20. *The Search for Chemotherapeutic Amidines. Part VI. Pyridines.*

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Two amidines containing pyridine nuclei, *αε-bis-(5-amidino-2-pyridyloxy)pentane dihydrochloride* and 2-p-*amidinostyrylpyridine*, have been synthesised. Attempts to convert αγ-bis-(5-cyano-2-pyridyloxy)propane into the corresponding diamidine failed.

Two of the most interesting of the therapeutic series of diamidines prepared in these laboratories are 4 : 4′-diamidino-αγ-diphenoxypropane and 4 : 4′-diamidino-αε-diphenoxypentane ("Propamidine" and "Pentamidine"). Both compounds have found use in the treatment of trypanosome infections in man, and the former has also been used as a bacteriostatic agent. The pyridine analogues [I; $n = 3$ or 5, $R = R_1 = C(:NH)\cdot NH_2$] were therefore deemed worthy of preparation.

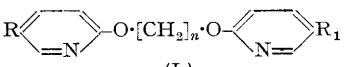
2-Amino-5-cyanopyridine was obtained by a modification of the method of Caldwell (*J. Amer. Chem. Soc.*, 1944, **66**, 1479), *viz.*, by treatment of 5-iodo-2-aminopyridine with cuprous cyanide in pyridine. The required cyanide was not obtained when the solvent was omitted. The cyanide gave the hitherto-unknown *2-hydroxy-5-cyanopyridine* on treatment with sodium nitrite in cooled sulphuric acid. We were unable to condense the hydroxy-compound with αε-dibromopentane, and only small yields of the required cyanide were obtained by treatment of the silver salt of the hydroxypyridine with dibromopentane in dioxan. The use of dry alcohol effected a slight improvement, whereas ethylene glycol as solvent afforded none of the required compound. A startling improvement was accomplished by using specially dried reactants and solvents. The hydroxypyridine (silver salt) and dibromopentane (both dried intensively over phosphoric oxide) gave, in dioxan previously dried by refluxing with sodium for 48 hours, excellent yields of *αε-bis-(5-cyano-2-pyridyloxy)pentane* (I; $n = 5$, $R = R_1 = CN$).

Conversion into the corresponding iminoether occurred satisfactorily in chloroform and

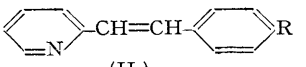
absolute alcohol treated with dry hydrogen chloride. It was found best not to purify the iminoether base, as considerable reconversion into the dicyanide occurred on attempted recrystallisation, a manifestation of the ease with which many iminoether bases decompose into alcohol and cyanide. *αε-Bis-(5-amidino-2-pyridyloxy)pentane dihydrochloride* was obtained by treatment of the iminoether base with ammonium chloride.

Under conditions similar to those used for the pentane derivative, *αγ*-bis-(5-cyano-2-pyridoxy)propane (I; $n = 3$, $R = R_1 = CN$) was prepared, but in poor yields. All attempts to convert this compound into the corresponding iminoether failed.

Yet another interesting amidine of the carbocyclic series is 4 : 4′-diamidinostilbene (" Stilbamidine "), and we accordingly decided to investigate the analogue [II, $R = C(:NH)\cdot NH_2$],



(I.)



(II.)

in which the pyridine ring might possibly supply the second basic centre which is presumably necessary for trypanocidal activity (see Ashley *et al.*, *J.*, **1942**, 103).

p-Cyanobenzaldehyde condensed easily and smoothly with α-picoline in the presence of acetic anhydride (cf. Chiang and Hartung, *J. Org. Chem.*, **1945**, **10**, 21) to give 2-p-*cyanostyrylpyridine* (II, R = CN), and the corresponding iminoether was obtained by the usual method. This, on treatment with ammonium chloride, gave 2-*p*-amidinostyrylpyridine, isolated as its *dihydrochloride*, readily soluble in water. It showed no trypanocidal activity, but the diamidine [I; $n = 5$, $R = R_1 = C(:NH)\cdot NH_2$] exhibited slight activity against *T. equiperdum* in mice. It also inhibited the growth of *Staph. aureus* at a dilution of 1 : 64,000, but even so, this activity is less than half that of the carbocyclic analogue (" Pentamidine ").

Experimental.

2-Amino-5-cyanopyridine.—5-Iodo-2-aminopyridine (11 g.) and dry cuprous cyanide (5 g.) were refluxed for 30 mins. in dry pyridine (12 c.c.). The pyridine was distilled off, and the residue rapidly distilled in a vacuum with a naked flame, using a wide tube receiver and without employing an air leak, the required compound being obtained as a pale yellow solid (4·6 g., 75·5%), m. p. 164°.

2-Hydroxy-5-cyanopyridine.—Sodium nitrite (15·4 g.) was added in small quantities to a well-stirred solution of 2-amino-5-cyanopyridine (15 g.) in sulphuric acid (18·5 c.c., *d* 1·84) and water (180 c.c.) kept at 0—5°. The solution remained clear at first, but evolution of nitrogen soon occurred with the separation of a white solid. The thick suspension was allowed to reach room temperature and was then heated on the steam-bath for 30 mins. The cooled suspension was filtered, and the residue recrystallised (charcoal) from water (400 c.c.), giving the required *compound* (13·1 g., 87·5%), m. p. 259—260° (Found : C, 59·8; H, 3·3; N, 23·1. $C_6H_4ON_2$ requires C, 60·0; H, 3·3; N, 23·3%). To a solution of this compound (1·2 g.) in N-sodium hydroxide (10·0 c.c.) and water (25 c.c.), 0·1N-silver nitrate solution was added, with stirring. The semi-gelatinous suspension was heated on the steam-bath for 30 mins. to complete the reaction, and the silver salt was filtered off and washed well with water and alcohol.

αε-Bis-(5-cyano-2-pyridyloxy)pentane.—The silver salt (11·35 g.) was dried in the steam-oven for 24 hours and over phosphoric oxide in a vacuum for 12 hours, then refluxed for 24 hours in pure dry dioxan (125 c.c.) with 1 : 5-dibromopentane (4·0 c.c.) which had been dried over phosphoric oxide. The suspension was filtered hot, and the filtrate deposited a white solid, which was washed with ether. The filtrate from this was diluted with 5 vols. of water, giving a soft solid; washing with ether removed an oil, and the solid remaining was combined with that obtained above and re-crystallised (charcoal) from alcohol (400 c.c.), giving the required *compound* as a white crystalline powder, m. p. 170—171° (5·4 g., 70%) (Found : C, 66·4; H, 5·3; N, 18·10. $C_{17}H_{16}O_2N_4$ requires C, 66·5; H, 5·2; N, 18·15%).

αε-Bis-(5-amidino-2-pyridyloxy)pentane Dihydrochloride.—The foregoing cyano-compound (1·5 g.) in dry chloroform (40 c.c.) and dry alcohol (1·25 c.c.) was saturated at −10° with dry hydrogen chloride. The tube was securely stoppered and kept at 0° for 13 days. The product was added to excess of sodium carbonate solution at 0°, and the liberated iminoether base extracted with chloroform. The chloroform solution was washed free from alkali with ice-water, dried (sodium sulphate), and the solvent removed under reduced pressure at a low temperature. The residue was dissolved in alcohol (25 c.c.) by warming, and an aqueous solution of ammonium chloride (0·588 g., 2·05 mols.) added. The resulting solution was kept at *ca.* 50° for 12 hours, and the product poured into acetone (300 c.c.). The sticky precipitate hardened to a white solid which was crystallised from alcohol (50 c.c.), giving the required *amidine dihydrochloride* (1·0 g., 50%) (Found : C, 48·2; H, 5·7; N, 19·75; Cl, 16·6. N : Cl = 3·0 : 1. $C_{17}H_{24}O_2N_6Cl_2,\frac{1}{2}H_2O$ requires C, 48·1; H, 5·9; N, 19·8; Cl, 16·7%).

αγ-Bis-(5-cyano-2-pyridyloxy)propane.—Quantities and conditions were as for the corresponding pentane derivative, but dry 1 : 3-dibromopropane (3·5 c.c.) was used. The reaction mixture, after refluxing, was filtered hot, and the cooled filtrate, from which no solid separated, was then diluted with 5 vols. of water. The collected precipitate was extracted with boiling alcohol, leaving a residue, m. p. 230—240°, probably impure unchanged 2-hydroxy-5-cyanopyridine. The alcoholic extract deposited a semi-crystalline mass which was recrystallised from alcohol (50 c.c.) to give the required *compound*, m. p. 134—135° (2·9 g., 22%) (Found : C, 64·2; H, 4·5; N, 20·0. $C_{15}H_{12}O_2N_4$ requires C, 64·2; H, 4·3; N, 20·0%).

2-p-*Cyanostyrylpyridine.*—A mixture of *p*-cyanobenzaldehyde (2 g.), α-picoline (1·44 c.c.), and acetic

anhydride (1·45 c.c.) was refluxed for 10 hours. The mixture was cooled, treated with ice-water, and the solid collected, washed well with water, crystallised twice from alcohol, and dried in a vacuum at 78° to give the *cyanide*, m. p. 128—130° (1·3 g., 42%) (Found : N, 13·6. $C_{14}H_{10}N_2$ requires N, 13·6%).

2-*Styrylpyridine-4'-iminoether*.—A mixture of 2-*p*-cyanostyrylpyridine (0·8 g.), lime-dried alcohol (0·57 c.c.), and dry chloroform (10 c.c.) was saturated with dry hydrogen chloride below 0° and then left at 0° for 6 days. After treatment with sodium hydroxide in the usual manner, the chloroform solution of the iminoether was evaporated in a vacuum. The iminoether (m. p. 85—87°) which crystallised on cooling was washed with ligroin and dried in a vacuum (0·78 g., 80%). It was used for the preparation of the amidine without further purification.

2-p-*Amidinostyrylpyridine*.—A mixture of 2-styrylpyridine-4'-iminoether (0·75 g., 1 mol.), ammonium chloride (0·17 g., 1·05 mol.), and aqueous alcohol (1 c.c.; 4 c.c.) was heated at 60—65° for 7 hours and then left in a warm place (30—40°) for a further 16 hours. Concentrated hydrochloric acid was then added until the mixture was just acid to Congo-red, and the precipitated *dihydrochloride* of the amidine (0·57 g.) was filtered off, washed with acetone, and crystallised from alcohol containing a little water (30 c.c.; 1·5 c.c.). The amidine dihydrochloride separated at 0° in very pale yellow crystals, m. p. 282—283° (0·26 g., 42%) (Found : N, 13·9; Cl, 23·2; N : Cl = 3·04 : 2. $C_{14}H_{13}N_3,2HCl$ requires N, 14·2; Cl, 24·0%).

The authors wish to thank Drs. A. J. Ewins, F.R.S., and H. J. Barber for their interest in this work, Mr. S. Bance, B.Sc., A.R.I.C., and his staff for the semi-microanalyses, and the Directors of May and Baker Ltd. for permission to publish these results.

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[*Received, May 6th*, 1946.]