tron impact) m/e 285 ($M^+ - H_2O$); mass spectrum (chemical ionization, methane) m/e 304 ($M + H^+$). The compound was too unstable for elemental analysis.

A sample of amine **3** was converted to a hydrochloride, mp 194–230°, by treatment of a solution of it in ethanol–ether with gaseous hydrogen chloride at 0°. Repeated recrystallization of the salt from ethanol–ether gave white crystals, mp 241–243°; the mixture melting point with (±)-cherylline hydrochloride (see below) was undepressed. The amine regenerated upon basicification of this salt was identical with cherylline in thin layer chromatographic behavior.

(±)-Cherylline (1). Method A.—A solution of 120 mg (0.396 mmol) of amine **3** in 50 ml of 4% aqueous ammonia was refluxed for 7 hr. The resulting pale yellow solution was acidified with concentrated hydrochloric acid, neutralized with sodium bicarbonate, and extracted thoroughly with ethyl acetate. The residue obtained upon evaporation of the solvent was crystallized from ether–hexane to give 93 mg (82%) of crude (±)-cherylline, mp 125–200°, identical in thin layer chromatographic behavior with authentic material[9] except for a trace of a polar impurity (ethyl acetate–chloroform–ethanol, 85:11:4). Several recrystallizations from benzene–methanol gave pure (±)-cherylline: mp 209–212° (reported[5] mp 215–216°); identical with (−)-cherylline[9] in tlc, nmr, uv, and mass spectrum; ir (KBr) 2.99, 6.21, 6.29, 6.64, 7.86, 7.98, and 8.91 μ; nmr (acetone-d_6, 60 MHz) δ 2.28 (s, 3), 2.38, 2.82 (ABX pattern, 2, J_{AB} = 11.0 Hz, J_{AX} = 7.5 Hz, J_{BX} = 5.5 Hz), 3.42 (s, 2), 3.52 (s, 3), 3.98 (dd, 1, J = 5.5 and 7.5 Hz), 6.27 (s, 1), 6.45 (s, 1), 6.62, 6.93 (AA'BB' pattern, 4, J = 8.5 Hz); uv max (ethanol) 226 nm (sh, ε 14,000), 280 (3900), 285 (4000), and 295 (sh, 2500); mass spectrum m/e 285, 242, 241, 227, 225, 211, 210, 181.

Anal. Calcd for $C_{17}H_{19}NO_3$: C, 71.56; H, 6.71; N, 4.91. Found: C, 71.40; H, 6.71; N, 4.69.

A sample of (±)-cherylline was converted to its hydrochloride and recrystallized from ethanol–ether to give 1 HCl, mp 240–243° (the salt first melted at 185°, resolidified at *ca.* 190°, then melted again at the specified temperature).

***p*-Hydroxy-α-{[(3-hydroxy-4-methoxybenzyl)amino]methyl}-benzyl Alcohol (10).**—A mixture of 836 mg (5.50 mmol) of isovanillin[14] (**9**), 1.04 g (5.50 mmol) of (±)-octapamine hydrochloride[14] (**8**), and 500 mg of sodium bicarbonate in 50 ml of methanol was stirred at 50° for 30 min. The reaction mixture was cooled in an ice bath, 1.00 g (26.3 mmol) of sodium borohydride was slowly added, and the resulting solution was stirred at room temperature for 30 min. Most of the solvent was evaporated under reduced pressure; the residue was dissolved in dilute hydrochloric acid, neutralized with sodium bicarbonate, and extracted with ethyl acetate to give 1.05 g (66%) of crude crystalline amine **10**, mp 115–135°. Two recrystallizations from ethyl acetate–methanol afforded the pure compound: mp 150–152°; ir (KBr) 3.0, 6.21, 6.29, 6.64, 7.97, 8.21, and 9.74 μ; nmr (DMSO-d_6, 90 MHz) δ 2.62 (d, 2, J = 6 Hz, NCH_2), 3.67 (s, 2, $ArCH_2N$), 3.74 (s, 3, OCH_3), 4.62 (t, 1, J = 6 Hz, ArCHO), 6.56–7.23 (m, 7, aromatic).

A sample of **10** was treated with excess acetic anhydride in pyridine at −10° to afford the tetraacetyl derivative as a colorless glass.

Anal. Calcd for $C_{24}H_{27}NO_8$: C, 63.01; H, 5.95; N, 3.06. Found: C, 62.76; H, 6.23; N, 2.79.

(±)-Cherylline (1). Method B.—To a solution of 100 mg (0.330 mmol) of phenolic amine **10** in 30 ml of ethyl formate–ethanol 3:1 was added 200 mg of potassium carbonate and 1 g of 3-Å molecular sieves. The mixture was refluxed under nitrogen for 8 hr. The solids were filtered and washed with ethanol, and the combined filtrates were evaporated under reduced pressure.

A suspension of the resulting white solid in 1,2-dimethoxyethane was treated with excess lithium aluminum hydride and the mixture was refluxed under nitrogen for 50 hr. The excess hydride was decomposed with saturated aqueous potassium sodium tartrate solution and the resulting suspension was refluxed for 3 hr. The solvent was decanted and the residue was dissolved in dilute hydrochloric acid, neutralized with sodium bicarbonate, and extracted with ethyl acetate. Crystallization of the crude product from ether–hexane afforded 62 mg (66%) of (±)-cherylline (**1**), identical in all respects with the material prepared by method A above.

(14) Aldrich Chemical Co., Milwaukee, Wis.

Registry No.—**1**, 26996-80-5; **1** HCl, 29002-62-8; **3**, 29002-63-9; **4**, 29002-64-0; **6**, 29002-65-1; **6** HCl, 29002-66-2; **7**, 29038-87-7; **10**, 29002-67-3.

Notes

A New Synthesis of 1,3-Dimethylcytosines

KEITARO SENGA, FUMIO YONEDA,* AND SADAO NISHIGAKI

Pharmaceutical Institute, School of Medicine, Keio University, Shinanomachi, Shinjuku-ku, Tokyo 160, Japan

Received October 20, 1970

The discovery of 3-methylcytidine[1] and 1-methyladenosine[2] as minor basic components of nucleic acid stimulated our interest in the chemistry of the iminopyrimidines, which have customarily been made by alkylation of the parent aminopyrimidines. In this note we will describe a new synthesis of 1,3-dimethylcytosine derivatives as a part of the exploitation of our preparative methods of pyrimidine derivatives of the imino type.

Heating of 6-amino-1,3-dimethyluracil (I) with phosphorous oxychloride at 240–250° for 10 hr afforded 6-chloro-1,3-dimethylcytosine (Ia) in 92% yield. The structure of Ia was assigned on the basis of the following evidence. Compound Ia shows a secondary amino stretching absorption band at 3250 cm^{-1} (Nujol). The nuclear magnetic resonance spectrum (CF_3COOH) of Ia shows singlets at 3.73 (CH_3), 3.86 (CH_3), and 6.67 ppm (C_5 H in pyrimidine), and two broad bands at 7.72 and 8.18 ppm ($=N^+H_2$). The mass spectrometry reveals a parent ion (m/e 173) and M + 2 ion, which suggests that one chlorine atom is contained in the molecule. The structure of Ia was finally established by catalytic dechlorination over palladium/carbon to the known 1,3-dimethylcytosine[3–6] (Ib) and by its conversion into the starting material I by treatment with aqueous sodium

(1) (a) R. H. Hall, *Biochem. Biophys. Res. Commun.*, **12**, 36b (1963); (b) R. H. Hall, *Biochemistry*, **4**, 661 (1965).

(2) (a) A. Hampton and D. I. Magrath, *J. Amer. Chem. Soc.*, **79**, 3250 (1957); (b) A. Hampton and M. H. Maguire, *ibid.*, **83**, 150 (1961).

(3) G. H. Hilbert, *ibid.*, **56**, 190 (1934).

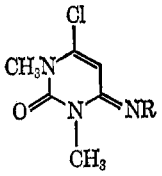
(4) The infrared spectroscopic data of Ib were reported by Angell: C. L. Angell, *J. Chem. Soc.*, 504 (1961).

(5) G. W. Kenner, C. B. Reese, and A. R Todd, *ibid.*, 855 (1955).

(6) P. Brookes and P. D. Lawley, *ibid.*, 1348 (1962).

TABLE I

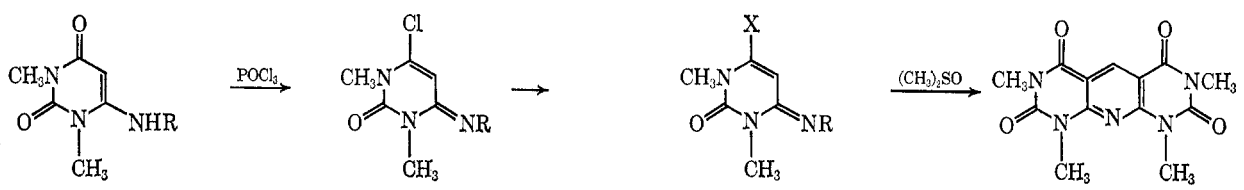
6-CHLORO-1,3-DIMETHYLCYTOSINES



Compd	R	Reaction time, hr	Reaction temp,[a] °C	Purification[b]	Yield, %	Mp, °C	Empirical formula	Calcd, % C	Calcd, % H	Calcd, % N	Found, % C	Found, % H	Found, % N
Ia	H	10	250	A[c]	92	150	$C_6H_8ClN_3O$	41.51	4.65	24.20	41.44	4.89	24.02
IIa	n-C_3H_7	5	240	B[d]	43	63	$C_9H_{14}ClN_3O$	50.12	6.54	19.49	50.01	6.35	19.30
IIIa	i-C_3H_7	3	240	B[d]	37	52	$C_9H_{14}ClN_3O$	50.12	6.54	19.49	50.08	6.58	19.68
IVa	C_6H_{11}	3	240	C[e]	92	108	$C_{12}H_{18}ClN_3O$	56.35	7.11	16.43	56.66	6.97	16.44
Va	C_6H_5	5	240	D[d]	88	103	$C_{12}H_{12}ClN_3O$	57.72	4.88	16.97	57.71	4.75	16.82
VIa	$C_6H_5CH_2$	3	240	C[f]	100	81	$C_{13}H_{14}ClN_3O$	59.21	5.35	15.93	59.33	5.36	16.10

[a] Temperature of oil bath. [b] A, recrystallization from chloroform; B, sublimation at 130° (1 mm); C, recrystallization from aqueous ethanol; D, sublimation at 200° (1 mm). [c] Pale yellow prisms. [d] Colorless powder. [e] Colorless prisms. [f] Colorless needles.

SCHEME I



I, R = H
II, R = n-C_3H_7
III, R = i-C_3H_7
IV, R = C_6H_{11}
V, R = C_6H_5
VI, R = $C_6H_5CH_2$

Ia, R = H
IIa, R = n-C_3H_7
IIIa, R = i-C_3H_7
IVa, R = C_6H_{11}
Va, R = C_6H_5
VIa, R = $C_6H_5CH_2$

Ib, R = H; X = H
Ic, R = H; X = NH_2
Id, R = H; X = C_6H_5NH
Ie, R = H; X = CH_3O
If, R = H; X = SH
Ig, R = C_6H_5; X = C_6H_5NH
Ih, R = H; X = SO_3H

bicarbonate. Similarly, heating several 6-(secondary amino)-1,3-dimethyluracils with phosphorous oxychloride gave the corresponding 6-chloro-1,3-dimethylcytosines in good yields (Table I).

The 6-chloro-1,3-dimethylcytosines obtained here served as starting materials for several nucleophilic reactions. For example, displacements of the chlorine in Ia by amino, anilino, alkoxy, and mercapto groups were carried out to yield the respective products. The results of these reactions are summarized in Table II. It is interesting to note that the imino group of 1,3-dimethylcytosines is considerably stable against acid hydrolysis. For example, heating of Ia in concentrated hydrochloric acid at 150–160° for 2 hr gave only a 13% yield of 1,3-dimethylbarbituric acid, with most starting material being recovered.

The reaction of Ia with excess aniline yielded 6-anilino-1,3-dimethyl-4-*N*-phenylcytosine (Ig), which was identical with the product obtained from 6-anilino-1,3-dimethylcytosine (Id) and aniline. 1,3-Dimethyl-6-thiocytosine (If) was also obtained in lower yield by the conventional thiation of I with phosphorous pentasulfide in pyridine. Oxidation of If with hydrogen peroxide in glacial acetic acid gave 1,3-dimethylcytosine-6-sulfonic acid (Ih), whose structure was established by alternative synthesis from Ia and sodium bisulfite. Heating of If in dimethyl sulfoxide gave 1,3,7,9-tetramethyl-2,4,6,8-tetraoxo-1,2,3,4,6,7,8,9-octahydropyrido[2,3-*d*:6,5-*d*′]dipyrimidine.[7-9]

Experimental Section[10]

General Procedure for Synthesis of 6-Chloro-1,3-dimethylcytosines (Ia–VIa).—A mixture of 0.1 mol of a 6-amino-1,3-dimethyluracil (I–VI) and 150 ml (1.64 mol) of phosphorous oxychloride was refluxed as described in Table I. After the excess of phosphorous oxychloride was evaporated under reduced pressure, the residue was dissolved in 80 ml of water. The solution was made alkaline with 5% aqueous ammonia, extracted with chloroform (ten 50-ml portions), dried over sodium sulfate, and concentrated to dryness. The residue was recrystallized from an appropriate solvent. When crystals were separated from the alkaline solution, they were collected by filtration, washed with water, dried, and recrystallized from an appropriate solvent.

(7) H. Bredereck, F. Effenberger, and R. Sauter, *Chem. Ber.*, **95**, 2049 (1962).

(8) R. C. Elderfield and M. Wharmby, *J. Org. Chem.*, **32**, 1638 (1967).

(9) K. Senga, F. Yoneda, and S. Nishigaki, *Chem. Pharm. Bull.*, **19**, 215 (1971).

(10) All melting points are uncorrected. Infrared spectra were recorded on a Japan Spectroscopic Co., Ltd., Model IR-E spectrometer; nmr spectra with a Japan Electron Optics Lab. Co., Ltd., Model JNM-C-60-H spectrometer.

TABLE II

REACTION OF 6-CHLORO-1,3-DIMETHYLCYTOSINE WITH SOME NUCLEOPHILES

Compd	R	Reactant (1 equiv)	Reaction time, hr	Temp, °C	Purification[a]	Yield, %	Mp, °C	Empirical formula	Calcd, % C	Calcd, % H	Calcd, % N	Found, % C	Found, % H	Found, % N
Ic	NH_2	Excess NH_3 in CH_3OH	8	95	A[b]	56	310	$C_6H_{11}ClN_4O$[e]	37.78	5.82	29.38	37.86	5.85	29.47
Id	C_6H_5NH	$C_6H_5NH_2$	3	190	B[c]	83	204	$C_{12}H_{14}N_4O$	62.59	6.13	24.33	62.69	6.10	24.11
Ie	CH_3O	CH_3ONa in CH_3OH	2	Reflux	C[b]	96	156	$C_7H_{11}N_3O_2$	49.69	6.55	24.84	49.86	6.70	25.11
If	SH	Aqueous $NaSH$	3	Reflux	D[d]	89	260	$C_6H_9N_3OS$	42.09	5.30	24.54	41.87	5.24	24.61

[a] A, recrystallization from a mixture of methanol and acetone; B, recrystallization from aqueous ethanol; C, recrystallization from benzene; D, recrystallization from aqueous dimethyl sulfoxide. [b] Colorless needles. [c] Colorless powder. [d] Pale yellow prisms. [e] Hydrochloride.

1,3-Dimethylcytosine (Ib).—A solution of 3.47 g (0.02 mol) of Ia and 2 ml of concentrated aqueous ammonia in 30 ml of methanol containing 0.3 g of 10% palladium/carbon was hydrogenated at room temperature and at atmospheric pressure. Hydrogenation was stopped when the theoretical volume (448 ml) of hydrogen was consumed. The solution was filtered and evaporated to dryness. The residue was dissolved in 15 ml of water, made alkaline with 5% aqueous ammonia, and extracted with chloroform (five 30-ml portions). The chloroform was dried over sodium sulfate, filtered, and evaporated to dryness to give 1.9 g (68%) of pale yellow powder. Sublimation at 200° (0.5 mm) afforded an analytical sample, mp 143–144°.

Anal. Calcd for $C_6H_9N_3O$: C, 51.78; H, 6.52; N, 30.20. Found: C, 52.07; H, 6.48; N, 29.94.

6-Amino-1,3-dimethylcytosine Hydrochloride (Ic).—A suspension of 1.74 g (0.01 mol) of Ia in 50 ml of saturated methanolic ammonia was heated in sealed tube as described in Table II. After cooling, the reaction mixture was evaporated to dryness. The residue was dissolved in 50 ml of 2 *N* hydrochloric acid with warming. After standing overnight at room temperature, the precipitated crystals were collected by filtration, washed with a small amount of chilled 2 *N* hydrochloric acid, and dried to give 1.0 g of pale yellow needles.

1,3-Dimethyl-6-thiocytosine (If). A.—A suspension of 2.6 g (0.015 mol) of Ia and 2.1 g (0.015 mol) of 40% aqueous sodium hydrosulfide in 30 ml of water was heated under the conditions described in Table II. After cooling, the precipitates were collected by filtration, washed with water, and dried to give 2.3 g of pale yellow powder.

B.—A mixture of 0.93 g (0.006 mol) of I and 2.3 g (0.012 mol) of phosphorous pentasulfide in 10 ml of pyridine was refluxed for 5 hr. After evaporating pyridine under reduced pressure, 50 ml of water was added to the resulting residue. The crystals which separated were collected by filtration, washed with water, and dried to give 0.35 g (34%) of If.

6-Anilino-1,3-dimethyl-4-*N*-phenylcytosine (Ig). A.—A mixture of 0.87 g (0.005 mol) of Ia, 0.94 g (0.01 mol) of aniline, and 3 drops of concentrated hydrochloric acid was heated at 200° for 3 hr. The reaction mixture was dissolved in 10 ml of ethanol and neutralized with aqueous ammonia. The precipitated crystals were collected by filtration, washed with water, dried, and recrystallized from aqueous ethanol to give 0.8 g (52%) of colorless needles, mp 179–181°.

Anal. Calcd for $C_{18}H_{18}N_4O$: C, 70.56; H, 5.92; N, 18.29. Found: C, 70.54; H, 5.82; N, 18.37.

B.—A mixture of 0.5 g (0.002 mol) of 6-anilino-1,3-dimethylcytosine (Id), 0.19 g (0.002 mol) of aniline, and 1 drop of concentrated hydrochloric acid was heated for 1.5 hr at 170°. The reaction mixture was crushed in water, collected by filtration, and washed with water. The crushed mass was recrystallized from aqueous ethanol to give 0.45 g (75%) of pale yellow needles, which was identical with the product obtained in A.

1,3-Dimethylcytosine-6-sulfonic Acid (Ih). A.—A suspension of 0.87 g (0.005 mol) of Ia and 1.04 g (0.01 mol) of sodium bisulfite in 10 ml of water was stirred at room temperature for 25 min. The precipitates were collected by filtration, washed with water, and dried. Recrystallization from aqueous dimethyl sulfoxide gave 1.1 g (100%) of colorless powder, mp >360°.

Anal. Calcd for $C_6H_9N_3O_4S$: C, 32.87; H, 4.14; N, 19.17. Found: C, 32.97; H, 4.19; N, 19.37.

B.—To a suspension of 0.68 g (0.004 mol) of If in 5 ml of glacial acetic acid 10 ml of 30% aqueous hydrogen peroxide was added dropwise at room temperature. After being stirred at room temperature for 1.5 hr, the precipitates were collected by filtration, washed with water, and dried to give 0.75 g (85%) of Ih.

1,3,7,9-Tetramethyl-2,4,6,8-tetraoxo-1,2,3,4,6,7,8,9-octahydropyrido[2,3-*d*:6,5-*d'*]dipyrimidine.—A suspension of 0.51 g (0.003 mol) of If in 10 ml of dimethyl sulfoxide was refluxed for 5 hr. After cooling, the precipitated crystals were collected by filtration, washed with 50 ml of acetone, and dried to give 0.27 g (60%) of pale yellow crystals.

Registry No.—Ia, 28795-51-9; Ib, 6749-87-7; Ic, 28795-53-1; Id, 28795-54-2; Ie, 28795-55-3; If, 28860-32-4; Ig, 28795-56-4; Ih, 28795-57-5; IIa, 28795-58-6; IIIa, 28795-59-7; IVa, 28795-60-0; Va, 28795-61-1; VIa, 28795-62-2.