hydrochloride and dicyandiamide were fused as shown. After softening at 108° (bath, 148°), complete fusion occurred at 143° (bath, 150°). The bath temperature was raised gradually to 182° while heating was maintained for 1.25 hours. The cooled reaction product was dissolved in 225 ml. of water, carbon added and the reaction mixture filtered. Addition of 25.0 g. of sodium nitrate precipitated 25.2 g. of oily crystals which were separated and recrystallized from 250 ml. of water. There was obtained 11.6 g. (38%) of the guanidine, m.p. 164–167°; recrystallized (isopropyl alcohol), m.p. 173–174°.

The guanidines isolated in this study are shown in Table VI.

Acknowledgment.—The authors are indebted to Dr. G. Ungar and his staff for the data on the hypoglycemic activity of the compounds.

YONKERS 1, N. Y.

[CONTRIBUTION FROM THE LABORATORY OF CHEMISTRY OF NATURAL PRODUCTS, NATIONAL HEART INSTITUTE, NATIONAL INSTITUTES OF HEALTH AND VARIAN ASSOCIATES]

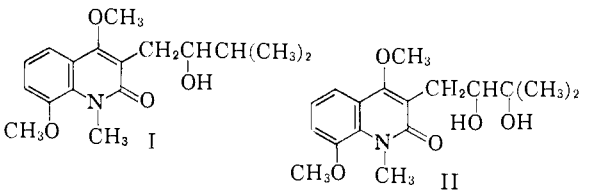
Alkaloids of *Lunasia amara* Blanco. Hydroxylunacridine

BY SIDNEY GOODWIN, J. N. SHOOLERY AND E. C. HORNING

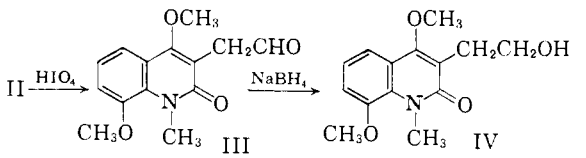
RECEIVED DECEMBER 1, 1958

Hydroxylunacridine has been shown to have the structure II.

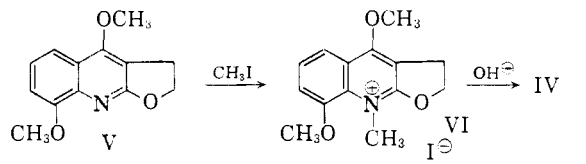
One of the leaf alkaloids of *Lunasia amara* Blanco[1] was found to have the empirical formula $C_{17}H_{23}O_5N$ and to contain two methoxyl groups, one N-methyl group and two active hydrogen atoms. The ultraviolet absorption spectrum was identical with that of lunacridine (I), indicating that the aromatic system was that of a 3-alkyl-4,8-dimethoxy-1-methyl-2-quinolone. The nuclear magnetic resonance spectrum confirmed this relationship; the signals of the aromatic hydrogen nuclei, and of the methoxyl and N-methyl hydrogen nuclei, were identical with those observed for lunacridine. In addition, the n.m.r. spectrum indicated that the side chain arrangement was $-CH_2CHOHCOH(CH_3)_2$. The compound was therefore given the name hydroxylunacridine and considered to be II.



Periodic acid oxidation of hydroxylunacridine yielded acetone, isolated and identified as the 2,4-dinitrophenylhydrazone, and a second carbonyl-containing cleavage product which was also isolated as a 2,4-dinitrophenylhydrazone. The analytical data for the latter compound corresponded to those for an aldehyde derivative of the expected structure III. When the periodic acid oxidation was followed by sodium borohydride reduction *in situ*, a crystalline compound, $C_{14}H_{17}O_4N$, m.p. 120–121.5°, was isolated and was presumed to be the alcohol IV.



This alcohol, a key compound in the structure determination of the alkaloid, may be prepared from γ-fagarine by a sequence of reactions suggested by *Lunasia* chemistry; specifically the dihydrofurano ring opening reaction analogous to the observed conversion of the methyl lunacrinium ion to lunacridine.[2] The requisite dihydro-γ-fagarine (V) may be obtained either by the Grundon–McCorkindale synthesis[5] or from the catalytic reduction of γ-fagarine. The natural material was used here to prepare V which in turn was converted to the methiodide VI. Treatment of the methiodide with dilute sodium hydroxide solution yielded the alcohol IV, m.p. 120–121°, which proved to be identical with the compound isolated



from the degradation of hydroxylunacridine. In addition to the usual comparison, IV from γ-fagarine was converted to the aldehyde 2,4-dinitrophenylhydrazone which was identical with the product obtained through the periodic acid oxidation of hydroxylunacridine.

Nuclear Magnetic Resonance Spectrum.[6]—Although the n.m.r. spectrum was used to predict the structure of the side chain of hydroxylunacridine, it

(1) The leaves and bark of *Lunasia* sp. contain a number of alkaloids not previously described or studied. A summary of the alkaloids isolated from *L. amara* leaves, including hydroxylunacridine, is in preparation. References to earlier work, and a review of current knowledge relating to the "water-soluble" quaternary *Lunasia* bases, the major alkaloid lunacrine, and the related compound lunacridine, are included in a summary by J. R. Price.[2] Structures have been proposed for lunacrine and lunacridine.[3,4]

(2) J. R. Price, "Recent Advances in Heterocyclic Chemistry," Academic Press, Inc., New York, N. Y., 1958, p. 92.

(3) S. Goodwin and E. C. Horning, THIS JOURNAL, **81**, 1908 (1959).

(4) S. Goodwin, J. N. Shoolery and L. F. Johnson, *ibid.*, **81**, 3065 (1959).

(5) M. F. Grundon and N. J. McCorkindale, *J. Chem. Soc.*, 2177 (1957).

(6) The resonance frequencies are given relative to benzene at 60 mc. and the solvent was deuterio-chloroform. The equipment and operating conditions were the same as those described for lunacrine and lunine.[4]

is perhaps more instructive to discuss the spectrum from the point of view of the chemically proved structure II rather than as a structure proof. The spectrum of the side chain consisted of eighteen lines; three of which were temperature dependent (t.d.) and were therefore assigned to the two hydroxyl hydrogen atoms. The eighteen lines were arranged in the following patterns: a, 67.3 and 70.4 c.p.s. (both t.d.); b, 161.0, 163.3, 166.4, 170.9, 173.4 and 176.3 c.p.s.; c, 189.5, 192, 203.4, 206, 219 (t.d.), 222.8, 227 and 237 c.p.s.; and d, 307 c.p.s. The t.d. doublet (pattern a) must arise from spin–spin interaction of the secondary hydroxyl-hydrogen atom and the tertiary (CH) hydrogen; that is, from the –OH of the >CHOH system. The tertiary hydroxyl-hydrogen atom has been assigned to the remaining temperature dependent line (219 c.p.s. in pattern c). Pattern c, exclusive of the t.d. line, has the appearance of the AB portion of the spectrum of an ABX system of spin coupled nuclei. Analysis by the Anderson method afforded the parameters, δ_{AB} = 23.4, J_{AB} = 14.0, J_{AX} = 2.2 and J_{BX} = 10.0 c.p.s. This pattern has been assigned to the CH_2 group of the side chain; the hydrogen atoms are nonequivalent by virtue of attachment of the CH_2 group to an asymmetric carbon atom and restricted rotation implemented by intramolecular hydrogen bonding of the secondary hydroxyl group with the carbonyl group of the 2-quinolone. That hydrogen bonding does occur is indicated by the low resonance frequency of the secondary hydroxyl-hydrogen atom; the unshielding effects of hydrogen bonding are well known. Pattern b has been assigned to the tertiary hydrogen of the side chain (X of the ABX notation). The six lines of this pattern are arranged as a pair of triplets of roughly 1:2:1 relative intensities. This type of multiplet is to be expected of a system involving two small and approximately equal coupling constants (J_{AX} = 2.2 and J_{CHOH} = 3.1 c.p.s.) and one large coupling (J_{BX} = 10 c.p.s.). By inspection of the data for pattern b, it will be noted that the average of the small spacings in the triplets is 2.7 c.p.s. and that the spacing between the centers of the triplets is 10.1 c.p.s. These values are in excellent agreement with the coupling constants derived from patterns a and c and required for b. The resonance frequency of the six C-methyl hydrogen atoms was found to be 307 c.p.s.; there was no fine structure. In the original analysis of the spectrum it was, of course, very significant that the C-methyl hydrogen atoms were not involved in spin–spin interaction because it indicated that the methyl groups were attached to a carbon atom (or atoms) having no attached hydrogen atoms.

The establishment of the structure of hydroxylunacridine provides additional evidence for the previously assigned positions of the methoxyl groups of lunacridine (and hence of lunacrine as well), and indicates the validity of n.m.r. data in suggesting side-chain structures and aromatic substitution in this series.[4]

Acknowledgment.—We are indebted to Mr. L. F. Johnson, Varian Associates, Palo Alto, for the n.m.r. data; to Miss A. A. Velasquez for assistance in the isolation of the hydroxylunacridine used in this study and to Dr. I. J. Pachter, Smith, Kline and French Laboratories, Philadelphia, for a generous quantity of γ-fagarine. We are grateful to Dr. H. Rapoport and Mr. K. G. Holden, University of California, Berkeley, for information related to their investigation of *Balfourodendron* sp.; one of their alkaloids is apparently identical with hydroxylunacridine.

Experimental[7]

Hydroxylunacridine.—The analytical sample[1] was crystallized from ethyl acetate–cyclohexane, m.p. 100–102°, $[\alpha]^{24}_{589}$ +31.5°, $[\alpha]^{24}_{436}$ +84.2° (*c* 6.99, ethanol).

Anal. Calcd. for $C_{17}H_{23}O_5N$: C, 63.53; H, 7.21; N, 4.36; OCH_3, 19.31; $(N)CH_3$, 4.68; active hydrogen (two), 0.63. Found: C, 63.67; H, 7.05; N, 4.29; OCH_3, 21.02; $(N)CH_3$, 5.75; active hydrogen, 0.63.

The infrared spectrum had λ^{Chf}_{max} 2.79(v.w.), 3.0(broad), 6.13(s), 6.21(m) and 6.33(s); absorption in the 6 μ region resembled that found for lunacridine. The ultraviolet absorption spectrum was essentially identical with that of lunacridine.[3] The λ_{max} and λ_{min} values and log ε values in ethanol were λ_{max} 2.39 (4.39), 257 (4.39), 284 (3.93), 293 (3.90), 332 (3.55) and λ_{min}. 224 (4.32), 246 (4.32), 275 (3.88), 291 (3.90), 310 (3.37). The values observed for lunacridine were λ_{max} 239 (4.38), 257 (4.41), 285 (3.94), 294 (3.91), 333 (3.55) and λ_{min} 224 (4.31), 246 (4.31), 275 (3.89), 291 (3.90), 310 (3.36).

Periodic Acid Oxidation of Hydroxylunacridine. (A) Acetone 2,4-Dinitrophenylhydrazone.—A solution of 0.33 g. of hydroxylunacridine, 0.67 g. of periodic acid, 2 ml. of acetic acid and 5 ml. of water was allowed to stand for 3 hours and then diluted with water, made alkaline with 10% sodium hydroxide solution, and steam distilled (internal) into a receiver containing an aqueous sulfuric acid solution of 2,4-dinitrophenylhydrazine. The yellow precipitate crystallized from ethanol as yellow needles, m.p. 126–128°. The m.p. was not depressed on admixture with authentic acetone 2,4-dinitrophenylhydrazone, m.p. 128°, and the infrared spectra of the derivatives were identical.

(B) 4,8-Dimethoxy-1-methyl-3-(β-hydroxyethyl)-2-quinolone.—A solution of 0.45 g. of periodic acid in 5 ml. of water was added to a 5-ml. solution of 0.28 g. of hydroxylunacridine in ethylene glycol dimethyl ether. After standing for several hours, a large excess of sodium borohydride was added. The reaction mixture was diluted with water and extracted with chloroform. The oily residue (0.19 g.) solidified on scratching and afforded colorless needles, m.p. 120–121.5°, on crystallization from aqueous methanol. This product was identical with that obtained from γ-fagarine.

(C) 4,8-Dimethoxy-1-methyl-3-carboxaldehydoethyl-2-quinolone 2,4-Dinitrophenylhydrazone.—The periodic acid oxidation was carried out in aqueous acetic acid (1:1). The crude oily product was isolated by chloroform extraction and was treated with 2,4-dinitrophenylhydrazine in methanol–sulfuric acid solution. Crystallization of the resultant solid from ethyl acetate afforded long yellow needles melting at 214–216°.

Anal. Calcd. for $C_{20}H_{19}O_7N_5$: C, 54.42; H, 4.34; N, 15.87. Found: C, 54.45; H, 4.40; N, 15.79.

Dihydro-γ-fagarine.—A solution of 0.40 g. of γ-fagarine in 10 ml. of ethanol was added to a pre-treated mixture of 50 mg. of palladium–charcoal (10%) and 10 ml. of ethanol in an atmosphere of hydrogen. The mixture was stirred at room temperature and atmospheric pressure until the theoretical volume of hydrogen had been absorbed (50 min.). The product was isolated in the usual manner and on crystallization from aqueous methanol a first crop of 0.29 g. of colorless needles, m.p. 172–173° (reported[5] m.p. 168–170°), was obtained.

4,8-Dimethoxy-1-methyl-3-(β-hydroxyethyl)-2-quinolone.—Dihydro-γ-fagarine methiodide was prepared by allowing

(7) All melting points were taken with a Kofler stage. The instrumental work, exclusive of the n.m.r. spectrum, was carried out by Mrs. K. Warren and Miss P. Wagner. Analyses were performed in the laboratories of Mr. W. Manser, Zurich, Switzerland, and Mr. J. F. Alicino, Metuchen, N. J.

a solution of 0.13 g. of dihydro-γ-fagarine in excess methyl iodide to stand at room temperature for two days. A solution of 0.15 g. of the crude methiodide in 10 ml. of methanol was treated with 5 ml. of 10% sodium hydroxide solution and warmed on a steam-bath for 10 min. After evaporation of most of the methanol, the product was extracted with ether. The ether extracts were washed with 0.1 *N* sulfuric acid solution and sodium bicarbonate solution. The product crystallized from aqueous methanol as colorless needles, m.p. 120–121°. This material was identical (mixed m.p. and infrared spectrum) with that obtained from hydroxylunacridine.

Anal. Calcd. for $C_{14}H_{17}O_4N$: C, 63.86; H, 6.51; N, 5.32. Found: C, 63.89; H, 6.47; N, 5.23.

4,8-Dimethoxy-1-methyl-3-carboxaldehydoethyl-2-quinolone 2,4-Dinitrophenylhydrazone.—A mixture of 4,8-dimethoxy-1-methyl-3-(β-hydroxyethyl)-2-quinolone (70 mg.) from γ-fagarine, chromic acid (50 mg.) and pyridine (7 ml.) was allowed to stand at room temperature. The product was isolated by ether extraction of the reaction mixture after dilution with water. Treatment of the crude product with a solution of 2,4-dinitrophenylhydrazine in ethanol–sulfuric acid solution gave a yellow crystalline derivative, m.p. 213–215°, after crystallization from ethyl acetate. This was identical (mixed m.p., infrared spectrum) with the product from hydroxylunacridine.

BETHESDA 14, MD.
PALO ALTO, CALIF.

[CONTRIBUTION FROM THE DEPARTMENT OF CHEMISTRY, UNIVERSITY OF CALIFORNIA, BERKELEY]

Isolation of Alkaloids from *Balfourodendron riedelianum*. The Structure of Balfourodine

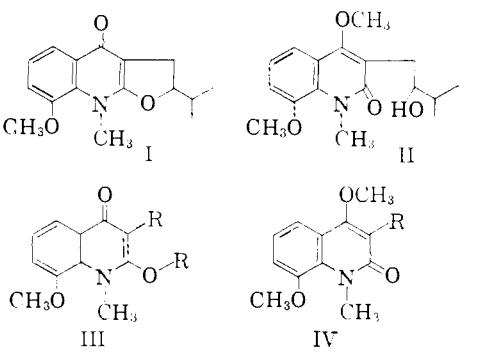
BY HENRY RAPOPORT AND KENNETH G. HOLDEN[1]

RECEIVED DECEMBER 18, 1958

The alkaloids of *Balfourodendron riedelianum* were separated from the crude plant extract by a systematic extraction procedure which fractionated the alkaloids according to their basicities and polarities. The structures of two of the alkaloids, balfourodine and balfourolone, have been determined, and it has further been shown that balfourolone is an artifact of the isolation procedure arising from the base-catalyzed ring opening of an O^4-methylbalfourodinium salt whose structure is also discussed.

Balfourodendron riedelianum, a member of the Rutaceae family, is a small tree or shrub indigenous to Brazil and Argentina where it has found popular use for the treatment of stomach and intestinal ailments. Mundt[2] has reported the isolation of a small amount of alkaloidal material from the mature bark. In the present work the isolation of alkaloids was undertaken in a systematic manner and the structures of the alkaloids present in greatest concentration were determined.

Separation of the alkaloids from other plant material was accomplished by continuous extraction of an aqueous solution of the bark extract[3] with ether and more polar solvents at various *p*H's. By this method a fractionation of the alkaloids according to basicity and polarity was effected as shown in Fig. 1. The crude fractions thus obtained then were separated into fairly pure alkaloidal fractions by chromatography on alumina. Of immediate interest were the chief fractions from B and C, B_1 and C_1, since they, besides being the major alkaloidal fractions, had ultraviolet spectra which were suggestive of 2- and 4-quinolones,[4] respectively. Further investigation showed that the ultraviolet spectra of B_1 and C_1 were qualitatively identical with those of lunacrine (I) and lunacridine (II),[5] respectively. This suggests the part structures III for B_1 and IV for C_1. In view of these considerations and the ready availability of B_1 and C_1, a more detailed examination of these fractions was undertaken.



Compound B_1 upon further purification gave balfourodine, $C_{16}H_{19}O_4N$, m.p. 188–189°. Like lunacrine,[5] balfourodine contains one methoxyl and one N-methyl group, but differs in having an additional oxygen atom. The same relationship holds between lunacridine[5] and balfourolone, $C_{17}H_{23}O_5N$, m.p. 99–100°, obtained by crystallization from fraction C_1. Since the additional oxygen atom, in both compounds, has no effect on the ultraviolet absorption, it most certainly could not be on the quinolone nucleus. Therefore, a glycol grouping in balfourolone was strongly indicated.

Treatment of balfourolone with periodic acid followed by distillation of part of the reaction mixture gave acetone as its *p*-nitrophenylhydrazone. This establishes the part structure $R_2C(OH)C(OH)(CH_3)_2$ for balfourolone. The other fragment of the periodate oxidation was an aldehyde, $C_{14}H_{15}O_4N$, which was characterized as its semi-

(1) Public Health Service Predoctoral Research Fellow of the National Heart Institute.

(2) G. A. Mundt, *Anales farm. y bioquim. (Buenos Aires)*, [2] **21**, 25 (1954).

(3) We are indebted to Dr. Glenn E. Ullyot of Smith, Kline and French Laboratories, Philadelphia, and Dr. Oscar Ribeiro of Instituto de Quimica Agricola, Rio de Janeiro, for their assistance in procuring this material.

(4) E. A. Steck, G. W. Ewing and F. C. Nachod, THIS JOURNAL, **71**, 238 (1949).

(5) S. Goodwin and E. C. Horning, *ibid.*, **81**, 1908 (1959). We are indebted to Dr. Sidney Goodwin of the National Heart Institute for samples of lunacrine and lunacridine as well as for pre-publication information concerning her investigation of the alkaloids of *Lunasia amara* Blanco.