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The Preparation of Spermidine and Monoacetylspermidine

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The reaction of acrylonitrile with monoacetylputrescine gave N^1-(2-cyanoethyl)-N^4-acetyl-1,4-butanediamine, catalytic reduction of which afforded monoacetylspermidine which was deacetylated by hydrochloric acid to produce spermidine trihydrochloride.

The cyanoethylation of γ-aminobutyronitrile and catalytic reduction of the resulting N-(2-cyanoethyl)-γ-aminobutyronitrile to produce N-(3-aminopropyl)-1,4-butanediamine (spermidine) has been described by Danzig and Schultz.[1] The formation of spermidine in the reduction step is limited to narrow conditions of temperature and pressure, although spermidine trihydrochloride (I) was reported as resulting in 28.7% yield by a selected procedure.

Modifications of Danzig and Schultz's method have been tried for possible application to the preparation of C^{14}-labeled spermidine with the use of acrylonitrile-1-C^{14} in the cyanoethylation step. Lithium aluminum hydride reduction of N-(2-cyanoethyl)-γ-aminobutyronitrile in ether–tetrahydrofuran gave a 20–25% yield of spermidine trihydrochloride along with larger amounts of sirupy products. Catalytic reduction of a suitable nitrile having only one cyano group in the molecule offered the possibility of an improved yield. Accordingly N^1-(2-cyanoethyl)-N^4-acetyl-1,4-butanediamine (II) was prepared by reaction of acrylonitrile with N-acetyl-1,4-butanediamine (monoacetylputrescine). Reduction of N^1-(2-cyanoethyl)-N^4-acetyl-1,4-butanediamine, in ethanol saturated with ammonia, by Raney nickel and hydrogen at 116–122° and 2400 p.s.i. afforded N^1-(3-aminopropyl)-N^4-acetyl-1,4-butanediamine [monoacetylspermidine (III)] which was isolated as its crystalline dihydrochloride in 54% yield. Deacetylation of monoacetylspermidine by hot hydrochloric acid readily gave pure spermidine trihydrochloride. The over-all yield was about 32% based on the acrylonitrile.

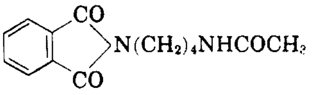
Crystalline monoacetylputrescine hydrochloride was prepared through reaction of hydrazine[2] with N^1-acetyl-N^4-phthaloyl-1,4-butanediamine (IV) which resulted from the reduction of γ-phthalimidobutyronitrile in acetic anhydride solution over platinum catalyst. γ-Phthalimidobutyronitrile was prepared[3] readily from potassium phthalimide and commercially available γ-bromobutyronitrile. The over-all yield of spermidine trihydrochloride was about 11% based on the γ-phthalimidobutyronitrile.

The reaction of monoacetylputrescine with γ-bromopropylphthalimide gave sirupy products which made the reaction unsuitable for the preparation of spermidine. The reaction of monobenzoylputrescine with γ-bromopropylphthalimide similarly was found by von Braun and Pinkernelle[4] to be unsatisfactory for the preparation of spermidine.

$H_2N(CH_2)_4NH(CH_2)_3NH_2\cdot 3HCl$ I

$CH_3COHN(CH_2)_4NH(CH_2)_2CN$ II

$CH_3COHN(CH_2)_4NH(CH_2)_3NH_2$ III



IV

EXPERIMENTAL

N^1-Acetyl-N^4-phthaloyl-1,4-butanediamine. A mixture of 1 g. of platinum oxide with a solution of 10 g. of γ-phthalimidobutyronitrile in 100 ml. of acetic anhydride was shaken with hydrogen at 25–26° and atmospheric pressure for 4.5 hours, 2.6 moles of hydrogen being absorbed. The filtered solution was added to 400 ml. of ice-water and the mixture was kept at room temperature for several hours. After filtration the solution was concentrated *in vacuo* (bath 80°) to a small volume, then diluted with water and the evaporation *in vacuo* was repeated to remove acetic acid. The crystalline residue was dissolved in absolute ethanol and the solvent was evaporated *in vacuo*. A solution of the substance in hot benzene, after filtration and concentration to 30 ml., deposited 5.3 g. of N^1-acetyl-N^4-phthaloyl-1,4-butanediamine. The filtrate after slight concentration gave at room temperature 0.2 g. of somewhat impure crystals of phthalimide, which was purified from ethanol, m.p. 233–234° (corr.).

Anal. Calc'd for $C_8H_5NO_2$: C, 65.30; H, 3.43; N, 9.52. Found: C, 65.43; H, 3.75; N, 9.61.

The filtrate from the phthalimide upon dilution with *n*-hexane deposited more crystals of N^1-acetyl-N^4-phthaloyl-1,4-butanediamine. Recrystallized once as colorless needles from benzene, the yield of compound melting at 141–144° was 5.6 g. or 46%. The analytical sample, m.p. 149–150° (uncorr.), was dried in an evacuated desiccator over calcium chloride.

Anal. Calc'd for $C_{14}H_{16}N_2O_3$: C, 64.60; H, 6.20; N, 10.77. Found: C, 64.03; H, 6.10; N, 10.65.

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[1] Danzig and Schultz, *J. Am. Chem. Soc.*, **74**, 1836 (1952).

[2] Ing and Manske, *J. Chem. Soc.*, 2348 (1926); Balenović, Bregant, Cerar, Fleš, and Jambrešić, *J. Org. Chem.*, **18**, 297 (1953).

[3] Goldberg and Kelly, *J. Chem. Soc.*, 1369 (1947).

[4] von Braun and Pinkernelle, *Ber.*, **70**, 1230 (1937).

Monoacetylputrescine hydrochloride. To a solution of 5 g. of N^1-acetyl-N^4-phthaloyl-1,4-butanediamine in 35 ml. of warm absolute ethanol was added 1.4 ml. of 85% hydrazine hydrate. After being refluxed for one hour, the mixture was kept at 25° for several hours, and the white solid was filtered off and washed thoroughly with absolute ethanol. The filtrate was concentrated *in vacuo* (bath 45°) to dryness. To a solution of the residue in 12 ml. of water was added sufficient hydrochloric acid to make the mixture just acid to Congo Red. After removal of phthaloylhydrazide, the solution was concentrated *in vacuo* to a thick sirup which was dissolved in 10 ml. of water. The solution, after removal of more phthaloylhydrazide, was concentrated *in vacuo* to a thick sirup which was taken up in absolute ethanol. Evaporation of the solvent *in vacuo* gave a crystalline residue, a solution of which in absolute ethanol upon concentration to 7 ml. and treatment with a drop of 1.5 *N* ethanolic hydrogen chloride solution deposited crystals of monoacetylputrescine hydrochloride; concentration of the filtrate gave more of the substance. Recrystallized from absolute ethanol, the yield of almost pure compound was 2.3 g. or 72%. The analytical sample, dried *in vacuo* over calcium chloride, melted at 140–141° (corr.).

Anal. Calc'd for $C_6H_{15}ClN_2O$: C, 43.24; H, 9.07; Cl, 21.28; N, 16.81. Found: C, 43.08; H, 8.86; Cl, 21.42; N, 17.06.

N^1-(2-Cyanoethyl)-N^4-acetyl-1,4-butanediamine hydrochloride. To a solution of 2 g. of monoacetylputrescine hydrochloride in 11 ml. of absolute ethanol were added 6.1 ml. of 1.962 *N* sodium hydroxide and a solution of 0.64 g. of recently distilled acrylonitrile in 6 ml. of absolute ethanol. After being kept at room temperature for 16 hours, the solution was diluted with 6 ml. of absolute ethanol, refluxed for one hour and then kept at room temperature for 2 hours. Sufficient 1.5 *N* ethanolic hydrogen chloride solution (9 ml.) was added to make the solution acid to Congo Red. Some crystals of sodium chloride were removed and the solvent was evaporated *in vacuo*. A solution of the residue in hot absolute ethanol, after filtration and concentration, gave at room temperature crystals of N^1-(2-cyanoethyl)-N^4-acetyl-1,4-butanediamine hydrochloride. A second crop was obtained by diluting the filtrate with ethyl ether and crystallizing the precipitated sirup from ethanol. The pure compound resulted from one recrystallization of the crude product from absolute ethanol; yield 1.85 g. or 70%; m.p. 143–144° (uncorr.).

Anal. Calc'd for $C_9H_{18}ClN_3O$: C, 49.19; H, 8.26; Cl, 16.14; N, 19.13. Found (dried at 25° *in vacuo* over calcium chloride): C, 49.28; H, 8.06; Cl, 16.32; N, 18.90.

Monoacetylspermidine dihydrochloride. A solution of 0.5 g. of N^1-(2-cyanoethyl)-N^4-acetyl-1,4-butanediamine hydrochloride in 33 ml. of absolute ethanol, which had been saturated with ammonia at 20–23°, was shaken for 45 minutes at 116–122° with *ca.* 0.4 g. of Raney nickel catalyst and hydrogen at 2400 p.s.i. The catalyst was removed by filtration and the solvent was evaporated, through a column, on the steam-bath. A solution of the residue in 10 ml. of absolute ethanol was kept at 20° during the addition of 4 ml. of 1.5 *N* ethanolic hydrogen chloride solution which precipitated some solid and made the solution acid to Congo Red. After the mixture had been diluted with 50 ml. of ethyl ether and kept at room temperature for one hour, the crude crystals of monoacetylspermidine dihydrochloride were collected on a funnel and washed with absolute ethanol-ether (1:2). The filtered solution of the dihydrochloride in 15 ml. of hot absolute ethanol, after concentration to *ca.* 4 ml., deposited at room temperature colorless crystals of the pure compound which was dried in an evacuated desiccator over calcium chloride; yield 318 mg. or 54%; m.p. 200–202° (uncorr., bath preheated to 170°).

Anal. Calc'd for $C_9H_{23}Cl_2N_3O$: C, 41.54; H, 8.91; Cl, 27.25; N, 16.15. Found: C, 41.31; H, 8.79; Cl, 27.18; N, 15.93.

Spermidine trihydrochloride. A solution of 0.63 g. of monoacetylspermidine dihydrochloride in 30 ml. of 37% hydrochloric acid was refluxed for 4 hours and then concentrated *in vacuo* to 1 ml. This solution, after dilution with 0.5 ml. of water and 7 ml. of ethanol, yielded at room temperature a first crop of pure spermidine trihydrochloride. The filtrate upon concentration and dilution with ethanol gave more of the compound which was recrystallized from aqueous ethanol. The yield of pure compound melting at 256–258° (corr.) was 0.525 g. or 85%. An additional 5% of somewhat impure product was obtained from the mother liquors.

Anal. Calc'd for $C_7H_{22}Cl_3N_3$: Cl, 41.77; N, 16.50. Found (dried at 100° *in vacuo*): Cl, 41.69; N, 16.24.

The product yielded a picrate melting at 213–214° (corr.). Dudley, Rosenheim, and Starling[5] reported a melting point of 210–212° for spermidine picrate.

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(5) Dudley, Rosenheim, and Starling, *Biochem. J.*, 21, 97 (1927).