diluted with water. The acid was filtered off, washed with water and dried; yield 0.94 g. It was recrystallized from ethyl acetate; m.p. 123–126°.

Anal. Calcd. for $C_{17}H_{16}N_2O_6$: N, 8.14. Found: N, 7.98.

DL-*threo*-2-Acetamido-1-(4-nitro-1-naphthyl)-1,3-propanediol (V).—A mixture of 24.2 g. (0.08 mole) of α-acetamido-β-hydroxy-4-nitro-1-propionaphthone, 32.6 g. (0.16 mole) of aluminum isopropoxide and 250 ml. of anhydrous isopropyl alcohol was distilled slowly until the acetone test was negative. During the distillation a total of 180 ml. of distillate was collected. To the warm residue was added 120 ml. of isopropyl alcohol and 45 ml. of water. The mixture was refluxed for several minutes. Apparently ammonia was formed. The hot mixture was filtered through a layer of Super-cel. The filter-cake was extracted three times with hot 250-ml. portions of 80% isopropyl alcohol. The extracts were combined and concentrated thoroughly at 20 mm. and 80° (water-bath). The residue, a thick oil, partially solidified after standing several hours. It was mixed with 50 ml. of ethyl acetate, cooled and filtered to yield 2.1 g. of solid. Additional material was obtained by concentration of the ethyl acetate. The solids were combined and recrystallized from water.

DL-*threo*-2-Amino-1-(4-nitro-1-naphthyl)-1,3-propanediol (VI).—A mixture of 1.75 g. of the acetamido derivative and 80 ml. of 5% hydrochloric acid was heated on a steam-bath for 3 hours, then mixed with 2 g. of Darco and filtered. An excess of 20% aqueous sodium hydroxide was added to the cooled filtrate. The cold mixture was filtered and the solid washed with water and dried. The product was recrystallized from dilute methanol.

DL-*threo*-2-Dichloroacetamido-1-(4-nitro-1-naphthyl)-1,3-propanediol (VII).—A solution of 1.05 g. (0.004 mole) of the free base in 25 ml. of methyl dichloroacetate was warmed on the steam-bath for 25 minutes. The solution was concentrated at 20 mm. and 90° (water-bath). The solid residue was washed with chloroform and then recrystallized from water.

DETROIT, MICHIGAN RECEIVED SEPTEMBER 13, 1950

[CONTRIBUTION FROM THE DEPARTMENT OF CHEMISTRY, UNIVERSITY OF MARYLAND]

Quinolinequinones.[1] I. Quinones and Hydroquinones Related to Pentaquine[1a]

BY NATHAN L. DRAKE AND YOLANDA T. PRATT

Certain quinones and hydroquinones related to pentaquine (I) have been prepared by the reactions indicated in Fig. 1. The high *in vitro* antimalarial activities of compounds IIIb and Vb, in contrast to the inactivity of I, lend support to Schönhöfer's theory that the *in vivo* action of the 8-aminoquinolinesupon the erythrocytic forms of malaria plasmodia is due to the quinonoid products to which these drugs are converted by the host.

On the basis of certain correlations of chemical structure with activity against the erythrocytic forms of avian plasmodia, Schönhöfer postulated that the action of 6-methoxy-8-aminoquinolines (*e.g.*, I) is due to the quinonoid products (*e.g.*, IV) to which they are converted by the host organism.[2,3,4]

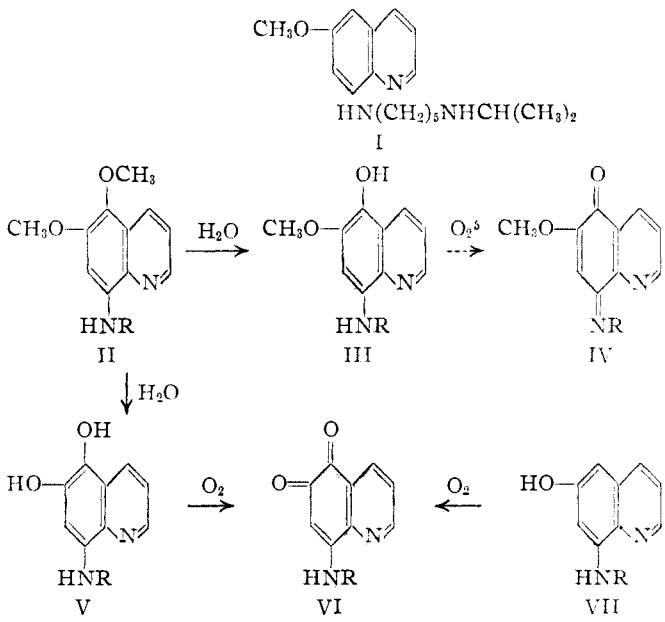


Fig. 1.—In the a series R = H; in the b series R = $-(CH_2)_5NHCH(CH_3)_2$.

It has also been suggested that the methemoglobinemia resulting from administration of these drugs might be caused by such quinonimines.[3] In view of these theories and the fact that certain naphthoquinones display antimalarial activity, the quinones and quinonimines of quinoline and its derivatives are of considerable interest.

With the ultimate objective of preparing potential drugs based on quinonimines such as IV or on quinones such as VI as well as on the quinolinequinone (dihydroquinolinedione) analogs of the naphthoquinone antimalarials, a general study of the quinolinequinones has been initiated in these laboratories. In this paper are considered certain quinones and hydroquinones related to pentaquine (I) as shown in Fig. 1.

The hydroxy compounds (IIIa, IIIb, Va and Vb) were prepared as hydrobromides by hydrolysis of the related 5,6-dimethoxy-8-aminoquinolines, IIa and IIb, with constant-boiling hydrobromic acid under nitrogen. The salts were isolated directly from the reaction mixture without liberation of the highly unstable free bases.

The possibility of obtaining 5-hydroxy-6-methoxy-8-aminoquinolines (III) by selective hydrolysis of the 5,6-dimethoxy compounds (II) was suggested by the observation that the 5-methoxyl group in 5,6-dimethoxy-8-nitroquinoline

(1a) This work was supported by Contract RG 191 and continuation grants from the National Institutes of Health.

(1) Dihydroquinolinediones.

(2) F. Schönhöfer, *Z. physiol. Chem.*, **274**, 1 (1942).

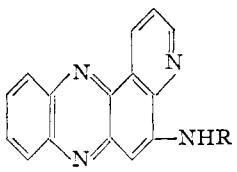
(3) K. C. Blanchard, in Wiselogle, "A Survey of Antimalarial Drugs 1941-1945," J. W. Edwards, Ann Arbor, Mich., 1946, Vol. I, p. 129 ff.

(4) The isolation of 5,6-quinolinequinone from the urine of rabbits and humans after the administration of quinoline has been claimed [H. Führer, *Arch. exp. Path. Pharm.*, **55**, 27 (1906); B. Schuenemann, *ibid.*, **100**, 51 (1923)].

(5) The broken arrow signifies that the reaction products have not been isolated and characterized.

is extremely labile,[6] and by the fact that in acidic media the influence of the 8-amino group is similar to that of the 8-nitro group. It was found that under controlled conditions both IIa and IIb yielded products that were completely soluble in alkali; furthermore, analytical data showed that only one of the methoxyl groups had been cleaved. The melting point of the free base of the product from IIa corresponded to that previously reported for 5-hydroxy-6-methoxy-8-aminoquinoline (IIIa) prepared by an unequivocal procedure.[7] Further evidence for both structures IIIa and IIIb was the fact that upon exposure to air in alkaline solution these compounds were very rapidly converted to quinonoid products which could be formed only if the hydroxyl group is located at the 5-position. Although the quinonimines IVa and IVb have not yet been isolated in the pure state, the presence of these oxidation products was indicated by characteristic color changes observed upon reduction with hydrosulfite and subsequent oxidation by air.

For complete hydrolysis of both methoxyl groups of the 5,6-dimethoxy-8-aminoquinolines, IIa and IIb, to form the corresponding 5,6-dihydroxy compounds, Va and Vb, a reaction period of about 4 hours at the reflux temperature was required. Very little cleavage of the 8-amino groups was noted under these rigorous conditions.[8] The two 5,6-dihydroxy-8-aminoquinolines (V) were rapidly oxidized to the quinones, VIa and VIb, after exposure of the alkaline solutions to air for a short time. Although 8-amino-5,6-quinolinequinone (VIa) was readily isolated as the crystalline free base, the quinone with the pentaquine side-chain (VIb) was not prepared in the pure state. Both quinones were characterized by way of the *o*-phenylenediamine condensation products (VIII).



VIIIa, R = H
VIIIb, R = $—(CH_2)_5NHCH(CH_3)_2$
VIIIc, R = $—COCH_3$

In a second method for the synthesis of the 8-amino-5,6-quinolinequinones (VI), the readily available 6-hydroxy-8-aminoquinolines, VIIa and VIIb, were treated with oxygen in sodium carbonate and sodium bicarbonate solutions, respectively; oxidation occurred relatively rapidly at room temperature. Inasmuch as the yield of VIa was about 50%, the procedure constitutes a practical synthesis for this compound. The more complex quinone VIb was very difficult to isolate and no attempt was made to increase the low yield (15%) obtained. The oxidation of demethylated pentaquine (VIIb) by this process is of theoretical interest, but apparently compound VIb is more satisfactorily prepared from Vb. The oxidation products were identified by the comparison of derivatives with those of authentic quinones prepared from the 5,6-dihydroxy-8-aminoquinolines (Va and Vb).

8-Amino-5,6-quinolinequinone (VIa) is soluble in potassium hydroxide, but only sparingly soluble in sodium carbonate or ammonia solutions. The same solubility properties have been observed for the analogous 4-amino-1,2-naphthoquinone[9] which has been shown to exist predominately in the ortho quinonoid form but which tautomerizes to the para quinonimine structure under the influence of strong alkali. It is probable, therefore, that 8-amino-5,6-quinolinequinone has the structure VIa except in the presence of strong alkali. This quinone (VIa) forms an N-acetyl derivative which condenses with *o*-phenylenediamine to yield VIIIc. The latter is also obtained by acetylating VIIIa, the *o*-phenylenediamine condensation product of the original quinone (VIa). The amino group of VIa is readily removed by heating in acidic or alkaline media. In contrast to 5,6-quinolinequinone,[10] neither the 8-amino derivative (VIa) nor its reduced form (Va) gives a blue color with ammonia solution. Although Va would be the expected 1,4-addition product of quinolinequinone with ammonia, it is apparently not an intermediate in the formation of the blue compound obtained by treating 5,6-quinolinequinone with aqueous ammonia.

If the Schönhöfer theory is valid, IVb and VIb, provided they are sufficiently stable, should be very effective antimalarials and should also appear as metabolites of pentaquine (I) and demethylated pentaquine (VIIb), respectively. Compounds VIb and VIIb are likewise possible metabolites of pentaquine since it has been found[11] that atebrin is partly demethylated in the human body to the corresponding 7-hydroxyacridine. Although a quinonimine could also be formed from pentaquine by oxidation at the 7-position, the antimalarial activities of certain derivatives of the 6-methoxy-8-aminoquinolines indicate that it is more probable that transformation products of type IVb or VIb would be active.[3] Furthermore, the ease with which the 6-hydroxy-8-aminoquinolines (VII) are oxidized to 8-amino-5,6-quinolinequinones (VI) by oxygen not only provides chemical evidence in favor of the Schönhöfer theory, but also indicates that the 5-position of the 8-aminoquinolines is the more likely point of attack.

Under the conditions of either *in vitro* or *in vivo* assays it would be expected that the hydroquinones, IIIb and Vb, would be rapidly oxidized to the quinones, IVb and VIb, as described above. It

(6) R. C. Elderfield, *et al.*, THIS JOURNAL, **68**, 1584 (1946).

(7) W. A. Jacobs and M. Heidelberger, *ibid.*, **44**, 1077 (1922).

(8) It is interesting to compare these results with those of Jacobs and Heidelberger[7] who hydrolyzed 5,8-diamino-6-methoxyquinoline with hydrochloric acid. Under mild conditions cleavage of only the 5-amino group occurred and 5-hydroxy-6-methoxy-8-aminoquinoline (IIIa) was obtained. Under more rigorous conditions both amino groups were removed by hydrolysis and the product was 5,8-dihydroxy-6-methoxyquinoline. If the intermediate in the latter reaction as well as in our drastic hydrolysis of 5,6-dimethoxy-8-aminoquinoline (IIa) is IIIa, the relative stability of the 8-amino group of IIIa toward hydrobromic acid is somewhat suprising in view of its instability toward hydrochloric acid.

(9) L. F. Fieser and M. Fieser, THIS JOURNAL, **56**, 1565 (1934).

(10) J. Mathēus, *Ber.*, **21**, 1886 (1888); H. Führer, *Arch. Pharm.*, **244**, 602 (1906).

(11) D. L. Hammick and D. Firth, *Nature*, **154**, 461 (1944); D. L. Hammick and S. F. Mason, *ibid.*, **156**, 718 (1945).

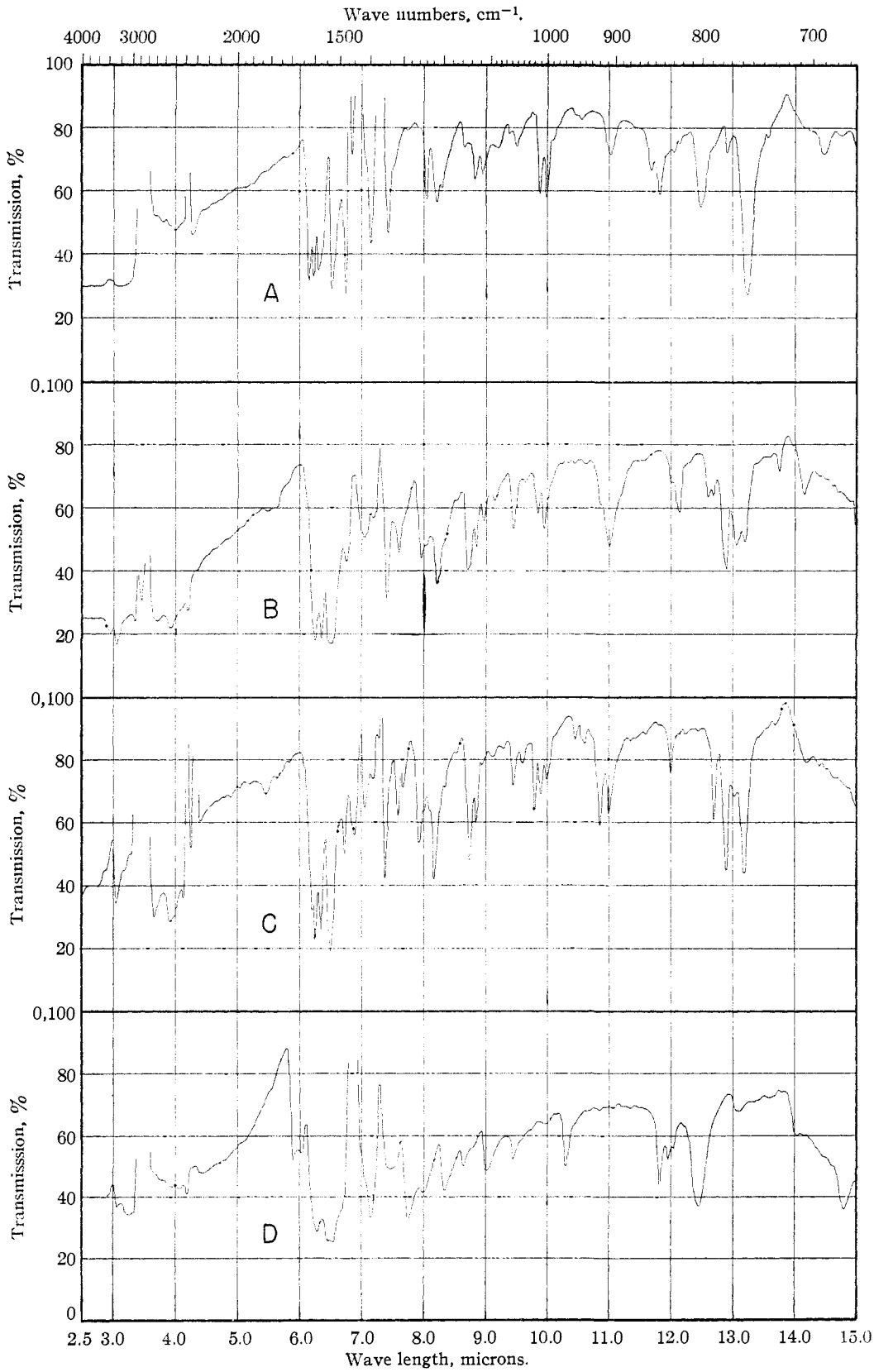


Fig. 2.—Infrared absorption spectra: A, compound VIIIa; B, hydrobromide of compound VIIIb from Vb; C, hydrobromide of compound VIIIb from VIIb; D, compound VIa.

was felt, therefore, that the quinonimines and quinones could be best subjected to biological tests in the form of their more accessible hydroquinones. That this assumption is justified, at

least for the *in vitro* test discussed below, is indicated by the fact that the hydroquinone Va showed essentially the same activity as the derived quinone VIa.

In studies of antimalarial activity it was of particular interest to determine the direct action of the compounds upon parasites *in vitro*. In tests of this nature the drugs undergo limited chemical alterations and are not subject to gross physiological distribution effects which are frequently a determining factor in *in vivo* assays. The compounds prepared here were kindly tested by Greenberg, Taylor and Josephson[12] against the erythrocytic forms of *P. gallinaceum in vitro*. These workers had previously found that pentaquine (I) and 5-methoxypentaquine (IIb) were essentially inactive even at a concentration of 5000 micrograms per l. In contrast, 5-hydroxypentaquine (IIIb) and demethylated pentaquine (VIIb) were active at a concentration of 600 micrograms per l. while the dihydroxy compound, Vb, was active at 150 micrograms per l. Compounds IIIa, Va and VIa, which lack the side-chain on the 8-amino group, were active at roughly 2500 micrograms per l. A tentative explanation of the activity of demethylated pentaquine (VIIb) and the inactivity of pentaquine (I) is that under the conditions of the test VIIb is partially oxidized to the quinone VIb as described above but that pentaquine is not converted to the quinonimine IVb. In support of this it was found that when pentaquine (I) was treated with oxygen under the same conditions that led to the conversion of VIIb to VIb, I was recovered unchanged in practically quantitative yield.

The hydroquinones as well as the quinone VIb all caused the development of methemoglobin *in vitro*.[13] Compounds IIIb, Vb and VIIb were only about one-fifth as active *in vivo* as pentaquine (I) against *P. gallinaceum* in chicks when administered orally.[12] It is possible that the quinonimine IVb and the quinone VIb, formed from IIIb and Vb, respectively, are too rapidly destroyed to serve as efficient drugs *in vivo*. Dr. L. H. Schmidt kindly tested IIIb and Vb against *P. cynomolgi* in monkeys and found that neither compound showed curative action when administered orally or intramuscularly in conjunction with quinine.[14] Since current interest in the 6-methoxy-8-aminoquinolines is based primarily upon their curative action, it is hoped that future work will determine whether the inactivity of IIIb and Vb is due to actual ineffectiveness against the exoerythrocytic forms of *P. cynomolgi*, which are responsible for relapses, or to the instability or unfavorable distribution of these compounds or their quinones in the host organism.

The results of the *in vitro* assays, however, appear to support the Schönhöfer theory for the action of 8-aminoquinolines against *P. gallinaceum*. Confirmation of the postulate that the quinonimine IVb, or the quinone VIb, is actually a metabolite of pentaquine (I) is contingent upon the isolation and identification of such metabolic products or their derivatives. The spectra of some of the more stable compounds prepared here have been determined (Figs. 2 and 3) in the hope that work on these or related substances in other laboratories would thereby be facilitated.

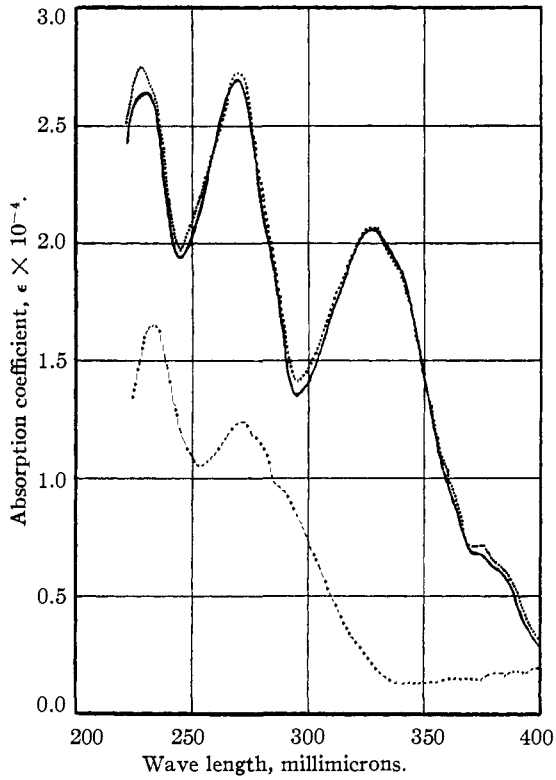


Fig. 3.—Ultraviolet absorption spectra in 0.1 *N* hydrochloric acid: ——, compound VIIIb from Vb; ....., compound VIIIb from VIIb; ---·---, compound VIa.

Experimental[15,16,17]

5-Hydroxy-6-methoxy-8-aminoquinoline (IIIa, **DR 15,896**).[18]—A solution of 0.5 g. of 5,6-dimethoxy-8-aminoquinoline[8] (IIa) in 2.5 ml. of 48% hydrobromic acid was heated in a bath maintained at 100° (internal temperature about 93°). Nitrogen was first bubbled through the solution and then passed over the surface after the product began to precipitate. After 5 hours a small sample of the reaction mixture was found to be completely soluble in alkali. The mixture was cooled in ice and the orange-yellow crystalline product (0.55 g., 63%) was filtered and dried under reduced pressure. It gradually darkened when placed in a melting-point bath at 230° and heated rapidly; it blackened and melted at 239–240° with effervescence.

Anal. Calcd. for $C_{10}H_{10}N_2O_2 \cdot 2HBr$: C, 34.11; H, 3.44; methoxyl, 8.81. Found: C, 34.24, 34.36; H, 3.66, 3.55; methoxyl, 9.05, 9.06.

No attempt was made to determine optimum conditions or maximum yields.

The free base was obtained by treating an aqueous solution of the above salt with an excess of sodium acetate solution containing a trace of sodium hydrosulfite. The yellow precipitate was filtered and washed with water. After excess water was removed with the aid of a rubber dam to prevent oxidation, the product was dried under di-

(12) J. Greenberg, D. J. Taylor and E. S. Josephson, *J. Infectious Diseases*, in press.

(13) E. S. Josephson and co-workers, personal communication.

(14) L. H. Schmidt, personal communication.

(15) The authors wish to thank Mrs. Mary Aldridge and Mr. Byron Baer for the microanalyses.

(16) All melting points are corrected.

(17) The hydrobromic acid used in these experiments was redistilled and stored under carbon dioxide.

(18) This is the designation assigned at the Drug Repository of the National Institutes of Health.

minished pressure. It melted at 179–181° with previous darkening and sintering.[7]

When an alkaline solution of this compound was shaken in air a few minutes, the color changed from orange to dark green and a very insoluble green solid, presumably the quinhydrone, separated out. Upon the addition of hydrosulfite this precipitate dissolved and the orange color was restored.

5-Hydroxy-6-methoxy-8-(5′-isopropylaminoamylamino)-quinoline (IIIb, DR 15,895).—The dihydrobromide monohydrate of IIb (DR 15,332)[19] was prepared by treating a solution of the free base in absolute alcohol with 4% hydrobromic acid and precipitating with anhydrous ether. It was recrystallized from alcohol and ether; m.p. 142.5–143°.

Anal. Calcd. for $C_{19}H_{29}O_2N_2 \cdot 2HBr \cdot H_2O$: C, 44.63; H, 6.50; N, 8.22. Found: C, 44.65, 44.56; H, 6.43, 6.41; N, 8.07, 8.14.

A solution of 0.5 g. of the above salt in 2.5 ml. of 48% hydrobromic acid was heated in a bath maintained at 90° (internal temperature, 86–87°) while a slow stream of nitrogen was admitted under the surface of the liquid. A test sample withdrawn after 6 hours contained only a trace of alkali-insoluble material. After an additional half-hour of heating, the solution was cooled under nitrogen and treated with 7.5 ml. of absolute alcohol and the crystalline salt of the product was precipitated by the addition of 45 ml. of anhydrous ether.[20] The gold-yellow product weighed 0.45 g. (85%) and was completely soluble in alkali. It darkened rapidly when placed in a melting-point bath at 170° and melted at 191.5–192.5° with preliminary sintering.

Anal. Calcd. for $C_{18}H_{27}N_3O_2 \cdot 3HBr \cdot H_2O$: C, 37.39; H, 5.58; methoxyl, 5.37. Found: C, 37.38, 37.45; H, 5.58, 5.49; methoxyl, 5.31, 5.25.

As an indication of homogeneity it was found that no significant change in melting point or carbon and hydrogen analysis occurred upon recrystallization by the method used for Vb.

A dilute solution of this compound when treated with excess alkali was orange for only a second and then became dark. The color changed to orange again after the solution was shaken a short time in air. This series of changes was reversed by the addition of hydrosulfite to the oxidized product. The transient intermediate dark color was apparently due to the quinhydrone.

5,6-Dihydroxy-8-aminoquinoline (Va, DR 15,898).—In the preparation of this compound the time required for hydrolysis depended upon the relative amount of hydrobromic acid used because of the insolubility of the dihydrobromide of the monomethoxy compound IIIa. The yield of Va was not diminished when a large excess of hydrobromic acid was employed.

Two grams of 5,6-dimethoxy-8-aminoquinoline (IIa) was heated with 30 ml. of 48% hydrobromic acid at the reflux temperature for 3.5 hours. Air was excluded as in the preparation of IIIa. The reaction mixture was cooled in ice under nitrogen and the crystalline precipitate (3.0 g., 91%) was filtered off. The product contained 0.16% methoxyl.

A sample of this material (0.5 g.) was purified by dissolving in 15 ml. of methanol, treating with 2.5 ml. of 48% hydrobromic acid and precipitating with 75 ml. of ether. The bright yellow dihydrobromide (0.45 g., 82% over-all yield) apparently contained a trace of IIIa. When placed in a melting-point bath at 225° and heated rapidly, the salt gradually became greenish-gray above 245° and melted as a black resin at about 270°.

Anal. Calcd. for $C_9H_8N_2O_2 \cdot 2HBr$: C, 31.98; H, 2.98; methoxyl, 0.00. Found: C, 32.27, 32.41; H, 3.29, 3.37; methoxyl, 0.30, 0.21.

5,6-Dihydroxy-8-(5′-isopropylaminoamylamino)-quinoline (Vb, DR 15,873).—A solution of 0.5 g. of the dihydrobromide of IIb in 2.5 ml. of 48% hydrobromic acid was heated under reflux for 3.5 hours. When the solution was cooled under nitrogen and treated with 7.5 ml. of absolute alcohol and 45 ml. of anhydrous ether,[20] 0.45 g. (84%) of gold-yellow crystals was obtained. Upon the addition of 10 to 15 ml. of ether to the mother liquor more of the crystalline product precipitated. The first crop melted at 208.5–210° with preliminary darkening and sintering if placed in the bath at 190°. Although according to the analytical data this product contained a trace of methoxy compound (IIIb), the above procedure is convenient for the preparation, in high yield, of material suitable for most purposes without recrystallization.

Anal. Calcd. for $C_{17}H_{25}N_3O_2 \cdot 3HBr$: C, 37.38; H, 5.17; methoxyl, 0.00. Found: C, 37.24, 37.19; H, 5.52, 5.23; methoxyl, 0.33, 0.32.

When this experiment was repeated with a reflux period of 4 hours, the same yield of a product melting below 170°[21] but completely free of methoxyl, was obtained. A sample of this material (0.38 g.) was recrystallized by dissolving in 6 ml. of absolute alcohol, treating with 2 ml. of 48% hydrobromic acid and precipitating with 30 ml. of anhydrous ether.[20] The product (0.31 g., 78% over-all) melted at 210.5–211.5° (dec.). After a second recrystallization the melting point was 212–213° (dec.) and the over-all yield was 69%.

Anal. Calcd. for $C_{17}H_{25}N_3O_2 \cdot 3HBr$: C, 37.38; H, 5.17; Br, 43.90. Found: C, 37.47, 37.43; H, 5.42, 5.47; Br, 43.94, 44.09.

8-Amino-5,6-quinolinequinone (VIa, DR 15,897) from Va.—The sample of Va best suited for this conversion was that obtained by hydrolyzing IIa as described above but increasing the reaction period to 4 hours. The product from 1.0 g. of IIa was dissolved in water and treated with an equal volume of 1 *N* potassium hydroxide. The solution was then shaken intermittently during a 20-minute period in an open flask. Some of the quinone precipitated as red needles at this point and the remainder separated upon the addition of excess acetic acid to the suspension. After the mixture had been cooled in ice, the quinone was filtered off. The yield was 0.69 g. (81% from 5,6-dimethoxy-8-aminoquinoline). Upon recrystallization from 550 ml. of water (a trace of dark material was filtered from the hot solution), there was obtained 0.51 g. (60% over-all yield) of 8-amino-5,6-quinolinequinone; it did not have a definite melting point.

Anal. Calcd. for $C_9H_6N_2O_2$: C, 62.08; H, 3.47; N, 16.09. Found: C, 62.32, 62.19; H, 3.52, 3.58; N, 15.96, 15.93.

This quinone was only sparingly soluble in most organic solvents and dissolved very slowly in dilute hydrochloric acid. It was readily soluble in potassium hydroxide but only very slightly soluble in sodium carbonate or ammonia solutions. It did not develop a blue color with dilute ammonia.

8-Amino-5,6-quinolinequinone (VIa, DR 15,897) from VIIa.—One gram of 6-hydroxy-8-aminoquinoline (VIIa) was almost completely dissolved in a hot solution of 0.8 g. of sodium carbonate monohydrate in 70 ml. of water. After it was cooled to room temperature, the turbid solution was treated with a rapid stream of oxygen for 3 hours. The resulting dark red precipitate (0.48 g.) was filtered off and washed with water. The combined filtrate and washings were further treated with oxygen overnight. The second crop of red precipitate was isolated as before and combined with the first giving a total yield of 0.70 g. (65%) of crude quinone.

One-half gram of this crude product was triturated and washed with 6 ml. of 0.1 *N* HCl and finally washed with water. The red solid was then dissolved in dilute potassium hydroxide and the solution was filtered and acidified with acetic acid. The resultant red needles, which weighed 0.40 g. (58% yield from 6-hydroxy-8-aminoquinoline VIIa), were not pure as shown by the analytical data but were satisfactory for the preparation of derivatives.

Anal. Calcd. for $C_9H_6N_2O_2$: C, 62.08; H, 3.47. Found: C, 62.62, 62.58; H, 3.98, 4.00.

The above hydrochloric acid extract was made strongly alkaline and then treated with excess acetic acid. The gray

(19) Drake, *et al.*, THIS JOURNAL, **68**, 1529 (1946).

(20) The solvents were previously freed of air by boiling and then cooled under carbon dioxide. The precipitation was carried out under carbon dioxide so that air was excluded as much as possible without the use of special equipment. External cooling was applied when the alcohol and hydrobromic acid were mixed. The crystals were filtered rapidly and the funnel containing the product was quickly placed over potassium hydroxide in a desiccator which was immediately evacuated. Some of the salts were hygroscopic if impure.

(21) The results were essentially the same when the nitrogen used for the hydrolysis was first passed through Fieser's solution to remove all traces of oxygen.

precipitate (0.07 g.) thus obtained did not display quinonoid properties but will be further investigated.

A second sample of the original crude quinone (VIa) (0.18 g.) was recrystallized from 140 ml. of water. The purified material weighed 0.12 g. (43% from 6-hydroxy-8-aminoquinoline) and gave satisfactory analytical data.

Anal. Calcd. for $C_9H_6N_2O_2$: C, 62.08; H, 3.47. Found: C, 61.82, 61.77; H, 3.85, 3.90.

The solubility properties of this product were identical with those of the quinone (VIa) prepared from 5,6-dihydroxy-8-aminoquinoline (Va). For confirmation of the structure the oxidation product was acetylated and the resulting acetylaminoquinolinequinone was treated with *o*-phenylenediamine to yield VIIIc as described below. By means of melting point and mixed melting point determinations the two acetyl derivatives were shown to be identical with those obtained from 8-amino-5,6-quinolinequinone (VIa) prepared by the oxidation of 8-amino-5,6-dihydroxyquinoline (Va).

The acetylation of the quinone VIa was carried out by heating a suspension of 0.20 g. of 8-amino-5,6-quinolinequinone (Va) in 5 ml. of acetic anhydride at the reflux temperature. After about 15 minutes practically all of the quinone had dissolved and heating was continued for 15 minutes longer. The solution was cooled somewhat and treated successively with water and solid sodium bicarbonate to remove excess acetic anhydride and most of the acetic acid formed. The mixture was then extracted with ethylene dichloride. The dark brown extract was washed once with a small volume of water and then shaken a short time with Drierite. Before removal of the drying agent, Norite was added and the suspension was shaken at room temperature. Upon evaporation of the filtered solution under diminished pressure, there was obtained 0.18 g. (70%) of an orange crystalline solid. This was recrystallized from about 30 ml. of absolute alcohol (Norite) and yielded 0.15 g. of orange-yellow product which melted at 210.5–211.0°. This sample was submitted for analysis since the melting point of a small portion was unchanged after a second recrystallization. The data corresponded to that calculated for 8-acetylamino-5,6-quinolinequinone (DR 15,966).

Anal. Calcd. for $C_{11}H_8O_3N_2$: C, 61.11; H, 3.73. Found: C, 61.11, 61.27; H, 4.04, 4.02.

5-Aminopyrido[3,2-a]phenazine (VIIIa) was prepared by treating 8-amino-5,6-quinolinequinone (VIa) with *o*-phenylenediamine as follows. A suspension of the quinone (0.20 g.) in 20 ml. of acetic acid was boiled 1 or 2 minutes and treated with 0.15 g. of pure powdered *o*-phenylenediamine. The resulting clear red solution was allowed to stand at room temperature overnight. It was then treated with water and excess sodium bicarbonate. The orange solid, which separated in quantitative yield, was filtered off and washed with water. An analytical sample prepared by several recrystallizations from 95% alcohol melted at 257–258°.

Anal. Calcd. for $C_{15}H_{10}N_4$: C, 73.16; H, 4.09. Found: C, 73.22, 73.07; H, 4.24, 4.29.

This product, as well as the other phenazine derivatives (VIIIb and VIIIc), gave a bright green color with concentrated sulfuric acid.

5-Acetylaminopyrido[3,2-a]phenazine (VIIIc) was obtained by two different procedures. In the first method a solution of 0.50 g. of 8-acetylamino-5,6-quinolinequinone in 1.5 ml. of acetic acid was treated at the boiling point with 0.30 g. of powdered *o*-phenylenediamine. After the solution had been allowed to stand at room temperature overnight, water was gradually added and a practically quantitative yield of yellow crystalline product separated. It was recrystallized from absolute alcohol as stout yellow needles which melted at 256–257°.

Anal. Calcd. for $C_{17}H_{12}N_4O$: C, 70.82; H, 4.20. Found: C, 70.75, 70.73; H, 4.59, 4.42.

The second method for preparing VIIIc consisted in heating a solution of about 0.05 g. of 5-aminopyrido[3,2-a]-phenazine (VIIIa) in 0.3 ml. of acetic anhydride at the reflux temperature for 10 minutes. When the cooled solution was treated with water, about 0.04 g. of orange solid was obtained. After recrystallization from absolute alcohol this product melted at 255.0–255.5°. The mixed melting point with the *o*-phenylenediamine condensation product of 8-acetylamino-5,6-quinolinequinone (above) was 255–256°.

8-(5′-Isopropylaminoamylamino)-5,6-quinolinequinone (VIb) from Vb.—A solution of 0.20 g. of the trihydrobromide of Vb in 10 ml. of water was treated with an equal volume of saturated aqueous sodium bicarbonate and intermittently shaken in an open flask for one-half hour at room temperature. The color became very dark brown almost immediately and then changed gradually to deep red. Concentrated aqueous ammonia was added until the *p*H was about 10. The solution was then quickly extracted several times with ethylene dichloride and the extract was washed with water containing sufficient ammonia to maintain a *p*H of 10. After superficial drying over Drierite, the ethylene dichloride solution was evaporated to dryness under reduced pressure. The residual red oil (VIb) weighed 0.09 g.

This crude product was converted to the phenazine VIIIb by dissolving in 2 ml. of acetic acid and treating with 0.15 g. of powdered *o*-phenylenediamine. The reaction mixture was warmed to about 80° and allowed to stand at room temperature for 2 days. It was then diluted with water, taken to *p*H 10 with aqueous ammonia with cooling and extracted with ethylene dichloride. The extract was washed with very dilute ammonia (*p*H 10) and shaken with Drierite. The yellow fluorescent solution was evaporated to dryness and the residue was dissolved in absolute alcohol and treated with Norite at the boiling point. After filtration several drops of 48% hydrobromic acid were added and the deep red crystals of the hydrobromide of 5-(5′-isopropylaminoamylamino)-pyrido[3,2-a]phenazine (VIIIb) were precipitated with anhydrous ether. The yield was 0.16 g. (77% from Va). This product was readily recrystallized from absolute alcohol–anhydrous ether. The melting point behavior was somewhat indefinite. The microtechnique (Kofler Micro Hot Stage) was used and the sample was slowly heated in the apparatus from 75° to 115° over a period of about 15 minutes to permit gradual dehydration. Original preparations melted at about 200–202°. After recrystallization the individual particles melted at 200–202° but the aggregates were converted to larger crystals which did not begin to round off until temperatures of 230–232° were reached. After further recrystallization no melting was observed below 231–233° at which temperatures the crystals began to round off; complete liquefaction was not observed near this range.

Anal. Calcd. for $C_{23}H_{27}N_5 \cdot 2HBr \cdot 2H_2O$: C, 48.34; H, 5.82. Found: C, 48.71, 48.60; H, 5.54, 5.63.

8-(5′-Isopropylaminoamylamino)-5,6-quinolinequinone (VIb) from VIIb.—The dihydrochloride of demethylated pentaquine (VIIb; DR 15,324)[19] was prepared by treating the free base with hydrogen chloride in alcohol solution and precipitating with anhydrous ether; or by hydrolyzing pentaquine dihydrochloride with 20% hydrochloric acid under reflux and isolating by the method used for IIIb. Upon recrystallization from absolute alcohol and anhydrous ether the dihydrochloride of VIIb melted at 194–196°.

Anal. Calcd. for $C_{17}H_{25}N_3O \cdot 2HCl$: C, 56.66; H, 7.56; N, 11.66. Found: C, 56.45, 56.44; H, 7.68, 7.70; N, 11.44, 11.57.

One-half gram of this salt was dissolved in 25 ml. of water, 10 ml. of saturated aqueous sodium bicarbonate was added and the turbid solution was treated with a rapid stream of oxygen at room temperature. After about 20 minutes an orange precipitate appeared. It gave a strong coupling test with diazotized sulfanilic acid[22] and was probably the starting material (VIIb). A few drops of octyl alcohol were added as required to control foaming. After 2.5 hours the reaction mixture still gave a positive coupling reaction but after an additional hour this test for VIIb was negative and the orange precipitate had been replaced by a greenish-yellow solid. The stream of oxygen was discontinued after a total reaction period of 4 hours.

The precipitate was filtered off and washed with 2% sodium bicarbonate solution and water and the washings were combined with the filtrate. The precipitate was freed of octyl alcohol by washing with petroleum ether. This solid by-product (0.01 g.) did not couple with diazotized sulfanilic acid and did not display quinonoid properties. It will be further investigated.

The aqueous filtrate from the oxidation reaction was taken

(22) B. B. Brodie, S. Udenfriend and J. V. Taggart, *J. Biol. Chem.*, **168**, 328 (1947).

to about *p*H 10 with concentrated ammonia solution and extracted with ethylene dichloride. The extract was washed with very dilute ammonia (*p*H 10) and dried a short time over Drierite. Before filtration the suspension was shaken with Norite. The filtered solution was then evaporated to dryness yielding the crude quinone (VIb) as a dark orange oil containing octyl alcohol.

For identification this oil was converted to the phenazine VIIIb. Half of the above product was dissolved in 2 ml. of acetic acid and treated with 0.20 g. of powdered *o*-phenylenediamine. The red solution was then warmed on the steam-bath for 15 minutes and allowed to stand at room temperature overnight. After it had been diluted with water, the solution was washed with petroleum ether to remove octyl alcohol. It was then treated with concentrated ammonia solution to bring the *p*H up to about 10 and extracted with ethylene dichloride. The fluorescent extract was washed with 1 *N* potassium hydroxide and dilute ammonia and finally shaken with Drierite and Norite. The residue obtained upon evaporation to dryness under reduced pressure was dissolved in absolute alcohol and treated with a few drops of 48% hydrobromic acid and a small volume of anhydrous ether. When the resulting turbid solution was cooled and scratched and seeded with the hydrobromide of VIIIb prepared from the oxidation product of Vb (see above), a red crystalline precipitate began to form. The suspension was allowed to stand in the refrigerator overnight and then treated with more ether before the product was filtered off. There was apparently a considerable quantity of impurity present since attempts to precipitate most of the salt resulted in the separation of an oil. The crystalline material was dissolved in hot absolute alcohol and reprecipitated by the addition of anhydrous ether. The red salt thus obtained from half of the original crude quinone weighed 0.06 g. (15% from VIIb). The micro melting point of this material was about 194°. After recrystallization from absolute alcohol and ether the melting point behavior showed the same changes as noted for the sample of the dihydrobromide dihydrate of 5-(5'-isopropylaminoamylamino)-pyrido[3,2-a]phenazine (VIIIb) prepared from Vb. The infrared and ultraviolet absorption spectra (Figs. 2 and 3) of the products from the two sources were very similar. It is quite probable that the slight differences between the two sets of curves are due to the presence of traces of impurities in the product obtained from VIIb.

Treatment of Pentaquine (I) with Oxygen.—A sample of pentaquine dihydrochloride was dissolved in water and treated with sodium bicarbonate solution and oxygen under conditions identical with those which led to the oxidation of demethylated pentaquine. After a few minutes much of the pentaquine precipitated from the solution but no other change was observed. At the end of the 4-hour reaction period a visual examination of the colors developed with diazotized sulfanilic acid indicated that one-third of the original pentaquine remained in solution and that no reaction had occurred. The precipitate was filtered off, washed and dissolved in alcohol. The alcohol solution was treated with concentrated hydrochloric acid and the hydrochloride of the base was precipitated with ether. The filtrate was made strongly alkaline and extracted with ethylene dichloride. The extract was washed, dried and evaporated to dryness and the residue was converted to the dihydrochloride. The hydrochlorides from both the precipitate and filtrate of the original reaction mixture melted only slightly below the melting point of pure pentaquine dihydrochloride and the melting points of mixtures with the latter were not depressed.

Absorption Spectra.[23]—The infrared spectra were measured by means of a Perkin–Elmer model 12C recording spectrometer with the pulverized samples suspended in mineral oil. The ultraviolet spectra were determined with a Beckman model DU quartz spectrophotometer, using approximately 0.1 *N* hydrochloric acid as the solvent.

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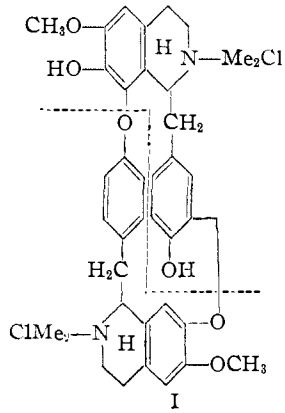
The Synthesis of *dl*-Coclaurine

BY JACOB FINKELSTEIN

In the search for a curare-like acting substance the benzylisoquinoline portion of the *d*-tubocurarine chloride molecule was considered. A natural occurring compound of identical structure with such biological activity has been reported. To verify this the compound had to be synthesized. The greatest handicap to the conventional benzylisoquinoline synthesis has been the difficulty in obtaining the β-phenylethylamines. In this paper several methods were studied and conditions are described for the easy preparation of the desired amine in good yields. The compounds with possible biological significance were tested but found to be devoid of any curare-like activity at the dose levels tried.

With the establishment of *d*-tubocurarine chloride as a useful drug in medicine, much chemical research has been undertaken to find simpler active compounds. Among the investigations undertaken in these laboratories along these lines, consideration was given to the possible existence of an active fragment in the *d*-tubocurarine chloride molecule. When the King[1] formula for the alkaloid I is bisected, each half is represented by the structure of 1-(4-hydroxybenzyl)-6-methoxy-7-hydroxy-2,2-dimethyl-1,2,3,4-tetrahydroisoquinolinium chloride.

A closely related substance, 1-(4-hydroxybenzyl)-6-methoxy-7-hydroxy-1,2,3,4-tetrahydroisoquinoline or *d*-coclaurine, is known. This alkaloid was isolated in pure form by Kondo and Kondo[2] who also established its chemical constitution and structure by degradative methods. Since then no further work has been reported. Additional



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interest in this active fragment idea was furnished from a report by Plugge[3] who tested extracts of the source of this alkaloid and found it to possess a weak and definite curare-like activity. The ex-

(1) King, *J. Chem. Soc.*, 265 (1949).

(2) Kondo and Kondo, *J. prakt. Chem.*, **126**, 24-52 (1930).

(3) Plugge, *Arch. exptl. path. Pharmakol.*, **32**, 266 (1893).