THE SYNTHESIS OF ^{14}C AND ^{3}H-LABELED N-(2-p-AZIDOPHENYLETHYL)-NORLEVORPHANOL

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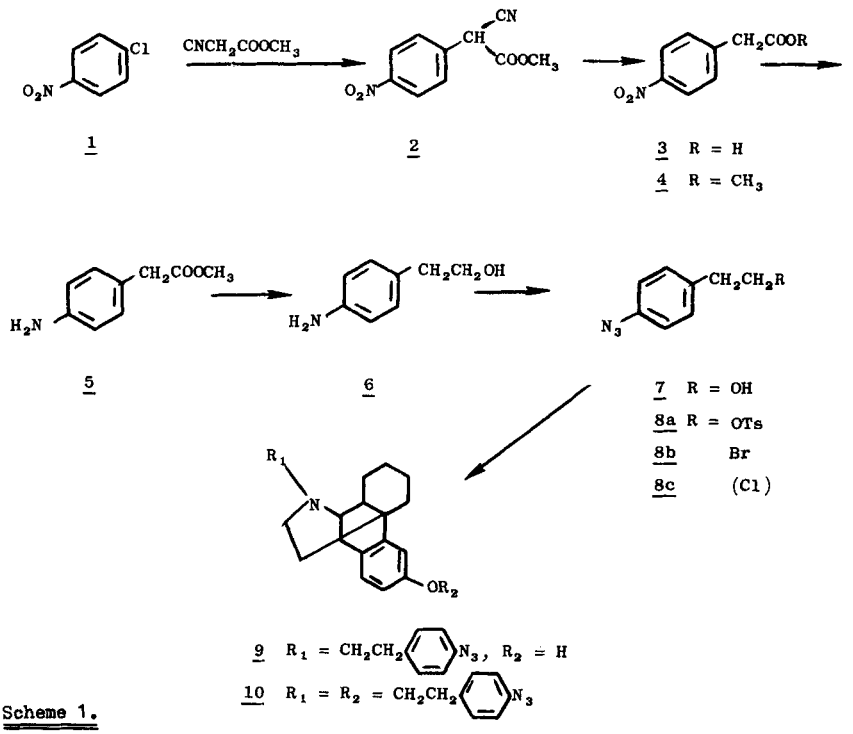
SUMMARY

Synthetic procedures for N-(2-p-azidophenylethyl)-norlevorphanol (9) labeled with ^{14}C and ^{3}H at the 1-position of the 2-phenylethyl moiety are presented. A process for synthesis of p-nitrophenylacetic-1-^{14}C acid from p-chloro-nitrobenzene and methyl cyano-^{14}C-acetate is described. The nitro acid was converted to 9 in a six step procedure, proceeding through 2-(p-aminophenyl)ethanol (6) as a key intermediate. The alcohol (6) was also prepared with ^{3}H label via reduction of methyl p-aminophenylacetate with sodium borotritide-aluminium chloride.

Introduction

The utility of N-(2-p-azidophenylethyl)-norlevorphanol (9) as an aid for the location and potential site mapping of opiate receptors was recently reported by Winter and Goldstein.[1] Further pursuit of this research suggested the preparation of the title compound with specific ^{14}C and tritium labels at the one position of the N-2-p-azidophenethyl moiety.

The labeled compounds were prepared by a modification (Scheme I) of the previously reported route[1] proceeding through methyl p-aminophenylacetate (5). In the tritium series the label was introduced via sodium borotritide reduction of unlabeled 5 to p-aminophenylethanol-1-$^{3}H_2$ (6). However, in the ^{14}C series it was necessary to synthesize (5)-1-^{14}C from p-nitrophenylacetic -1- ^{14}C acid (3). The acid (3) was conveniently obtained from alkylation[2] of methyl cyano-^{14}C-acetate with



Scheme 1.

p-chloronitrobenzene followed by acid hydrolysis and decarboxylation of the cyano ester intermediate (2). The specific activities of the starting sodium cyanide-^{14}C (used to prepare methyl cyanoacetate) and the terminal compound (9) were essentially equivalent showing that decarboxylation occurred selectively at the ester carbonyl in 2.

The use of the tosylate (8a) represented an advantage over the bromide[1] (8b) in the convenience and yield of its preparation as well as in the higher yield obtained in the subsequent alkylation of norlevorphanol. If the tosylation reaction was allowed to proceed for a longer time, significant amounts of a side product regarded as the chloride (8c) were formed. The chloride, however, was not displaced by norlevorphanol under similar mild reaction conditions. Alkylation of norlevorphanol with 8a also produced some of the O,N-bis alkylated compound (10), which was readily separable from 9 by chromatography.

Experimental Part

p-Nitrophenylacetic-1-^{14}C-Acid (3)

Sodium hydride (0.430 g, 10.2 mmol of 57% oil suspension) was freed of oil by pentane wash and treated with 1.0 ml of dry hexamethylphosphoramide. Methyl cyano-^{14}C-acetate (0.370 g, 3.74 mmol, prepared[3] from sodium cyanide-^{14}C, specific activity 30.8 mCi/mmol) in 2 ml of hexamethylphosphoramide was then added dropwise over 10 minutes. After gas evolution ceased, p-chloronitrobenzene (0.590 g, 3.74 mmol) was added and the mixture heated at 95-100° for 15 hours. The mixture was acidified (1.8 ml of glacial acetic acid followed by 1.8 ml of concentrated hydrochloric acid) and diluted with 80 ml of ice water. After 4 hours the brown precipitate (cyano ester 2) was collected by filtration and washed with water.

The damp material was refluxed for 60 hours with 3.6 ml of 50% acetic acid containing 0.18 ml of concentrated sulfuric acid to effect decarboxylation and hydrolysis (any radioactive gas trapped by sodium hydroxide solution). Water (8 ml) was added and the pH adjusted to 8 with 4.4 ml of 6N ammonium hydroxide. Some dark solid was removed by filtration and the yellow filtrate acidified with 2 ml of concentrated hydrochloric acid. The yellow precipitate was extracted into two 20-ml portions of ether. The combined ether extracts were washed with water (10 ml), dried over magnesium sulfate and evaporated to leave 0.596 g (88%) of p-nitrophenylacetic-1-^{14}C acid; NMR and IR spectra were identical to authentic material.

2-(p-Aminophenyl)ethanol (6)

A. (-1-^{14}C). The p-nitrophenylacetic acid (3) was esterified in 92% yield by refluxing 20 hr with methanol/p-toluenesulfonic acid to give methyl p-nitrophenylacetate (4). The nitro ester (0.265 g) was hydrogenated over platinum oxide in methanol at atmospheric pressure. After gas uptake was complete, catalyst and solvent were removed and the residue identified as amino ester (5) by TLC. The amino ester (0.214 g, 1.30 mmol), 0.073 g (1.93 mmol) of lithium aluminum hydride and 5 ml of ether were stirred

at ambient temperature for 15 hours followed by the sequential addition of 0.073 ml of water, 0.073 ml of 15% sodium hydroxide and 0.220 ml of water. The mixture was stirred 10 minutes and the ether layer separated by pipette. After a second 5-ml ether extraction, the combined extracts were dried over magnesium sulfate and evaporated to leave 0.142 g (80%) of tan solid; identical by TLC with material previously prepared.[1]

B. (unlabeled).* Diglyme (4.0 ml, dried over calcium hydride) was cooled to 0° and 1.000 g (7.5 mmol) of aluminum chloride was added portion-wise with stirring. After 5 minutes, 0.830 g (5.0 mmol) of methyl p-aminophenylacetate was added, followed in 10 minutes by the drop-wise addition of a slurry of 0.760 g (20.5 mmol) of sodium borohydride in 15 ml of diglyme. After the vigorous bubbling subsided the mixture was stirred 15 hours. Water (5 ml) was slowly added at 0-5° and the mixture extracted with three 10-ml portions of dichloromethane. The combined organic extracts were dried over magnesium sulfate and evaporated *in vacuo* to leave 0.450 g (64%) of pale yellow crystals, identical by NMR and TLC with material previously prepared.[1]

2-(p-Azidophenyl)ethanol (-1-^{14}C and 1,1-$^{3}H_2$) (7)

A solution of 0.142 g (1.04 mmol) of 6 in 0.71 ml of 4N hydrochloric acid was chilled to 0° in an ice-salt bath, followed by the addition of 0.072 g (1.04 mmol) of sodium nitrite in 0.36 ml of water. After 20 minutes the diazonium salt was treated with 0.069 g (1.04 mmol) of sodium azide in 0.36 ml of water. The mixture was stirred at 0° for 1 hour and then extracted with three 6-ml portions of dichloromethane. The combined extracts were washed with 1 ml of ice water, dried over magnesium sulfate and evaporated to leave 0.144 g (85%); single spot by TLC, identical with material previously reported.[1]

Footnotes * A sodium borotritide reduction was carried out by The Radiochemical Centre, Amersham, England using the above process developed by the authors. The crude reaction product was used directly in the following step to prepare the tritiated p-toluenesulfonate (8a, -1,1-$^{3}H_2$).

2-(p-Azidophenyl)ethyl(-1-^{14}C and -1,1-$^{3}H_2$)-p-toluenesulfonate (8a)

To an ice cold solution of 0.144 g (0.88 mmol) of the azido alcohol (7) in 2.4 ml of pyridine was added 0.340 g (1.78 mmol) of p-toluenesulfonyl chloride. The mixture was kept at 0-5° for 3 hours, diluted with 13 ml of ice water, and extracted with 10 ml of dichloromethane. The extract was dried over magnesium sulfate and evaporated to leave 208 mg (75%) of crystalline product; TLC showed a single spot. Material from another run was recrystallized from cyclohexane, m.p. 48°; IR 4.75μ (azide), 7.35μ and 8.45μ (sulfonate); NMR $CDCl_3$, δ 6.8-7.8 (8H, aromatic), 4.23 (2H, triplet, CH_2O), 2.90 (2H, triplet, $ArCH_2$), 2.44 (3H, CH_3). The compound was unstable to light. Longer reaction times caused some formation of chloride (8c); NMR 3.64 (2H, triplet, CH_2Cl); 8b[1] 3.50 (2H, triplet, CH_2Br).

Norlevorphanol (3-Hydroxy-N-methylmorphinan)

A solution of 9.69 g of levorphanol tartrate dihydrate in 255 ml of warm water was adjusted to pH 8-9 with concentrated ammonium hydroxide. The white precipitate of levorphanol free base was collected, washed with water and dried to leave 5.40 g (96%). A mixture of the 5.40 g (21.0 mmol) of levorphanol, 4.60 g (33.4 mmol) of anhydrous potassium carbonate and 110 ml of chloroform was stirred and treated dropwise with 22.0 ml (0.23 mmol) of ethyl chloroformate. The solution was stirred for 6 hours at room temperature and 15 hours under reflux. After cooling to room temperature, the solution was washed with 70 ml of water, dried over magnesium sulfate and the solvent removed to leave 9.40 g of the crude N,O carbonate. The syrup was dissolved in 80 ml of methanol, treated with 40 ml of 10% sodium hydroxide and the solution kept at ambient temperature for 1 hour. Water (260 ml) was added followed by adjustment of the pH to 1-2 with 12 N hydrochloric acid. The syrupy precipitate was extracted into two 60-ml portions of chloroform, which was washed with 60 ml of water, dried over magnesium sulfate and evaporated in vacuo to afford 6.30 g (100%) of syrup identified as N-carbethoxynorlevorphanol by infrared; λ^{film} 3.0μ (OH), 5.90 (N-COOEt).

The urethane was hydrolyzed by refluxing the above 6.30 g in a mixture of 65 ml glacial acetic acid and 95 ml of 12 N hydrochloric acid for 24 hours. The solvent was evaporated in vacuo and the residue was taken up in 50 ml of hot water and filtered. The filtrate was adjusted to pH 8 with concentrated ammonium hydroxide and the resulting white, crystalline precipitate was collected, washed with water and dried to afford 3.90 g (76% from levorphanol free base) of norlevorphanol, m.p. 240-250° (lit.[4] m.p. 260-262); picrate, yellow crystals, m.p. 197-201°. Anal. calc'd for $C_{22}H_{24}N_4O_8$: C, 55.9; H, 5.12, N. 11.9. Found: C, 55.9; H, 5.34; N, 12.0.

N-(2-p-Azidophenylethyl-1-^{14}C and -1,1-$^{3}H_2$)norlevorphanol (9)

A mixture of 0.208 g (0.655 mmol of azidotosylate (8a)), 0.105 g (0.760 mmol) of powdered anhydrous potassium carbonate, 0.189 g (0.778 mmol) of norlevorphanol and 1.1 ml of methanol was stirred at 41-43° for 60 hours in a stoppered flask protected from light. The mixture was stirred for 30 minutes with 4.5 ml of water and 10 ml of a 2:1 benzene-ether mixture. The organic layer was drawn off and another 10 ml extraction carried out. The combined organic extracts were washed with 3.0 ml of water, dried over magnesium sulfate and treated with ethereal hydrogen chloride to precipitate the hydrochloride salt as a gum. The supernatant was removed by decantation and the gum dissolved in 3.0 ml of methanol. Water (4.5 ml) was added and the mixture was adjusted to pH 8 with 1.5 N ammonium hydroxide. The precipitated oil was extracted into two 1.5 ml portions of chloroform, which was dried over magnesium sulfate and evaporated to afford 0.138 g (54%) of a gummy product; TLC showed an intense uv absorbing spot at R_f 0.45 and a weak spot at R_f 0.70. The material was separated by thick plate chromatography (15% methanol - 85% chloroform in silica gel) to afford 0.076 g (30%) of chromatographically pure product. The NMR and IR spectra were compatible with the desired product (9), while the faster moving contaminant was shown to be the O,N-bis alkylated compound (10) by NMR; triplet, δ 4.2 for CH_2OAr and correct integration of aromatic and

aliphatic protons. Specific activity of the ^{14}C-labeled product was 31.5 mCi/mmol and the tritiated material was 1.25 Ci/mmol, with radiochemical purities of 90%. Rechromatography of both labeled and cold material usually showed about 5% impurity at the origin, probably due to thermal or photolytic decomposition of 9.

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