[CONTRIBUTION FROM THE DEPARTMENT OF CHEMISTRY OF THE UNIVERSITY OF MICHIGAN]

Synthesis of Potential Anticancer Agents. III. Nitrogen Mustards Derived from 8-Aminoquinolines[1-3]

ROBERT C. ELDERFIELD AND ERNEST F. LEVON[4]

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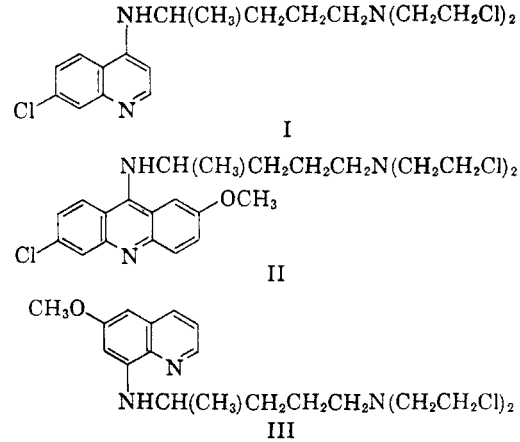
A series of 8-[3-bis(2-chloroethylamino)propionamido]quinolines has been prepared for evaluation as anticancer agents. 8-Amino-6-methoxyquinoline apparently cannot be reductively alkylated with acetals or aldehydes. Certain aspects of the synthesis of 8-(3-aminopropylamino)-2-methoxyquinoline have been clarified.

In recent years considerable interest has been displayed in the so-called antimalarial mustards as possible chemo-therapeutic agents in the management of neo-plastic disease. These substances may be broadly defined as embracing standard antimalarial drugs into the molecules of which an alkylating function such as the bis-2-chloroethylamino (nitrogen mustard) is incorporated. The rational underlying this approach was based on the hope that the antimalarial moiety of the molecule would act as a carrier to localize the action of the alkylating function.[5-7]

Fulton and co-workers[8] prepared the 2-anilino and 2-methyl derivatives of 4-[2-bis(2-chloroethylamino)ethyl]quinoline as candidate amebocides. The compounds showed no outstanding properties in this regard and apparently have not been examined as anticancer agents. Creech[6] has reported a derivative of 7-chloroquinoline carrying the same side-chain among several antimalarial mustards which showed appreciable activity against ascites tumors in mice. Creech also noted that antitumor activity in animals was retained in the 4-aminoquinoline mustards through various alterations in the side-chain and nuclear substitution. Jones[7,9] has reported the results of preliminary clinical trials of the nitrogen mustard analogs of chloroquine (I), quinacrine (II), and pamaquine (III), which although quite favorable were not conclusive.



In their syntheses Jones, Price, and Sen[7] were unable to obtain salts which analyzed for sufficient chlorine. Their 4-aminoquinoline and 9-aminoacridine mustards were prepared by a fundamentally different series of reactions and also gave somewhat unsatisfactory analytical data. Thus one would expect to encounter difficulty in the preparation and handling of compounds of this type.

Neeman[10] has prepared several amide analogs of the 8-amino-6-methoxyquinoline (IV) antimalarials carrying the carbonyl group in the side-chain, either at the 8-nitrogen or at the terminal end. Snyder and Freier[11] prepared several 3-dialkylaminopropionyl derivatives of IV. These as well as other acylated antimalarials had essentially no antimalarial activity.[12] Buchi[13] and Gaind and coworkers[14] have reported dialkylaminopropionyl derivatives of the other aminoquinolines as potential local anesthetics and several workers have described α-aminoacetamidoquinolines as spasmolytics or local anesthetics. The interesting point in

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(2) For paper II in this series see *J. Org. Chem.*, **24**, 1410 (1959).

(3) This work is based on a dissertation submitted by Ernest F. LeVon in partial fulfillment of the requirements for the degree of Doctor of Philosophy in the University of Michigan.

(4) Dow Chemical Company Fellow, 1955-1957. U.S. P.H.S. Predoctoral Fellow, 1958.

(5) R. Jones, Jr., C. C. Price, and A. K. Sen, *J. Org. Chem.*, **22**, 783 (1957).

(6) H. J. Creech, *Ann. N. Y. Acad. Sci.*, **68**, 868 (1958).

(7) R. Jones, Jr., U. Jonsson, M. Browning, H. Lessner, C. C. Price, and A. K. Sen, *Ann. N. Y. Acad. Sci.*, **68**, 1133 (1958).

(8) J. D. Fulton, L. P. Joyner, H. King, J. M. Osbond, and J. Wright, *Proc. Roy. Soc. (London)*, B, **137**, 339 (1950).

(9) R. Jones, Jr., H. J. Creech, C. C. Price, A. K. Sen, R. M. Peck, R. F. Nankwitz, Jr., R. Rhines, D. McKenzie, and W. F. Dunning, *Proc. Am. Assoc. Cancer Research*, **2**, 132 (1956).

(10) M. Neeman, *J. Chem. Soc.*, 2525 (1955).

(11) H. R. Snyder and H. E. Freier, *J. Am. Chem. Soc.*, **68**, 2485 (1946).

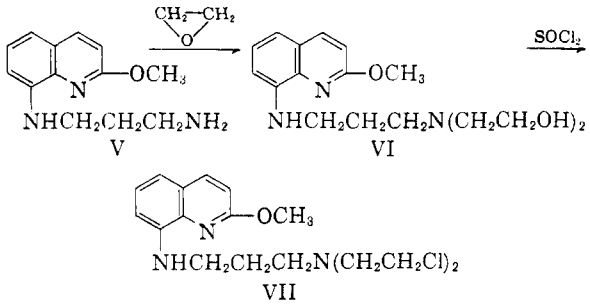
(12) *A Survey of Anti-malarial Drugs*, **1941-1945**, F. Y. Wiselogle (Editor), J. W. Edwards, Inc., Ann Arbor, Mich., 1946.

(13) J. Buchi, R. Lieberherr, and L. Ragaz, *Helv. Chim. Acta*, **34**, 1380 (1951).

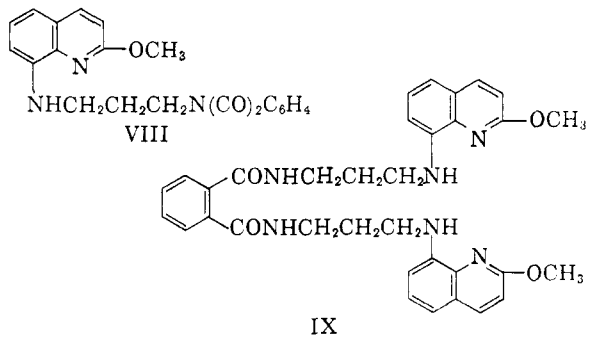
(14) K. N. Gaind, J. N. Ray, and B. Sarin, *J. Indian Chem. Soc.*, **17**, 619 (1941).

this connection is that the introduction of the amide function resulted in sharply reduced toxicity.[11] It therefore was desirable to prepare representative acylamido derivatives of 8-aminoquinolines which also carried the nitrogen mustard function. Interest in these compounds is two-fold. It was hoped that anticancer activity would be retained with lower toxicity and, secondly, that the compounds, or their precursors, might provide a better route to compounds of the type of III than that used by Jones and co-workers.[5] The first of these goals has apparently been achieved, as preliminary data with experimental animal tumors have shown a high order of activity coupled with relatively low toxicity for many of the substances.[15] Results of the approach to the second objective will be reported subsequently.

At the outset, attention was directed toward possible modifications of the Jones, Price, and Sen procedure[5] (V–VII) or alternate procedures for the preparation of anlogs of III. Initial effort was devoted to the preparation of 8-[3-bis(2-chloroethylamino)propylamino]-2-methoxyquinoline (VII) on the basis of preliminary pharmacological data on 8-(3-aminopropylamino)-2-methoxyquinoline and 8-(3-diethylaminopropylamino)-2-methoxyquinoline[16] which indicated that VII might be the compound of choice in this series.



A supply of crude 2-methoxy-8-(3-phthalimidopropylamino)quinoline (VIII) was available from other work. Fractional crystallization of this resulted in the isolation of a small amount of 8-bis(3-phthalimidopropylamino)-2-methoxyquinoline. Hydrazinolysis of VIII was accomplished by the method of Ing and Manske[17] and V was characterized as its dibenzenesulfonyl derivative. As a side-product in the hydrazinolysis a small amount of what is believed to be phthalobis[3-(2-methoxy-8-quinolylamino)propyl]amide (IX) analogous to the 6-methoxy derivative reported by Barber and Wragg[18] from a similar reaction. IX showed carbonyl absorption in the infrared at 1665 cm.$^{-1}$ similar to that of phthalamide and phthalhydrazide and a strong band at about 1522 cm.$^{-1}$ which probably corresponds to the band at 1538 cm.$^{-1}$ in VIII. Conversion of IX to a benzenesulfonyl derivative eliminated the band at 1522 cm.$^{-1}$ and regenerated carbonyl bands at 1705 and 1768 cm.$^{-1}$ typical of phthalimides. Barber and Wragg[18] noted that the 6-methoxy analog of IX regenerated the phthalimide corresponding to VIII by elimination of a molecule of primary amine. In view



of the difficulty encountered in handling V and its derivatives attention was shifted to 8-amino-6-methoxyquinoline (IV).

Mild conditions of hydroxyethylation of primaquine[6-methoxy-8-(4-amino-1-methylbutylamino)-quinoline] gave no isolatable products and in one instance resulted in no reaction. The forcing conditions used by Jones[5] were not employed because of the danger of alkylating the secondary 8-amino group as well as the terminal primary amine. Reductive alkylation offered an alternative means of introducing the side-chain as a preformed moiety. This has been accomplished by various workers from 8-aminoquinolines and ketals[19–22] or enol ethers derived from ketals.[23] 1-Bis(2-hydroxyethylamino)-4-pentanone[6] failed to yield the ketal under conditions which are successful with 1-diethylamino-4-pentanone. When 3-chloropropionaldehyde diethyl acetal was allowed to react with diethanolamine, the expected 3-diethanolaminopropionaldehyde acetal was obtained in good yield. However, attempted condensation of this acetal and IV failed. The mixtures obtained when the reaction was carried out in the presence of various acidic catalysts did not absorb catalytically activated hydrogen under conditions which are successful with ketals and infrared spectra, indicated only the recovery of starting materials. When the reaction of IV with butyraldehyde was

(15) Private communication from Dr. Ralph Jones, Jr., Jackson Memorial Hospital, University of Miami, Miami, Florida.

(16) H. B. Hughes and L. H. Schmidt, *Proc. Soc. Exp. Biol. Med.*, **73**, 581 (1950).

(17) H. R. Ing and R. H. F. Manske, *J. Chem. Soc.*, 2348 (1926).

(18) H. J. Barber and W. R. Wragg, *J. Chem. Soc.*, 1331 (1947).

(19) R. C. Elderfield, W. R. Vaughan, B. B. Millward, and J. H. Ross, *J. Org. Chem.*, **23**, 1378 (1958).

(20) H. J. Barber, D. H. O. John and W. R. Wragg, *J. Am. Chem. Soc.*, **70**, 2282 (1948).

(21) S. Tatsuoka, J. Ueyanagi, and T. Kinoshita, *J. Pharm. Soc. Japan*, **69**, 33 (1949).

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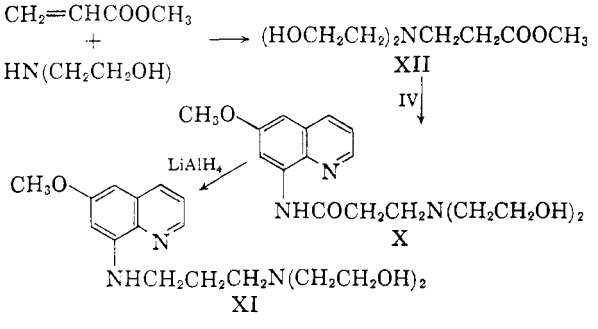
(23) D. Shiho, *J. Chem. Soc. Japan*, **65**, 135 (1944).

attempted, it occurred readily even in the absence of acidic catalysts. However, the product did not undergo catalytic reduction (as judged by hydrogen absorption) and no pure products could be isolated. We therefore conclude that preparation of aminoalkyl derivatives of IV cannot readily be accomplished by reductive alkylation with acetals or aldehydes.

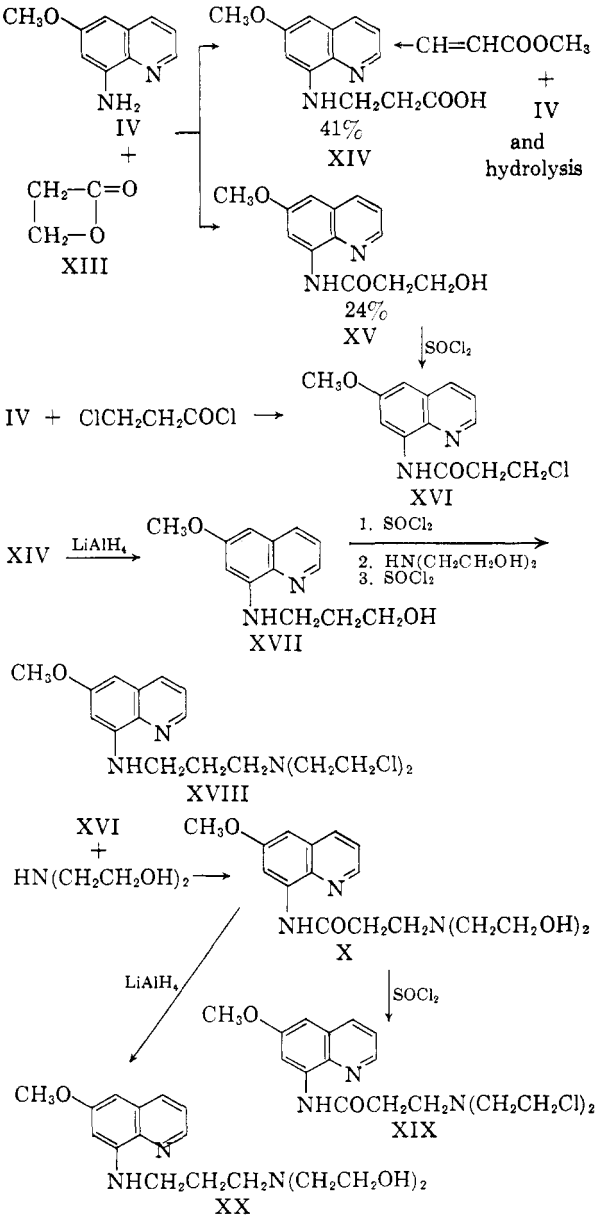
The Leuckart method for reductive alkylation with formic acid as the reducing agent was also investigated with butyraldehyde and IV. The only pure product isolated was the previously known formyl derivative of IV. Tomita[24] has reported that only low yields of pamaquine result from a modification of the Leuckart reaction under rather severe conditions.

Elderfield and co-workers[25] have suggested that alkyl derivatives of IV might be obtained by acylation of IV followed by reduction of the amide with lithium aluminum hydride. However, this approach was carried only through the acylation step and the proposed reduction was never carried out. It was hoped that aminolysis of a suitably substituted ester by IV would give a mustard diol carrying an amide function such as X which could be reduced to XI with lithium aluminum hydride. Addition of diethanolamine to methyl acrylate appeared to form methyl 3-bis(2-hydroxyethylamino)propionate (XII) very smoothly. Although XII dissociated on attempted distillation at 0.4–0.8 mm., it might have been useful as a crude product. Similar instability has also been reported for the adduct of diethanolamine and acrylonitrile.[26] Hydrolysis of crude XII gave 3-bis(2-hydroxyethylamino)-propionic acid which was also prepared from diethanolamine and propiolactone.[27] Because of the



instability of XII, exploratory attempts to acylate IV with ethyl butyrate were made. Several attempts under a variety of conditions gave no recognizable sign of the formation of the butyryl derivatives of IV. We therefore conclude that ester aminolysis with IV does not proceed readily.

Propiolactone (XIII) reacts with aromatic amines to give β-alanines and hydracrylamides.[27–29] The ratio of the products apparently varies with the amine and the conditions of the reaction. Reaction of IV with XIII in boiling benzene gave a mixture from which was isolated 41% of *N*-(6-methoxy-8-quinolyl)-β-alanine (XIV) and 24% of 6-methoxy-8-hydracrylamidoquinoline (XV). The reaction was very slow in boiling ether. The β-alanine (XIV) was identical with the product obtained by hydrolysis of the crude adduct of IV and methyl acrylate. Chlorination of XV with



(24) M. Tomita, S. Uyeo, H. Otaya, H. Maekawa, M. Fukuda, S. Echigo, S. Mizukami, and T. Matsui, *J. Pharm. Soc. Japan*, **71**, 829 (1951).

(25) R. C. Elderfield *et al.*, *J. Am. Chem. Soc.*, **77**, 4819 (1955).

(26) H. A. Bruson, *Org. Reactions*, 80 (1949).

(27) T. L. Gresham, J. E. Jansen, F. W. Shaver, R. A. Bankert, and F. T. Fiedorek, *J. Am. Chem. Soc.*, **73**, 3168 (1951).

(28) T. L. Gresham and F. W. Shaver, U. S. pat. 2,568,621 (1951).

(29) C. D. Hurd and S. Hayao, *J. Am. Chem. Soc.*, **74**, 5889 (1952); **76**, 5562 (1954).

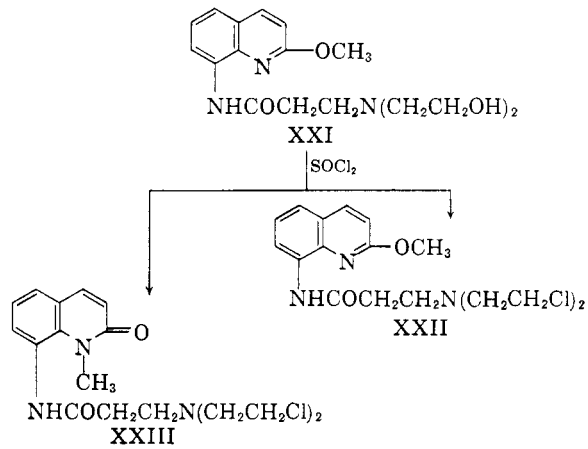
thionyl chloride gave 8-(3-chloropropionylamido)-quinoline which was identical with the product of the reaction of IV with 3-chloropropionyl chloride. When the reaction of IV with XIII was carried out in hot toluene or xylene substantial amounts of polymeric material resulted. However, XIV and XV were formed but in lower yields than was the case when the reaction was carried out in benzene.

The β-alanine (XIV) was reduced by lithium aluminum hydride in ether to give 8-(3-hydroxypropylamino)-6-methoxyquinoline (XVII) identical to the substance previously prepared by an alternate route.[30,31] This may possibly serve as an intermediate in the synthesis of the mustard derived from plasmocid (XVIII).

Reaction of XVI with two moles of diethanolamine gave up to 95% of the dihydrochloride of XIX which was in turn converted by thionyl chloride into 8-[3-bis(2-chloroethylamino)propionylamido]-6-methoxyquinoline (XX) an amide mustard analog of XVIII. Reduction of XIX with lithium aluminum hydride in tetrahydrofuran gave the mustard diol (XXI), which should be the ideal precursor to the plasmocid mustard (XVIII). Isolation and purification of XX as the dihydrochloride was easily accomplished in yields as high as 80% based on the amount of IV taken.

Contrary to the smooth reaction which we have noted between XVI and diethanolamine, Bergmann and Schapiro[32] report that reaction of XVI with diethylamine results in dehydrochlorination and that the crude product gives 8-(acrylamido)-6-methoxyquinoline on distillation. On the other hand, Gaind, Ray, and Sarin[15] found that 8-(3-chloropropionylamido)quinoline reacted with diethylamine without dehydrochlorination in the presence of sodium carbonate. They did not, however, distill the product, but isolated it as the picrate. Snyder and Freier[12] obtained Bergmann's acrylamide from IV and acrylolyl chloride and succeeded in adding diethylamine smoothly to the vinyl linkage. Again, the product was not distilled but isolated as the salt. Thus it is probable that Bergmann and Schapiro actually had 8-(3-diethylaminopropionylamido)-6-methoxyquinoline in hand but that this lost diethylamine on distillation.

In view of the encouraging results of preliminary screening data for XX against animal tumors,[15] the series was extended to include derivatives of 5,6-dimethoxyquinoline, 6-methoxylepidine, and 2-methoxyquinoline analogous to XVI, XIX, and XX. These all proceeded smoothly with the exception of the conversion of the mustard diol (XXI) derived from 2-methoxquinoline to the mustard (XXII). When XXI was treated with thionyl chloride in chloroform, two main products were isolated as the hydrochlorides. Recrystallization of the higher melting fraction gave a product which melted sharply at about 200° and furnished analytical data in substantial agreement with those



demanded by XXII. However, the infrared spectrum showed two carbonyl bands at 1705 and 1670 cm.$^{-1}$ and methoxyl analyses indicated the absence of the 2-methoxyl function. Sheinker and Pomerantsev[33] report infrared absorption in the carbonyl region at 1650 and 1655 cm.$^{-1}$ for carbostyril and 1-methyl-2-quinolone respectively. 1-Methyl-8-nitro-2-quinolone absorbs at about 1665 cm.$^{-1}$. Therefore the mustard melting at 200° is probably the isomeric 1-methyl-2-quinolone (XXIII). The first carbonyl absorption (1705 cm.$^{-1}$) is probably due to the side chain amide and the other (1670 cm.$^{-1}$) to the 2-quinolone. Other amides prepared in the course of this work showed carbonyl absorptions from 1660 to 1700 cm.$^{-1}$ However, none having the 1-methyl-2-quinolone structure were available. Recrystallization of the second hydrochloride gave the salt of XXII. This showed the presence of the methoxyl group on analysis and showed only one carbonyl band in the infrared at 1680 cm.$^{-1}$. On the other hand reaction of the hydrochloride of XXI with thionyl chloride gave XXII smoothly.

Details as to the physiological properties of these compounds will be reported elsewhere.

EXPERIMENTAL[34-36]

2-Methoxy-8-(3-phthalimodopropylamino)quinoline (VIII) *and 2-methoxy-8-bis(3-phthalimidopropylamino)quinoline.* The

(30) J. Crum and R. Robinson, *J. Chem. Soc.*, 561 (1943).

(31) W. H. Yanko, H. S. Mosher, and F. C. Whitmore, *J. Am. Chem. Soc.*, **67**, 664 (1945).

(32) F. Bergmann and D. Schapiro, *J. Org. Chem.*, **7**, 419 (1942).

(33) Y. N. Sheinker and Y. I. Pomerantsev, *Zhur. Fiz. Khim.*, **30**, 79 (1956).

(34) All melting points and boiling points are uncorrected unless noted otherwise.

(35) Infrared spectra were taken as nujol mulls on a Perkin-Elmer double beam recording spectrophotometer, Model 21.

(36) Microanalyses by Spang Microanalytical Laboratory, Ann Arbor, Michigan, Mrs. Anna Griffin of the University of Michigan, Drs. Weiler and Strauss, Oxford, England, or Schwarzkopf Microanalytical Laboratory, Woodside, N. Y.

crude phthalimide[37] prepared from 8-amino-2-methoxyquinoline[38] and 3-phthalimidopropyl bromide according to the general procedure of Baldwin,[39] was separated by repeated recrystallization from benzene or acetone-alcohol into pure VIII, m.p. 113–114.5° and a small amount of the bisphthalimidopropylamino compound, m.p. 137–138°. Melting point previously reported for VIII is 112–114.5°.[37] The disubstituted material was analyzed.

Anal. Calcd. for $C_{32}H_{28}N_4O_5$: C, 70.06; H, 5.14. Found: C, 70.13; H, 4.96.

The infrared spectra were complementary with respect to the absence of a strong band at 1538 cm.$^{-1}$ in the spectrum of the 2-methoxy-8-bis(3-phthalimidopropylamino)-quinoline which was present in the spectrum of VIII. This band was probably due to the 8-nitrogen-hydrogen bond. The carbonyl bands were at 1715 aud 1765 cm.$^{-1}$ in the monosubstituted compound and at 1705 and 1760 cm.$^{-1}$ in the other. VIII also showed an N—H band at about 3380 cm.$^{-1}$

Hydrazinolysis of VIII. 8-(3-Aminopropylamino)-2-methoxyquinoline has been the subject of pharmacological study[16] but its chemistry has not been recorded.[37] Pure VIII (25.0 g.) was suspended in 250 ml. of ethanol, hydrazine hydrate (3.8 g.) was added, and the mixture was refluxed gently for 3.5 hr. After cooling in the refrigerator, the nearly solid cake was filtered, washed, and dried to give 24.2 g. (89%) of the intermediate phthalhydrazide salt, m.p. 185–195°, of 8-(3-aminopropylamino)-2-methoxyquinoline; reported[37] m.p. 178.5–181°. In another run the intermediate was recrystallized from a large volume of alcohol without appreciably altering its melting point. The recrystallized material was suspended in water, made basic with *N* sodium hydroxide, and extracted with ether. The ethereal extract was dried over anhydrous magnesium sulfate and acidified by dropwise addition of an alcoholic solution of hydrogen chloride. The hydrochloride of V (11.6 g.) melted at 156–159° dec., solidified and remelted at 245–251°. This salt has been reported as melting at 240–245° after sintering at 140–144°.[37]

In another run, the reaction mixture was acidified while still hot with 6*N* hydrochloric acid and the phthalhydrazide was filtered. However, when the hydrochloride of V was allowed to crystallize directly from the filtrate, it still contained up to 11% of phthalhydrazide. In view of this and the susceptibility of the 2-methoxyl group to acid, this method of working up the reaction mixture was abandoned.

When 250 ml. of absolute alcohol was substituted for 95% alcohol in the above hydrazinolysis, some difficulty with bumping caused by separated solid was encountered. After refluxing for 3 hr. most of the alcohol was distilled, the residue was mixed with excess sodium hydroxide solution and the free amine (V) was extracted with ether. After washing with water, the combined ethereal extracts were dried over anhydrous potassium carbonate. Acidification of the basic aqueous solution from the ether extraction gave 9.2 g. (82%) of phthalhydrazide, m.p. 343–344° dec. During the drying of the ether extracts, a colorless precipitate separated and was filtered off with the potassium carbonate. Acidification of the ether filtrate with 2*N* alcoholic hydrogen chloride (prepared from concd. aqueous hydrochloric acid) gave 15.6 g. of the hydrochloride of V, m.p. 159–160° dec., solidifying and remelting at 248–254°. The carbonate residue from the ether filtrate was washed with water leaving 2.1 g. of insoluble base, m.p. 147–150°. After six recrystallizations from ethanol it melted at 148–152°.

Anal. Calcd. for $C_{34}H_{36}N_6O_4$: C, 68.90; H, 6.12; N, 14.18. Found: C, 68.64; H, 5.92; N, 14.02.

A hydrochloride which decomposed before it could be analyzed darkened at about 174° and melted at 176–177.5° dec. On the basis of comparative infrared data[40] and by analogy with the corresponding derivative of 8-amino-6-methoxyquinoline[18] this compound is assigned the structure of phthalobis[3-(2-methoxy-8-quinolylamino)propyl]amide (IX).

The hydrochlorides of 8-(3-aminopropylamino)-2-methoxyquinoline. The double-melting hydrochloride continued to decompose at about 160° and remelt at about 250° dec. after recrystallization from alcohol even in the presence of excess acid. The lower melting point was eliminated on heating at 95° for 48 hr. during which a strong carbonyl band appeared in the infrared at 1660 cm.$^{-1}$ Titration with silver nitrate indicated an equivalent weight of about 190 (18.7, 18.8% Cl) which corresponds to 1.5 equivalents of hydrogen chloride per molecule of quinoline. It has been frequently observed that derivatives of 8-aminoquinoline do not react with stoichiometric amounts of acid.[41] We interpret this behavior as indicating that the original 2-methoxyquinoline derivative (V) rearranges to the *N*-methylquinolone under the influence of heat.

The unrearranged hydrochloride was also titrated against silver nitrate after only brief heating.

Anal. Calcd. for $C_{13}H_{17}NO_3 \cdot HCl$: Cl, 13.24; for $C_{13}H_{17}NO_3 \cdot 1.5\,HCl$: Cl, 18.60; for $C_{13}H_{17}NO_3 \cdot 2HCl$: Cl, 23.30; for $C_{13}H_{17}N_3O \cdot 2HCl \cdot 2H_2O$: Cl, 20.84; for $C_{13}H_{17}NO_3 \cdot 2HCl \cdot C_2H_5OH$: Cl, 20.24. Found: 20.5.

A monohydrochloride was obtained as a hygroscopic tan powder, m.p. 188–194°, when the free base was acidified with the calculated amount of alcoholic hydrogen chloride (based on the original hydrochloride as $C_{13}H_{17}N_3O \cdot 2HCl \cdot 2H_2O$).

Anal. Found: Cl, 13.0.

The monophosphate of 8-(3-aminopropylamino)-2-methoxyquinoline. When an aliquot of the free base in ether was acidified with an equivalent of alcoholic phosphoric acid a monophosphate, m.p. 240–250° with sintering at about 180°, was obtained in 97% yield. Recrystallization from ethanol changed the melting point to 206–225°.

Anal. Calcd. for $C_{13}H_{17}N_3O \cdot H_3PO_4$: C, 47.42; H, 6.08; N, 9.42. Found: C, 48.09; H, 6.40; N, 9.56.

The hydrochloride of V was treated with benzenesulfonyl chloride in aqueous sodium hydroxide. After several recrystallizations from ethanol the bissulfonamide melted at 146–148°.

Anal. Calcd. for $C_{25}H_{25}N_3O_5S_2$: C, 58.69; H, 4.93; N, 8.22; S, 12.53. Found: C, 58.79; H, 5.02; N, 8.01; S, 12.25.

3-(2,2-Dihydroxyethylamino)propionaldehyde diethyl acetal. A solution of 12.5 g. of diethanolamine and 10 g. of 2-chloropropionaldehyde diethyl acetal in 25 ml. of absolute ethanol was refluxed for 21 hr. After removal of volatile material at the water pump, the residue was dissolved in 10 ml. of water and extracted with four 25-ml. portions of chloroform. After drying the combined chloroform extracts over anhydrous potassium carbonate, the residue after removal of the solvent was distilled under reduced pressure to give 53% of an oil, b.p. 130–133° (0.5 mm.), n_D^{25} 1.457. Titration with hydrochloric acid to a methyl red end point gave a neutral equivalent of 232, calcd. 235.3. No solid derivative could be prepared.

Anal. Calcd. for $C_{11}H_{23}NO_4$: C, 56.14; H, 10.71; N, 5.95. Found: C, 56.38; H, 10.87; N, 6.11.

Reaction of propiolactone with 8-amino-6-methoxyquinoline. A. *In ether.* To a solution of 12.1 g. of IV in 100 ml. of ether cooled to 5°, 5 g. of propiolactone was slowly added. There was no evidence of a spontaneous reaction and the mixture was allowed to stand overnight. Addition of a solution of 2 g. of sodium hydroxide in 30 ml. of water caused a moderately vigorous reaction (hydrolysis of unchanged lactone). The ether layer was separated, washed with water, and dried.

(37) R. C. Elderfield and H. E. Mertel, unpublished work.

(38) K. Mislow and J. B. Koepfli, *J. Am. Chem. Soc.*, **68**, 1553 (1946).

(39) A. W. Baldwin, *J. Chem. Soc.*, 2959 (1929).

(40) See ref. (3) for details.

(41) R. C. Elderfield, *et al.*, *J. Am. Chem. Soc.*, **68**, 1524 (1946).

Evaporation of the ether left 12 g. of fairly pure unchanged IV. Neutralization of the basic solution with 5 ml. of concd. hydrochloric acid gave 0.25 g. (1.5%) of *N*-(6-methoxy-8-quinolyl)-β-alanine (XIV), m.p. 142–144° after recrystallization from dilute ethanol.

Anal. Calcd. for $C_{13}H_{14}N_2O_3$: C, 63.40; H, 5.73; N, 11.38. Found: C, 63.33; H, 5.72; N, 11.35.

When the reaction mixture was refluxed for 45 hr., the yield of XIV was 15%.

B. *In benzene.* To a solution of 92 g. of IV in 600 ml. of warm benzene 39 g. of propiolactone was added. After refluxing for 70 hr. the cooled solution was extracted with an aqueous solution of 22 g. of sodium hydroxide. The aqueous solution was extracted with five 50-ml. portions of benzene. Concentration of the combined benzene solution and extracts left 45 g. of dark viscous oil. Crystallization from 100 ml. of benzene with charcoal gave 10.7 g. (8.2%) of 6-methoxy-8-hydracrylamidoquinoline (XV), m.p. 97.5–98.5°. The mother liquors contained further amounts of XV along with unchanged IV.

Anal. Calcd. for $C_{13}H_{14}N_2O_3$: C, 63.40; H, 5.73; N, 11.38. Found: C, 63.39; H, 5.74; N, 11.51.

The hydrochloride of XV melted at 179–184° dec. after recrystallization from absolute ethanol.

Anal. Calcd. for $C_{13}H_{14}N_2O_3 \cdot HCl$: N, 9.91. Found: N, 9.86.

When the reaction was run in refluxing xylene, it darkened rapidly in 5–10 min. Therefore it was allowed to stand overnight at room temperature during which a dark precipitate appeared. Addition of aqueous sodium hydroxide dissolved the precipitate and the mixture was continuously extracted with benzene for 24 hr. From the extracts 6% of XV was obtained. Careful acidification of the aqueous layer gave 34% of XIV along with a lump of black tar which was picked out manually.

When the reaction was run in toluene by gradually raising the temperature to 105° over 90 min., 13% of XV and 30% of XIV were obtained.

8-(3-Hydroxypropylamino)-6-methoxyquinoline (XVII). Absolute ether (200 ml.) containing a few mg. of lithium aluminum hydride was placed in a flask connected to a Soxhlet extractor and a condenser protected by a calcium chloride tube. After standing for a few minutes to insure dryness, 2.0 g. of hydride was added to the ether and 5 g. of XIV was placed in the thimble which was then filled with sand. Additional ether was added from time to time to maintain the volume. After a few days all the XIV had been extracted and the Soxhlet was replaced by a reflux condenser protected by a calcium chloride tube. After refluxing for 13 days, excess hydride was decomposed by successive addition of ethyl acetate and water. After filtering, the ether layer was separated, washed with water until neutral, and dried over anhydrous sodium carbonate. Cautious acidification of the aqueous solution gave no unchanged XIV. To the dried ether solution 50 ml. of absolute ethanol was added followed by 25 ml. of 1.5*N* alcoholic hydrogen chloride. The hydrochloride of XVII crystallized as clumps of orange needles, m.p. 170–177° dec. in 60% yield. After recrystallization from absolute ethanol and dry ether containing a little hydrogen chloride it melted at 178–180° dec. with preliminary sintering. Reported melting point for material prepared by another method is 178°[30] and 171–172°.[31]

Anal. Calcd. for $C_{13}H_{16}N_2O_2 \cdot HCl$: C, 58.10; H, 6.38; Cl, 13.19. Found: C, 58.08; H, 6.24; Cl, 13.00.

6-Methoxy-8-(3-chloropropionamido)quinoline (XVI). A. *From 6-methoxy-8-hydracrylamidoquinoline.* To a solution of 5.0 g. of XV in 50 ml. of chloroform dried over calcium chloride in a flask equipped with a dropping funnel, stirrer, and condenser, a solution of 3 ml. of pure thionyl chloride in 50 ml. of dry chloroform was added with stirring during 30 min. while the temperature was held at 10°. After standing 15 min. at room temperature the mixture was refluxed for 1 hr. After cooling 13 g. of anhydrous sodium carbonate was added and, after stirring for a few minutes, water was added cautiously. The chloroform layer was separated, filtered, and concentrated to give 5.7 g. of residue. Recrystallization from 2-propanol gave 86% of colorless needles, m.p. 104–105°; reported[32] m.p. 104°.

Anal. Calcd. for $C_{13}H_{13}ClN_2O_2$: C, 58.98; H, 4.95; N, 10.58; Cl, 13.40. Found: C, 59.01; H, 4.93; N, 10.62; Cl, 13.02.

B. *From 8-amino-6-methoxyquinoline.* To a cooled solution of 10.0 g. of IV in 50 ml. of chloroform, previously dried over calcium chloride, and 5 ml. of pyridine 6 ml. of 3-chloropropionyl chloride was added gradually during 20 min. during which the temperature was maintained at 5–15°. After allowing the mixture to come to room temperature, a cold solution of 15 g. of potassium carbonate in 50 ml. of water was added with good stirring and cooling. The chloroform layer was separated and the aqueous layer was extracted with fresh chloroform. Concentration of the washed and dried chloroform solution gave 12.7 g. (84%) of XVI, m.p. 104.5–105.5° after recrystallization from 2-propanol.

8-[3-Bis(2-hydroxyethylamino)propionamido]-6-methoxyquinoline (X). To a solution of 8.0 g. of redistilled diethanolamine in 100 ml. of absolute ethanol was added 10.0 g. of XVI and the mixture was refluxed for 48 hr. After removal of the alcohol under reduced pressure, the residue was taken up in chloroform and the solution was washed with water until the wash water was neutral. After drying over anhydrous sodium carbonate, removal of the solvent left 16 g. of pale yellow viscous material which was perfectly satisfactory for the preparation of the mustard, XIX. For characterization the crude product was converted to the hydrochloride by solution in 150 ml. of absolute ethanol at 65° and acidification with 80 ml. of 1.5 *N* alcoholic hydrogen chloride. A copious precipitate (95%) of fine yellow needles formed which was collected and washed with absolute ethanol. It was recrystallized from methanol-ether. Physical constants and analytical data for this and related compounds are given in Table I.

The picrate, prepared in benzene and recrystallized from methanol, melted at 169–171°.

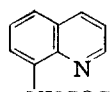
Anal. Calcd. for $C_{23}H_{26}N_6O_{11}$: C, 49.11; H, 4.66; N, 14.94. Found: C, 49.07; H, 4.54; N, 14.99.

8-[3-Bis(2-chloroethylamino)propionamido]-6-methoxyquinoline (XIX). Crude X prepared from 40 g. of XVI was cooled in an ice bath and 30 ml. of ice-cold purified thionyl chloride was added all at once. The flask was closed with a calcium chloride tube and kept in the ice bath for an hour with occasional swirling during which most of the diol dissolved. The mixture was allowed to come to room temperature overnight and concentrated under reduced pressure. The residue was warmed in 130 ml. of absolute ethanol and 20 ml. of 1.5*N* alcoholic hydrogen chloride was added during which the hydrochloride of XIX crystallized. After standing overnight the bright yellow granular crystals were collected and recrystallized by solution in 100 ml. of methanol and 20 ml. of 1.5*N* alcoholic hydrogen chloride, filtration and dilution with 100 ml. of absolute ether. The melting point varied somewhat with the rate of heating.

8-[3-Bis(2-chloroethylamino)propionamido]-5,6-dimethoxyquinoline. The chloroamide and diol were prepared from 5,6-dimethoxy-8-aminoquinoline[19] as in the preceding instance. Dry 8-[3-bis(2-hydroxyethylamino)propionamido]-5,6-dimethoxyquinoline hydrochloride (8.0 g.) was moistened with chloroform previously dried over calcium chloride and chilled in an ice-salt bath in a flask protected with a calcium chloride tube. To the mixture 16 ml. of purified thionyl chloride was added in one portion with swirling and cooling. The hydrochloride dissolved to give an orange solution in 5–10 min. The mixture was kept in the ice bath for 40 min. and then at room temperature for an hour after which the chloroform and excess thionyl chloride were removed under reduced pressure at 40°. The residue was crystallized from 80 ml. of absolute ethanol and 100 ml. of absolute ether. The very hygroscopic dihydrochloride was recrystallized from ethanol-ether in the presence of excess hydrogen chlo-

TABLE I

DERIVATIVES OF 8-AMINOQUINOLINE



NHCOCH$_2$CH$_2$R

R	Nuclear Substituent(s)	M.P. °	Yield, %	Analyses							
				Calcd.				Found			
				C	H	N	Cl	C	H	N	Cl
Cl	6-OCH$_3$	104.5–105.5	84	58.98	4.95	10.58	13.40	59.01	4.93	10.62	13.02
N(CH$_2$CH$_2$OH)$_2$[a]	6-OCH$_3$	175–178 (dec.)	95	50.25	6.20	10.34	17.45	50.24	6.28	10.14	17.31
N(CH$_2$CH$_2$Cl)$_2$[a]	6-OCH$_3$	184–187 (dec.)	80	46.07	5.23	9.48	32.00	46.16	5.40	9.58	31.84
Cl	5-OCH$_3$, 6-OCH$_3$	119–120	86	57.05	5.13	9.51	12.03	57.02	5.32	9.48	12.11
N(CH$_2$CH$_2$OH)$_2$[b]	5-OCH$_3$, 6-OCH$_3$	143–144	83	48.65	4.76	14.18		48.68	4.97	14.20	
N(CH$_2$CH$_2$Cl)$_2$[a]	5-OCH$_3$, 6-OCH$_3$	173–177	82	45.68	5.32	8.88	29.97	45.45	5.61	8.72	29.81
Cl	4-CH$_3$, 6-OCH$_3$	157.5–159	73	60.32	5.42	10.05	12.72	60.60	5.71	9.97	12.93
N(CH$_2$CH$_2$OH)$_2$	4-CH$_3$, 6-OCH$_3$	127.5–128.5	79	62.23	7.25	12.10		62.31	7.24	12.32	
N(CH$_2$CH$_2$Cl)$_2$[a]	4-CH$_3$, 6-OCH$_3$	176–178; 207–220[c]	83	47.28	5.51	9.19	31.02	47.28	5.57	9.24	31.24
Cl	2-OCH$_3$	113–114	83	58.98	4.95	10.58	13.40	59.13	4.88	10.89	13.39
N(CH$_2$CH$_2$OH)$_2$[d]	2-OCH$_3$	184.5–186 (dec.)	78	55.20	6.54	11.36	9.58	55.40	6.59	11.10	9.54
N(CH$_2$CH$_2$Cl)$_2$[d]	2-OCH$_3$	163–163.5 (dec.)	54	50.20	5.45	10.33	26.15	50.13	5.46	10.38	26.27[e]

[a] Isolated and analyzed as the dihydrochloride. [b] Isolated and analyzed as the picrate. The hydrochloride was amorphous. [c] Double melting point. [d] Isolated and analyzed as the monohydrochloride. [e] Calcd. CH_3O, 7.63. Found: 7.67.

ride. It showed a double melting point in an evacuated capillary, decomposing at about 106–107°, resolidifying and melting again at 183–177° dec.

8-[3-Bis(2-chloroethylamino)propionamido]-6-methoxylepidine. The chloropropionamide was prepared as in the above cases from 8-amino-6-methoxylepidine.[42] A solution of 15 g. of the chloropropionamide and 11.3 g. of redistilled diethanol amine in 225 ml. of absolute ethanol was refluxed for 40 hr. and concentrated to dryness. The residue was dissolved in chloroform and washed free of diethanolamine hydrochloride with water. After drying over anhydrous carbonate, the residue was crystallized from 150 ml. of absolute ethanol to give the diol as large prisms. The mustard was prepared in chloroform solution as described above. The dihydrochloride was recrystallized from methanol-ether with excess hydrogen chloride and showed a double melting point, decomposing at 178°, resolidifying as large plates, and melting again at 207–220° dec. in an evacuated capillary.

8-[3-Bis(2-chloroethylamino)propionamido]-2-methoxyquinoline (XXII) *and 8-[3-bis(2-chloroethylamino)propionamido]-1-methyl-2-quinolone* (XXIII). The chloropropionamide was prepared from 8-amino-2-methoxyquinoline as described above. The diol (XXI) was obtained as the monohydrochloride from methanol-ether with a slight excess of hydrogen chloride.

Direct treatment of the crude free base (XXI) with thionyl chloride gave no characterizable product. A solution of 2.0 g. of the monohydrochloride of XXI in water was made alkaline with sodium hydroxide and extracted with alcohol free chloroform. After drying the extract over anhydrous potassium carbonate, a solution of 10 ml. of redistilled thionyl chloride in 20 ml. of alcohol-free chloroform was gradually added. The mixture was protected by a calcium chloride tube. An oily precipitate that did not crystallize formed. Excess thionyl chloride was decomposed by addition of 10 ml. of methanol during which the oil solidified. It was too hygroscopic to collect on a filter satisfactorily. The tacky mass was dried in a desiccator to give a hard hygroscopic mass. Recrystallization from 2-propanol with excess alcoholic hydrogen chloride gave 112 mg. of material, m.p. 200–201.5° dec. The major portion of the product was recovered by concentrating the original chloroform-methanol filtrate and recrystallizing the residue from methanol-ether. This gave two slightly pink crops of 0.5 g. (46%) each. The first sintered from 180° and melted at 182–183° and the second melted at 163–164° dec. Recrystallization of the first fraction from methanol-ether gave material, m.p. 199.5–201.5° dec., which showed carbonyl absorption in the infrared at 1670 and 1705 cm.$^{-1}$ As no methoxyl was present on analysis, this is the *N*-methyl-2-quinolone (XXIII). Analytical data eliminate the carbostyril from consideration.

Anal. Calcd. for $C_{17}H_{21}Cl_2N_3O\cdot HCl$: C, 50.20; H, 5.45; N, 10.33; Cl, 26.15. Calcd. for $C_{16}H_{19}Cl_2N_3O_2\cdot HCl$: C, 48.93; H, 5.13; N, 10.70; Cl, 27.09. Found: C, 50.24; H, 4.99; N, 10.45; Cl, 25.95.

The second crop of crystalline material showed only a single carbonyl band in the infrared at 1680 cm.$^{-1}$ Recrystallization from methanol-ether left the melting point substantially unchanged. This was the monohydrochloride of the desired mustard (XXII).

When the hydrochloride of XXI was treated with thionyl chloride in chloroform as in the above examples, the hydrochloride of XXII was obtained in 54% yield with none of the quinolone being isolated.

ANN ARBOR, MICH.

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[CONTRIBUTION FROM THE DEPARTMENT OF CHEMISTRY OF THE UNIVERSITY OF MICHIGAN]

Synthesis of Potential Anticancer Agents. IV. Synthesis of Certain Substituted Amino- and Aziridinopyrimidines[1,2]

ROBERT C. ELDERFIELD AND RAJ NANDAN PRASAD[3]

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Candidate cytotoxic agents have been prepared by condensation of 2,4-dichloro-6-methyl-5-nitropyrimidine with various cyclic amines. The relative activating influence of a nitro, chloro, and bromo substituent in the 5-position of 2,4-dichloro-6-methylpyrimidine toward nucleophilic displacement of the chlorines has been studied.

Since 1946, when Gilman and Philips[4] reported the cytotoxic activity of bis-β,β'-dichloroethylmethylamine (nitrogen mustard), a number of cytotoxic substances containing the bis-β,β'-dichloroethylamine functions have been reported.[5] Most of these, except for some derivatives of amino acids[6–8] and sugars[9] are derivatives of parent compounds which are not of natural occurrence. Curiously, attention to the biologically important purines and pyrimidines has been largely directed to the preparation of analogs of them as possible antimetabolites and, until comparatively recently, few reports of incorporation of alkylating functions such as the bis-β,β'-dichloroethylamino or aziridino groups into these parent molecules have appeared. The rationale underlying the present work, a portion of which is presented,

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(2) For paper III in this series see *J. Org. Chem.*, 25, 1576 (1960).

(3) On leave of absence from the Chemistry Department, B. N. College, Patna University, India.

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