 mmol) was stirred under N_2 in 15 mL of dry $CHCl_3$ containing 4 Å molecular sieves. Triethylamine (0.42 mL, 3 mmol) then benzaldehyde (0.205 mL, 2 mmol) were syringed in, and the solution was stirred for 25 h under N_2. The reaction mixture was filtered and the solvent was rotary evaporated to give a white solid, which was triturated with 20 mL of ether, filtered and washed with ether. The combined ether filtrates were concentrated under reduced pressure to give a colorless liquid that was dissolved in 10 mL of anhydrous MeOH. The solution was cooled to 0° and sodium cyanoborohydride (75.5 mg, 1.2 mmol) was added, followed by 0.8 mL of 5N methanolic HCl. This solution was allowed to stand for 22 h under N_2, then the solvent was removed under reduced pressure, leaving a light brown mixture of salts. The product was isolated as described for 2a, yielding 282 mg (77%) of 5a, which was recrystallized from EtOAc to give shiny white flakes, mp 158-160°. NMR ($CDCl_3$) δ: 0.70 (m, 2H), 1.15 (m, 2H), 4.05 (s, 2H), 6.5 (m, 5H), 9.6 (broad s, 2H).

[O-^{3}H] Methanol:

To a solution of sodium methoxide (890 mg, 16.5 mmol) in anhydrous methanol (3.00 mL, 74.3 mmol) at 0° was added tritiated water (1 Ci, 0.2 mL, 11.1 mmol) in 1.0 mL (14.7 mmol) of anhydrous methanol. The methanol was distilled and an additional 1.0 mL of methanol was added to the distillation pot. The distillation was continued, yielding a total of 4.7 mL (115 mmol, 85%) of tritiated methanol; specific activity = 1.16×10^7 dpm/μmol.

[α-^{3}H] Cyclopropanecarboxylic acid (3b):

Cyclopropanecarbonitrile (1.4 mL, 20 mmol) was syringed into a solution containing sodium methoxide (297 mg, 5.5 mmol) in 4.7 mL of tritiated MeOH, and was refluxed for 16 h. The solution was cooled to room temperature and the MeOH was removed by distillation. After cooling, 5.5 mL of 6.1 N HCl was added along with 1 mL of methanol. The solution was distilled to 70° to remove all remaining MeOH, and then was refluxed for 18 h. After cooling, the solution was saturated with solid NaCl and extracted with 6 x 6 mL of ether, dried ($MgSO_4$) and filtered. The filtrate was rotary evaporated (through a liquid N_2 trap) to give 577 mg of 3b.

[1-^{3}H]-Cyclopropylamine·HCl (4b):

Thionyl chloride (0.46 mL, 6.47 mmol) was added to the unpurified 3b (577 mg, 6.47 mmol), venting the evolved gases through a syringe needle. After warming to room temperature, the solution was allowed to stand for 4 h, then was flushed with N_2. The solution was diluted with 3 mL of dry toluene and was added to 35 mL of refluxing toluene containing sodium azide (830 mg, 12.8 mmol). Work-up was identical to that for the deutero analogue (4a) and resulted in the isolation of 371 mg of an orange solid (4b), which was 97% radiopure by high voltage paper electrophoresis.

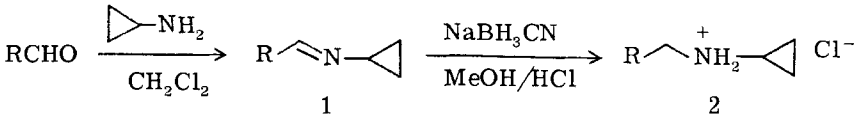
N-[1-^{3}H]-Cyclopropylbenzylamine·HCl (5b):

4b (371 mg, 397 mmol) and benzaldehyde (0.41 mL, 4 mmol) were allowed to react and the reaction was worked-up as in the synthesis of the deutero analogue (5a). After recrystallization from ethyl acetate, 324 mg (9% overall yield from cyclopropanecarbonitrile) of the product was obtained as shiny white flakes; specific activity $1.16 \cdot 10^7$ dpm/μmol. All of the radioactivity comigrated with carrier N-cyclopropylbenzylamine hydrochloride.

RESULTS AND DISCUSSION

The syntheses described herein provide easy routes to the preparation of selectively labeled cyclopropylamines and analogues. The synthesis of [phenyl-^{14}C] N-cyclopropylbenzylamine (2b) was carried out using a modification of the method of Bumgardner et al. (6). The more selective reducing agent, sodium cyanoborohydride, was used for the conversion of [phenyl-^{14}C]-N-benzylidenecyclopropylamine (1b) to 2b (Scheme I). Because of the stability

Scheme I



1

a R = phenyl b R = [U-^{14}C]phenyl

2

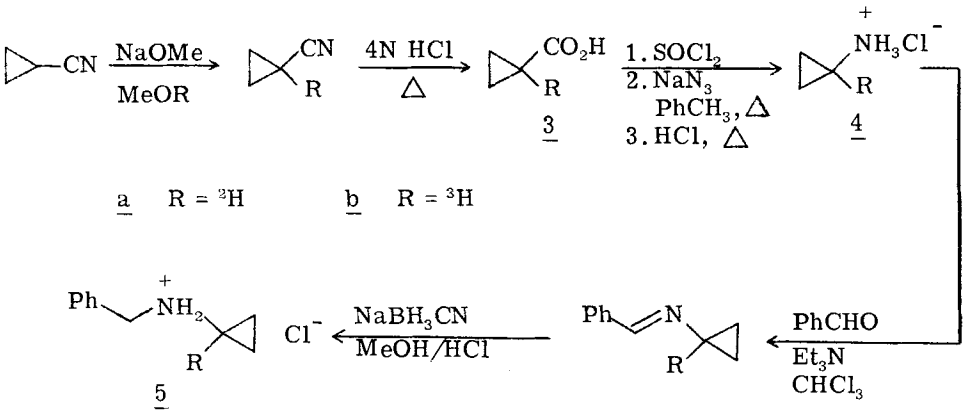
of the protonated intermediate, reaction time for this reduction need not be as lengthy as those for other reductive aminations (7). In fact, UV spectral data (monitoring at 306 nm) suggest that the reduction was complete after only 4 h.

In order to utilize the modified Bumgardner procedure for the synthesis of N-[1-^2H]-(5a) and N-[1-^3H]cyclopropylbenzylamine (5b), we required [1-^2H]-(4a) and [1-^3H]cyclopropylamine (4b). These compounds could be afforded by Curtius rearrangements of [α-^2H]-(3a) and [α-^3H]cyclopropanecarboxylic acid (3b). However, previous work (8,9) on the exchange of α-protons of carboxylate salts revealed that while, in general, proton exchange

using aqueous base is facile for these compounds, the exchange with cyclopropane carboxylates never exceeded 16%. Attempts in this laboratory to exchange the α-proton of cyclopropanecarboxylic acid or of methyl cyclopropanecarboxylate with lithium diisopropylamide or with lithium tetramethylpiperdide also were unsuccessful.

While exchange of α-protons in cyclopropanes containing strong resonance stabilizing electron withdrawing groups is feasible, most of these compounds could not easily be transformed into our key intermediates, 4a or 4b. The α-proton of cyclopropanecarbonitrile, however, is readily exchangeable in base (10). In the exchange of deuterium from CH_3O^2H for the α-proton in cyclopropanecarbonitrile, the course of the reaction was followed by observing the decrease in the NMR resonance of the α-proton (1.25 δ). The same conditions were used to prepare the α-^3H compound substituting CH_3O^3H which we prepared from ^3H_2O and $NaOCH_3$ in methanol. Acid hydrolysis of these labeled nitriles yielded the desired labeled cyclopropanecarboxylic acids which were converted by Curtius rearrangements to 4a and 4b (Scheme II). In order to

Scheme II



avoid difficult small scale work-up procedures during the conversion of 3a and 3b to 4a and 4b, we used slightly less than one equivalent of thionyl chloride to make the acid chloride. After thermal rearrangement with sodium azide and removal of the salts, the hydrolysis step was performed in a two phase reaction without isolation of the low boiling cyclopropane isocyanate intermediate. Once the amine hydrochlorides were formed, isolation was a simple matter of evaporating the solvent. This rearrangement and hydrolysis proceeded without any loss of label.

Treatment of labeled cyclopropylamine·HCl with a slight excess of triethylamine in dry chloroform containing molecular sieves, followed by the addition of one equivalent of benzaldehyde led to the production of labeled N-benzylidenecyclopropylamine. In all cases, the imine was converted to the product without purification, and without formation of any detectable impurities.

The radiopurity of [^{14}C-phenyl]-N-cyclopropylbenzylamine HCl and of N-[1-^{3}H]-cyclopropylbenzylamine·HCl was determined by high voltage paper electrophoresis. In both cases there were no neutral impurities at the origin, and all of the radioactivity migrated with carrier N-cyclopropylbenzylamine HCl. N-(1-^{2}H) Cyclopropylbenzylamine·HCl also comigrated on high voltage electrophoresis with the protic analogue.

ACKNOWLEDGMENT

We are grateful to the National Institutes of Health (MH 33475) for support of this research.

REFERENCES

1. Battersby A. R. - Accts. Chem. Res. 5: 148 (1972)

2. Silverman R. B. and Hoffman S. J. - J. Am. Chem. Soc. 102: 884 (1980)

3. Mills J., Kattau R., Slater I. H., and Fuller R. W. - J. Med. Chem. 11: 95 (1968)

4. Murphy D. L., Donnelly C. H., Richelson E., and Fuller, R. W. - Biochem. Pharmacol. 27: 1767 (1978)

5. Fuller R. W., Molloy B. B., Day W. A., Roush B. R., and Marsh M. M. - J. Med. Chem. 16: 101 (1973)

6. Bumgardner C. L., Lawton E. L., and Carver J. G. - J. Org. Chem. 37: 407 (1972)

7. Borch R. F., Bernstein M. D., and Durst H. D. - J. Am. Chem. Soc. 93: 2897 (1971)

8. Atkinson J. G., Csakavory J. J., Hebert G. T., and Stuart R. S. - J. Am. Chem. Soc. 90: 498 (1968)

9. Bottini A. T. and Davidson A. J. - J. Org. Chem. 30: 3302 (1965)

10. von Wijren W. Th., Steinberg H., and de Boer Th. J. - Tetrahedron 28: 5423 (1972)