(0.04 mole) of diethanolamine in 125 ml of 2-ethoxyethanol was refluxed for 18 hr. The hot reaction mixture was treated with charcoal, diluted with 50 ml of EtOH, and filtered. The hot filtrate was adjusted to pH 1 by addition of ethanolic HCl. The acidic solution was then chilled and the resulting precipitate was collected by filtration and air dried. The yield of IIIa, isolated as a dihydrochloride salt, was 2.1 g (30% yield), mp 205° dec. An analytical sample was obtained by recrystallization from absolute EtOH, mp 205° dec. *Anal.* ($C_{11}H_{18}N_6O_2 \cdot 2HCl \cdot H_2O$) C, H, N, Cl.

**9-[Bis(β-chloroethyl)aminoethyl]adenine (Ia).**—A finely powdered suspension of 1.79 g (0.005 mole) of IIIa·2HCl·$H_2O$ and 150 ml of $SOCl_2$ was heated on the steam bath for 1 hr. Excess $SOCl_2$ was then removed from the reaction mixture under reduced pressure as heating was continued on the steam bath. The residue was recrystallized from absolute EtOH to give 1.18 g (68% yield) of analytically pure dihydrochloride salt of Ia, mp 177–179°, $\lambda_{max}^{EtOH}$ 260 mμ (ε 15,000). *Anal.* ($C_{11}H_{16}Cl_2N_6 \cdot 2HCl \cdot H_2O$) C, H, Cl, N.

**9-[Bis(β-hydroxyethyl)aminoethyl]hypoxanthine (IIIc).**—A solution of 2.97 g (0.01 mole) of 6-methylthio-9-[bis(β-hydroxyethyl)aminoethyl]purine[9] (IIIb), 10 ml of 30% $H_2O_2$, 10 ml of concentrated HCl, and 100 ml of $H_2O$ was heated on the steam bath for 3 hr. The unreacted $H_2O_2$ was then decomposed by the slow addition of 10 g of $MnO_2$. After permitting to stand overnight, the solid was filtered and the filtrate was evaporated to dryness. The resulting yellow syrupy residue was covered with 100 ml of 95% EtOH, heated to boiling, and allowed to cool slowly. The white crystalline product IIIc, on cooling, separated as the analytically pure monohydrochloride salt, mp 124–126°. *Anal.* ($C_{11}H_{17}N_5O_3 \cdot HCl \cdot H_2O$) C, H, N.

**9-[Bis(β-chloroethyl)aminoethyl]hypoxanthine (Ib).**—A fine suspension of 1.60 g (0.005 mole) of IIIc·HCl·$H_2O$ in 150 ml of $SOCl_2$ was refluxed for 6 hr. During this time a complete solution was obtained, followed by the precipitation of crystalline solids. The reaction mixture was evaporated to dryness and the residue was recrystallized from 40 ml of absolute EtOH to give 1.40 g (75% yield) of analytically pure Ib·2HCl, mp 205° dec, $\lambda_{max}^{H_2O}$ 250 mμ (ε 11,300). *Anal.* ($C_{11}H_{15}Cl_2N_5O \cdot 2HCl$) C, H, $Cl^-$, N.

**2-Amino-6-methylthio-9-[bis(β-hydroxyethyl)aminoethyl]-purine (IIId).**—A solution of 12.2 g (0.05 mole) of 2-amino-6-methylthio-9-(β-chloroethyl)purine[9] and 10.5 g (0.1 mole) of diethanolamine in 250 ml of 2-ethoxyethanol was refluxed for 18 hr. The red solution was evaporated *in vacuo* on a steam bath. The resulting red residue was covered with 75 ml of absolute EtOH and allowed to stand overnight at 5°. The light tan solid was collected, washed with cold EtOH, and air dried to give 11.1 g (71% yield) of crude product, mp 157–159°. An analytical sample of IIId was obtained by recrystallization from absolute EtOH; mp 161–162°. *Anal.* ($C_{12}H_{20}N_6O_2S$) C, H, N.

**9-[Bis(β-hydroxyethyl)aminoethyl]guanine (IIIe).**—A solution of 6.24 g (0.02 mole) of IIId, 20 ml of 30% $H_2O_2$, and 200 ml of glacial AcOH was stirred at room temperature for 24 hr. The resulting light yellow solution was added to 1 l. of $Me_2CO$ with vigorous stirring. The mixture was then allowed to stand for 30 min and the supernatant liquid was separated by decantation. The resulting white residue was stirred with 200 ml of absolute EtOH for 30 min and the resulting hygroscopic solid was collected by rapid filtration. The solid was then dissolved in 100 ml of $H_2O$ and the solution was stirred with 8 g of Amberlite IR-45 for 30 min. The weakly basic ion-exchange resin was then separated by filtration. The process was repeated three times. The resulting aqueous filtrate was evaporated to dryness and the residue was recrystallized from absolute EtOH to give 2.1 g (37% yield) of IIIe, mp 192–194° dec. *Anal.* ($C_{11}H_{18}N_6O_3 \cdot H_2O$) C, H, N.

**9-[Bis(β-chloroethyl)aminoethyl]guanine (Ic).**—A finely divided suspension of 1.50 g (0.005 mole) of IIIe·$H_2O$ in 200 ml of $SOCl_2$ was refluxed for 5 hr. Although a complete solution was not formed during this period, a definite change in the appearance of the suspended solids was noted. The reaction mixture was evaporated to dryness under reduced pressure. The resulting off-white solid product was covered with 100 ml of absolute EtOH, heated to reflux, and then again evaporated to dryness. The crude product was recrystallized from 50 ml of absolute EtOH to give 1.10 g (62% yield) of analytically pure Ic·HCl, mp 218–220° dec, $\lambda_{max}^{H_2O}$ 253 mμ (ε 11,000), $\lambda_{max}^{H_2O}$ 269 mμ (ε 8200). *Anal.* ($C_{11}H_{16}Cl_2N_6O \cdot HCl$) C, H, N.

**Acknowledgment.**—The authors wish to express their appreciation to Mrs. Margaret L. Rounds and Mr. John R. Gravatt for their valuable assistance in performing analytical and instrumental measurements.

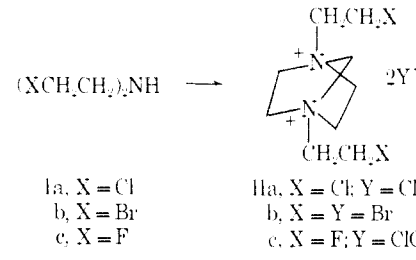
## Antineoplastic Agents. XXV. 1,4-Diazabicyclo[2.2.1]heptanes[1]

DYRAL C. FESSLER, GEORGE R. PETTIT,[2] AND JOSEPH A. SETTEPANI

*Departments of Chemistry, Arizona State University, Tempe, Arizona 85281, and University of Maine, Orono, Maine 04468*

*Received January 28, 1969*

Mannich-type reactions employing bis(2-chloroethyl)amine have been used to prepare a number of nitrogen mustards derived from acetophenones,[3a,b] acetylenic carbinols,[3c] natural products,[3d] coumarins,[3e] thiophenes,[3b] benzimidazoles,[3f] and cyclic ketones.[3g] During the course of our initial studies in this area, condensing 1 mole of formaldehyde with 2 moles of bis(2-chloroethyl)amine was found to yield a new quaternary ammonium salt, shown to be 1,4-bis(2-chloroethyl)-1,4-diazabicyclo[2.2.1]heptane diperchlorate (IIa).[1,4] The new heterocyclic compound IIa demonstrated signifi-



Ia, X = Cl; b, X = Br; c, X = F

IIa, X = Cl; Y = $ClO_4$; b, X = Y = Br; c, X = F; Y = $ClO_4$

cant activity against the Walker 256 carcinoma.[4] To provide further examples of this new heterocyclic system for evaluation as possible cancer chemotherapeutic agents and to determine the scope of the condensation reaction, an analogous study was extended to several N-alkyl- and N-benzyl-substituted 2-haloethylamines.

Treating ethanol solutions of amines Ia–c and IIIa–h with 37% formalin at room temperature led to quaternary ammonium salts IIa–c and IVa–h (Table I) in good yields. Generally, reaction was complete within 24 hr, but the less reactive[5] fluoro derivative Ic required 144 hr. Attempted condensation with the relatively poor nucleophile N-[2-chloroethyl-3,5-bis(trifluoromethyl)benzyl]amine led only to recovery of starting amine, even at extended reaction times. Structures assigned to each new compound were supported by pmr studies.[1]

(1) Part XXIV: G. R. Pettit, D. C. Fessler, and J. A. Settepani, *J. Org. Chem.*, in press.

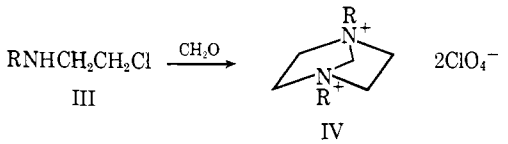
(2) To whom inquiries should be addressed at Arizona State University.

(3) (a) G. R. Pettit and J. A. Settepani, *J. Med. Pharm. Chem.*, **5**, 296 (1962); (b) W. Werner and M. Mühlstädt, *Justus Liebigs Ann. Chem.*, **693**, 197 (1966); (c) G. R. Pettit and B. J. Danley, *Can. J. Chem.*, **46**, 792 (1968); (d) G. R. Pettit and J. A. Settepani, *J. Org. Chem.*, **27**, 1714 (1962); (e) R. C. Elderfield and A. C. Menta, *J. Med. Chem.*, **10**, 922 (1967); (f) G. R. Revankar and S. Siddappa, *Monatsh. Chem.*, **98**, 169 (1967); (g) W. Werner, *J. Prakt. Chem.*, **37**, 154 (1968).

(4) (a) G. R. Pettit and J. A. Settepani, *Chem. Ind.* (London), 1805 (1964); (b) H. Böhme and H. Orth, *Arch. Pharm.*, **300**, 148 (1967).

(5) G. R. Pettit, J. A. Settepani, and R. A. Hill, *Can. J. Chem.*, **43**, 1792 (1965).

TABLE I



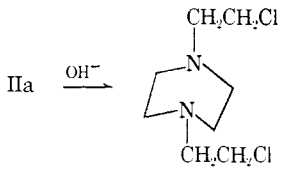
III IV

| Compd IV | R | Yield, % | Dec pt, °C | Formula[c] |
|---|---|---|---|---|
| a | $C_2H_5$ | 63 | 292–294 | $C_{11}H_{18}Cl_2N_2O_4$ |
| b |  | 81[a] | 271–273 | $C_{19}H_{26}Cl_2N_2O_4$ |
| c | $PhCH_2CH_2$ | 90[a] | 256–257.5 | $C_{21}H_{26}Cl_2N_2O_4$ |
| d | $3,4,5\text{-}(MeO)_3C_6H_2CH_2CH_2$ | 90[a] | 255–256.5 | $C_{27}H_{40}Cl_2N_2O_{10}$ |
| e | $PhCH_2$ | 61[a] | 191–193[b] | $C_{19}H_{24}Cl_2N_2O_4$ |
| f | $m\text{-}ClC_6H_4CH_2$ | 63[a] | 233–235 | $C_{19}H_{22}Cl_4N_2O_4$ |
| g | $3,5\text{-}(MeO)_2C_6H_3CH_2$ | 87[a] | 188–189 | $C_{23}H_{32}Cl_2N_2O_{12}$ |
| h | $3,4,5\text{-}(MeO)_3C_6H_2CH_2$ | 79[a] | 178–180 | $C_{25}H_{36}Cl_2N_2O_{14}$ |

[a] Calculated as the chloride salt. [b] Lit.[4b] 192°. [c] All compounds were analyzed for C, H, Cl, N. Analytical results were within 0.3% of the calculated values.

Biological evaluations are being performed under auspices of the Cancer Chemotherapy National Service Center. Results available at present indicate that chloroethyl derivative IIa and bromoethyl derivative IIb are the most active of the series, completely inhibiting growth of Walker 256 (subcutaneous) carcinoma in random-bred albino rats at dose levels of 23 and 50 mg/kg, respectively.[6] By comparison, the ethyl derivative IVa and quaternary salts IVf–h (investigated as chlorides) were considerably less active, demonstrating only slight activity at relatively high dose levels.

The greater activity displayed by haloethyl derivatives IIa,b might be explicable in terms of the diazabicyclo[2.2.1]heptane ring chemistry. Böhme and Orth have reported[4b] that basic hydrolysis of salt IIa produced formaldehyde and 1,4-bis(2-chloroethyl)piperazine (V), a potential alkylating agent. When evaluated



as described above, the maleate salt of piperazine V gave approximately 61% inhibition of tumor growth at a dose level of 150 mg/kg. Perhaps the activity of IIa may be due in part to *in vivo* formation of piperazine V.

### Experimental Section

Melting points were recorded employing a Kofler melting point apparatus. Purity of analytical samples (colorless) was confirmed by tlc on silica gel $HF_{254}$ (E. Merck, A. G. Darmstadt) spread on microscope slides. Chromatograms were performed with the top layer of a BuOH–$H_2O$–HOAc (4:5:1) mixture as solvent and developed with $I_2$. Microanalytical data were provided by Dr. A. Bernhardt, Max-Planck Institut, Mülheim, Germany. Ir spectra were determined in KBr by Miss K. Reimer (Arizona State University) and Dr. R. A. Hill (University of Maine). Pmr spectra were recorded in $D_2O$ (TMS external standard) with a Varian A-60. The secondary 2-haloethylamines were prepared employing a previously described procedure:[7] **N-(2-chloroethyl)-2-cyclopentylethylamine hydrochloride**, mp 245–246°, $\nu_{max}$ 2750 (broad, $NH_2^+$) $cm^{-1}$ [*Anal.* ($C_9H_{19}Cl_2N$) C, H, Cl, N]; **N-(2-chloroethyl)-2-phenylethylamine hydrochloride**, mp 193.5–194.5°, $\nu_{max}$ 2790 (broad, $NH_2^+$) and 1605 (weak) $cm^{-1}$, pmr $\delta$ 7.5 (s, 5 aromatic protons) and 4.1–3.0 (overlapping $A_2B_2$ patterns, 8 protons) [*Anal.* ($C_{10}H_{15}Cl_2N$) C, H, Cl].

The N-substituted 2-chloroethylamines were stored as their stable HCl salts and converted to the free bases as described for N-(2-chloroethyl)ethylamine.

**N-(2-Chloroethyl)ethylamine (IIIa).**—The HCl salt of IIIa (1.44 g) was added to an ice-cold 10% KOH solution (7 ml) and the resulting mixture was quickly extracted ($Et_2O$, three 10-ml portions). The combined $Et_2O$ extract was dried and concentrated at reduced pressure without heating to yield free base IIIa as a mobile oil.

**1,4-Diethyl-1,4-diazabicyclo[2.2.1]heptane Diperchlorate (IVa).**—A solution of N-(2-chloroethyl)ethylamine (0.01 mole), 37% formalin (2 ml), and 95% EtOH (3 ml) was stirred at room temperature 24 hr. Treatment of the colorless solution with 70% $HClO_4$ (0.8 ml) followed by chilling (ice bath) led to a crystalline solid (1.03 g, 63%) decomposing at 200–230°. Three recrystallizations from 95% EtOH afforded a pure sample as colorless plates: dec pt 292–294°.

**1,4-Bis(2-bromoethyl)-1,4-diazabicyclo[2.2.1]heptane Dibromide (IIb).**—Bis(2-bromoethyl)amine (0.008 mole) was left at room temperature in formalin–EtOH solution for 10 hr. After cooling, the salt which deposited was collected (2.3 g, 59%), dec pt 168–171°. A portion was recrystallized three times from aqueous EtOH for analysis, providing colorless plates: dec pt 178.5–179.5°. *Anal.* ($C_9H_{18}Br_4N_2 \cdot H_2O$) C, H, Br, N.

**1,4-Bis(2-fluoroethyl)-1,4-diazabicyclo[2.2.1]heptane Diperchlorate.**—To a solution of bis(2-fluoroethyl)amine hydrochloride (0.720 g, 0.005 ml)[8] in EtOH (10 ml) was added NaOH (0.2 g, 0.005 ml) in $H_2O$ (3 ml) followed by 37% formaldehyde (2 ml). After 6 days at room temperature, acidification with 70% $HClO_4$ (0.5 ml), dilution with EtOH (2 ml), chilling, and filtration afforded a colorless solid (0.55 g, 55%, dec pt 210–214). Two recrystallizations from aqueous EtOH followed by two from $H_2O$ produced an analytical sample, dec pt 223.5–225°. *Anal.* ($C_9H_{18}Cl_2N_2O_4$) C, H, Cl, F, N.

**1,4-Diazabicyclo[2.2.1]heptanes (Table I). General Procedure.**—A solution composed of N-[2-chloroethyl-2-(3,4,5-trimethoxyphenyl)]ethylamine (0.014 mole), 37% formalin (3.2 ml), and 95% EtOH (6.6 ml) was allowed to stand at room temperature 24 hr. The solution was concentrated at reduced pressure to an oil which was dried by addition and evaporation of PhH (three 50-ml portions). Upon trituration with dry $Et_2O$ the oil solidified and crystallized as needles (3.5 g, 90%) from EtOH–$Et_2O$; dec pt 215–220°. Treating a solution of the dichloride salt (0.5 g) in $H_2O$ (3 ml) with 70% $HClO_4$ (0.1 ml) yielded a colorless solid which crystallized from aqueous EtOH (0.47 g). Three recrystallizations from aqueous EtOH afforded a pure sample of IVd as colorless needles, dec pt 255–256.5°.

**Acknowledgment.**—This investigation was supported in part by Public Health service research grant CA-10115-02 from the National Cancer Institute, and in part by Grant T-79G from the American Cancer Society.

(6) In each case, the substance was given intraperitoneally in saline solution for 5 days following tumor transplant. Evaluation of tumor growth was made on the tenth day.

(7) G. R. Pettit, S. K. Gufta, and P. A. Whitehouse, *J. Med. Chem.*, **10**, 692 (1967).

(8) G. R. Pettit and R. L. Smith, *Can. J. Chem.*, **42**, 572 (1964).

## Fluorinated Pyrimidines. XXXII. Syntheses of 2′,3′-Dehydro-5-trifluoromethyl-2′-deoxyuridine and 5-Trifluoromethyluridine[1]

TASNEEM A. KHWAJA AND CHARLES HEIDELBERGER[2]

*McArdle Laboratory for Cancer Research, University of Wisconsin, Madison, Wisconsin 53706*

*Received February 3, 1969*

A number of fluorinated pyrimidines and their nucleoside derivatives have been synthesized in this labora-

(1) This work was supported in part by Grant CA-7175 from the National Cancer Institute, National Institutes of Health, U. S. Public Health Service.

(2) American Cancer Society Professor of Oncology.