yield 115 mg., m.p. 193–197° dec. Further recrystallization from the same solvent mixture gave the analytical sample, m.p. 195–197° dec., $\nu_{max}^{CH_2Cl_2}$ 1730 cm.$^{-1}$ (broad and unresolved). The substance showed no titratable groups in aqueous dimethylformamide solution and gave a negative Liebermann test.

Anal. Calcd. for $C_{39}H_{48}NO_{16}Cl$: C, 58.10; H, 6.00; N, 1.74; Cl, 4.40. Found: C, 57.95; H, 6.08; N, 1.91; Cl, 4.72.

Conversion of XIV into Oxonitine Triacetate (V).—The chloroformamide (XIV) of the previous experiment (29.5 mg.) was dissolved in 1 ml. of anhydrous formic acid and 1 ml. of acetic anhydride, and the mixture was heated overnight on the steam-bath. The solvent was removed under reduced pressure, and the product was taken up in methylene chloride–ether and washed with water and dilute sodium hydroxide. Crystallization from methylene chloride–petroleum ether afforded 20.3 mg. of material, m.p. 253–255°. Recrystallization from the same solvent pair raised the melting point to 256–257°, $[\alpha]_{5460}^{25}$ −64° (chloroform); $\nu_{max}^{CH_2Cl_2}$ 1730, 1661 cm.$^{-1}$. A mixed melting point determination with an authentic sample of oxonitine triacetate showed no depression, and the infrared spectra of the two specimens in methylene chloride, chloroform and carbon disulfide were identical.

Permanganate Oxidation of Ethyl-labeled Aconine Pentaacetate.—A sample of ethyl-labeled aconine pentaacetate (185 mg.), prepared as previously described, was oxidized with potassium permanganate in acetone by the method employed for oxidation of aconitine triacetate. Chromatography of the crude product on alumina afforded 42 mg. of unchanged starting material and 25 mg. of pure oxonine pentaacetate, m.p. 260–261°, $\nu_{max}^{CHCl_3}$ 1735, 1660 cm.$^{-1}$, identical with an authentic sample. Late fractions from the chromatogram showed infrared absorption at 1626 cm.$^{-1}$ and presumably contained N-acetylnoraconine pentaacetate. However, a pure sample of the latter compound could not be isolated.

Assay of the oxonine pentaacetate established the fact that only 6% of the activity originally present remained in the assay sample.

Anal. Calcd. for $C_{34}H_{47}NO_{15}$: C, 57.54; H, 6.68; N, 1.97; O, 33.82. Found: C, 57.71, 57.68; H, 6.43, 6.52; N, 2.26, 2.43; O, 33.54, 33.41.

[CONTRIBUTION FROM THE RESEARCH AND DEVELOPMENT DIVISION, SMITH, KLINE AND FRENCH LABORATORIES, PHILADELPHIA 1, PENNA.]

The Alkaloids of *Hortia arborea* Engl.[1]

BY I. J. PACHTER, R. F. RAFFAUF, G. E. ULLYOT AND O. RIBEIRO[2]

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Seven bases were isolated from the bark of *Hortia arborea* Engl. Four of these are of the furoquinoline type; dictamine, γ-fagarine, nor-γ-fagarine and skimmianine. The remaining three are of the quinazoline group: rutecarpine and two new alkaloids, hortiamine and hortiacine. Hortiamine, a red alkaloid, $C_{20}H_{17}N_3O_2$, yields 6-methoxy-1-oxo-1,2,3,4-tetrahydropyrid[3,4-b]indole and N-methylanthranilic acid when heated with ethanolic potassium hydroxide. The alkaloid was resynthesized through condensation of these degradation products. It is 10-methoxy-14-methyl-5-oxo-5,7,8,14-tetrahydroindolo[2,3-c]quinazo[3,2-a]pyridine. Hortiamine undergoes hydration in aqueous solvents to form a cyrstalline yellow compound, $C_{20}H_{19}N_3O_3$, which is 2-(*o*-methylaminobenzoyl)-6-methoxy-1-oxo-1,2,3,4-tetrahydropyrid[3,4-b]indole (6-methoxyrhetsinine). The hydrochloride of the alkaloid evolves gas upon fusion to yield a base, $C_{19}H_{18}N_2O_3$ identical with the second new alkaloid, hortiacine, which is shown to be 10-methoxyrutecarpine.

Several years ago our attention was directed to a Brazilian medicinal plant called "casca paratudo" which was said to possess excellent stimulant and stomachic properties. According to Hoehne,[3] "casca paratudo" is the bark of *Drimys winteri* Forst., a member of the family Magnoliaceae.

Samples of "casca paratudo" obtained from a commercial source[4] were found to contain alkaloids which, rather than being stimulant in action, were found to have sedative and hypotensive effects sufficient to warrant isolation and investigation of the individual bases. Initial fractionation studies showed that part of the alkaloids were of the furoquinoline type, typical of the family Rutaceae but not of the Magnoliaceae, and that botanical designation was therefore probably improper. Further investigation revealed that another Brazilian plant, *Hortia arborea* Engl., a member of the Rutaceae, also is called "casca paratudo." The plant is known for its bitter properties but is not reputed to have significant medicinal value. The materials used in the present study[5] were referred to the late botanist, Dr. J. G. Kuhlmann, who informed us that we were indeed dealing with *Hortia arborea* Engl.

The current work thus had fortuitous beginnings. Authentic *Drimys winteri* Forst.[6] was obtained eventually, but was found to possess neither alkaloids nor biological activity of note.

A concentrated ethanolic extract of milled trunk bark of *Hortia arborea* Engl. was partitioned between aqueous ammonia and chloroform. The chloroform layer was extracted with dilute hydrochloric acid, whereupon the alkaloids separated into two fractions: those whose salts remained in acid solution and those whose salts precipitated as yellow solid.

The acid-soluble alkaloids were liberated with aqueous ammonia, dissolved in benzene and precipitated as their hydrochlorides. The latter were dissolved in hot methanol, from which dictamnine (I) hydrochloride crystallized on cooling in 0.003% yield. Dictamnine was identified as its picrate[7] and by direct comparison with an authentic sample.[8]

The residual methanolic solution was evaporated to dryness and the bases regenerated. On stand-

(1) Presented, in part, at the 132nd Meeting of the American Chemical Society, New York City, N. Y., September, 1957, and at the International Congress for Pure and Applied Chemistry, Paris, July, 1957.

(2) Instituto de Quimica Agricola, Ministério da Agricultura, Rio de Janeiro, Brazil.

(3) F. C. Hoehne, "Plantas e Substâncias Vegetais Tóxicas e Medicinais," "Graphicors," São Paulo, Brazil, 1939.

(4) Flora Medicinal, Rio de Janeiro, Brazil.

(5) Specimens and barks derived from trees used in the present study have been deposited at the Herbarium of the Philadelphia Academy of Natural Sciences under numbers 859,232, 859,233 and 859,234.

(6) Specimen number 859,236 at the Herbarium of the Philadelphia Academy of Natural Sciences.

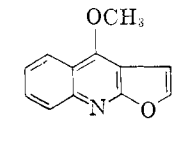
(7) Y. Asahina, T. Ohta and M. Inubuse, *Ber.*, **63**, 2045 (1930).

(8) We are grateful to Dr. J. R. Price of the C.S.I.R.O., Melbourne, Austr., for samples of dictamnine, γ-fagarine and skimmianine.

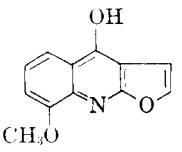
ing, a thick mass of crystals formed. The crystals were separated from residual oil and boiled with benzene. A small quantity of base, $C_{12}H_9NO_3$, m.p. 263–265° dec., remained insoluble. The base was phenolic, contained one methoxyl group and possessed an ultraviolet spectrum similar to those of the furoquinolines. Several years ago, Deulofeu and Bassi[9] reported that a substance of similar properties called nor-γ-fagarine (II) arose when γ-fagarine (III) hydrochloride was heated for 10 minutes at 130–135°. Upon repetition of their experiment, a product was obtained which proved to be identical with the compound in hand.

The alkaloids which disolved in benzene were subjected to chromatography and yielded γ-fagarine (III) in 0.002% yield and skimmianine (IV) in 0.001% yield. The properties of the alkaloids corresponded with data previously recorded[9–11] and identification was confirmed through direct comparison with authentic samples.[8]

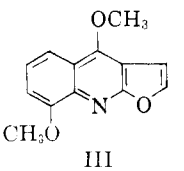
The occurrence of γ-fagarine in *Hortia arborea* Engl. and the ease with which it is converted into nor-γ-fagarine when heated with acid cause us to believe that the nor-γ-fagarine which was isolated is not an alkaloid but is an artifact which originated when the mixed crude hydrochlorides were heated with methanol.



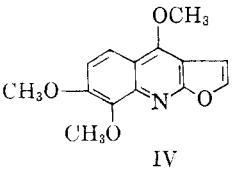
I



II



III



IV

The acid-precipitated yellow alkaloid salts were washed with ethanol and then boiled with water. Most of the material dissolved, but a tan solid was generated in the process. The tan solid was found reconvertible into yellow salts through treatment with excess acid. Hot water had thus effected a separation of the precipitated yellow material into strong bases, whose yellow salts dissolve in hot water, and weak bases, whose yellow salts hydrolyze in hot water.

The hot aqueous filtrate deposited bright yellow crystals upon cooling. Basification of the crystals in the presence of chloroform resulted in a blood-red chloroform solution from which beautiful orange-red needles of a new alkaloid, m.p. 209° dec., $C_{20}H_{17}N_3O_2$, were obtained. The name hortiamine is proposed for this base. It was isolated in 0.02% yield and is the major alkaloid of the bark. It is the one responsible for the hypotensive properties of the total bases.

(9) V. Deulofeu and D. Bassi, *Anales asoc. quim. arg.*, **40**, 249 (1952).

(10) V. Deulofeu, R. Labriola and J. De Langhe, THIS JOURNAL, **64**, 2326 (1942).

(11) K. Takeda, *J. Pharm. Soc. Japan*, **61**, 117 (1941); *C. A.*, **36**, 414 (1942).

Intensely red hortiamine solutions fade to pale orange and yield difficultly soluble yellow prisms when allowed to stand in the presence of water. The yellow prisms, $C_{20}H_{19}N_3O_3$, turn deep red when heated and melt at 209° dec., as does hortiamine. When boiled with anhydrous benzene, the yellow prisms dissolve slowly to generate a blood-red solution from which orange-red needles of hortiamine separate upon concentration. The red and yellow materials are thus readily interconvertible. Their empirical formulas differ by one mole of water.[12]

The ultraviolet spectrum of hortiamine is remarkably dependent upon the nature of the solvent employed, as shown in Fig. 1. Its infrared spectrum in mineral oil mull exhibits a carbonyl band at 5.96μ, but no bands in the NH region.

Prolonged heating with hydrochloric acid simply converts the alkaloid into a monohydrochloride. With 48% hydrobromic acid, however, it undergoes O-demethylation to produce a phenolic compound which dissolves in dilute sodium hydroxide to give a deep magenta-colored solution.

Hortiamine is insoluble in alkali. However, heating with ethanolic potassium hydroxide results in rapid solution accompanied by extensive degradation. A colorless product, m.p. 281–282°, $C_{12}H_{12}N_2O_2$, and an amino acid, m.p. 177–179°, were isolated from an alkaline hydrolysis mixture. The former dissolves in concentrated hydrochloric acid, but separates upon dilution of the solution with water. It possesses a methoxyl group and displays infrared bands at 3.00, 3.15 and 6.02μ. Its ultraviolet spectrum, with a maximum at 305 mμ and minima at 260 and 370μ, strongly resembles those of indole-2-carbonyl compounds. These properties coincide closely with those reported for or expected of 6-methoxy-1-oxo-1,2,3,4-tetrahydropyrid[3,4-b]indole (V).[13,14] An authentic specimen of the compound was prepared by the excellent method developed by Abramovitch and Shapiro[13] and was found to be identical with the hortiamine-derived product.

The amino acid was identified as N-methylanthranilic acid (VI) through mixed melting point and spectral comparisons with an authentic sample.

The two alkaline hydrolysis products account for all of the atoms of hortiamine. While there is more than one way to join V and VI to produce a $C_{20}H_{17}N_3O_2$ compound, the arrangement of atoms in structure 10-methoxy-14-methyl-5-oxo-5,7,8,14-

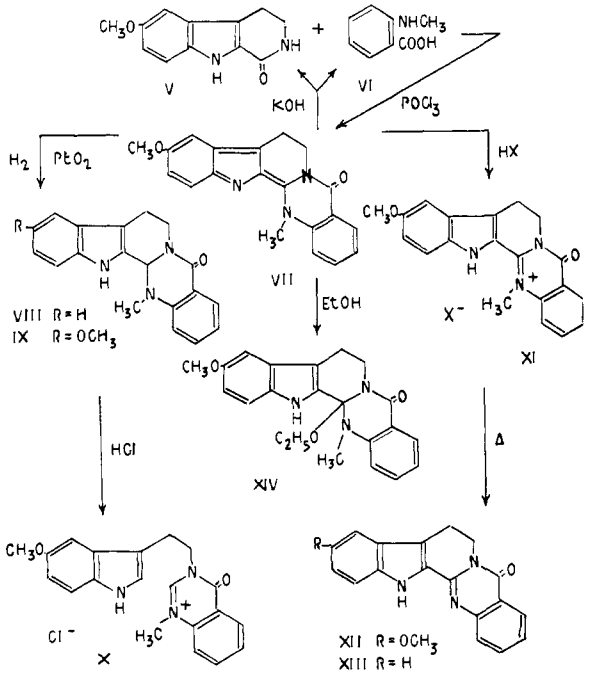
(12) During the course of our work, a paper on the alkaloids of *Hortia arborea* by L. D. Antonaccio and E. Tolmasquim appeared [*Anais acad. brasil. cienc.*, **28**, 183 (1956)]. The authors isolated a single base, m.p. 136–139°, and assigned an empirical formula but no structure. The chemical properties and spectral data which they report suggest that their material may be related to the yellow hydrate of hortiamine.

(13) H. S. B. Barrett, W. H. Perkin, Jr., and R. Robinson, *J. Chem. Soc.*, 2942 (1929); R. A. Abramovitch and D. Shapiro, *ibid.*, 4589 (1956).

(14) The ring systems described in this paper are named and numbered according to "The Ring Index" by A. M. Patterson and L. T. Capell, Reinhold Publishing Corp., New York, N. Y., 1940. "Chemical Abstracts" has employed the same nomenclature. The second edition of "The Ring Index" by A. M. Patterson, L. T. Capell and D. F. Walker, American Chemical Society, Washington, D. C., 1960, which has just come to our attention, suggests pyrido[3,4-b]indole in place of pyrid[3,4-b]indole and indolo[2′,3:3,4]pyrido[2,1-b]quinazoline in place of indolo[2,3-c]quinazo[3,2-a]pyridine.

tetrahydroindolo[2,3 - c]quinazo -[3,2-a]pyridine[14] (VII) is the only one which can account satisfactorily for the properties of the red alkaloid. Structure VII, it may be noted, is related to the colorless alkaloid evodiamine (VIII) derived from *Evodia rutecarpa*, Benth. and Hook., another member of the Rutaceae.

Catalytic hydrogenation of hortiamine produces colorless dihydrohortiamine which may be assigned structure IX; it is 10-methoxyevodiamine, and in fashion similar to evodiamine, it undergoes hydrolysis in aqueous ethanolic hydrochloric acid to form a substance for which, on the basis of previous work with evodiamine,[15,16] structure X is assigned. The compound is called 5-methoxyisoevodiamine chloride.



Hortiamine salts possess structure (XI) and, in mineral oil mull, display a single strong carbonyl band at 5.87 μ. They are actually quaternary salts and undergo N-demethylation upon pyrolysis to produce the 10-methoxy derivative (XII) of rutecarpine (XIII), another alkaloid derived from *Evodia rutecarpa* Benth. and Hook.

(15) Y. Asahina, *Acta Phytochim. (Japan)*, **1**, 67 (1923).

(16) Y. Asahina, *J. Pharm. Soc. Japan*, No. 503, 1 (1924); *C. A.*, **18**, 1667 (1924). In addition to the chemical evidence advanced by the Japanese,[15] the structure proposed for isoevodiamine chloride receives support from the following data. The infrared spectra of isoevodiamine chloride and its 5-methoxy derivative X show strong carbonyl absorption at 5.85 μ. Hortiamine salts (XI) absorb in the same region (5.87 μ). Less intense bands are also present in the isoevodiamine spectra at 6.03 μ and could be due to $>C{=}\overset{+}{N}<$ stretching. In the more highly conjugated hortiamine and anhydrorhetsinine[17,18] salts such absorption is absent, although bands of similar intensity are present at 6.2 μ. The isoevodiamines display no absorption in the 4 μ region characteristic of $=\overset{+}{N}H$. 5-Methoxyisoevodiamine chloride (X) is reversibly converted by alkali into an amorphous substance which no longer displays the 5.85 μ band but shows a strong band at 6.05 μ and a weaker one at 6.2 μ. The ultraviolet spectrum of this amorphous substance, with maxima at 278, 296, 310 and 340 mμ, is related both to methoxyevodiamine (IX) and to *o*-methylaminobenzoyl-5-methoxytryptamine, a compound to be described elsewhere.

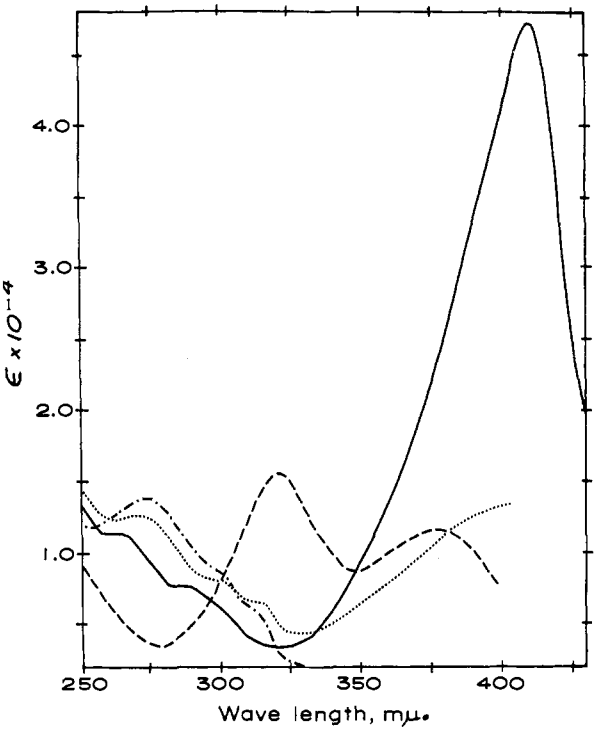
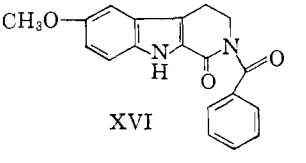


Fig. 1.—Hortiamine in acetonitrile, ———; in 95% ethanol, – – – –; in 100% ethanol, ... , dihydrohortiamine (10-methoxyevodiamine) in 100% ethanol, –·–·–·.

The spectrum of hortiamine in absolute acetonitrile (Fig. 1) corresponds to structure VII. Hortiamine is solvated by water and alcohol. The alcoholation process results in the formation of structure XIV and the spectrum in absolute ethanol is attributed to the formation, at room temperature, of an equilibrium mixture containing approximately 80% hortiamine ethanolate (XIV) and 20% hortiamine (VII). Hortiamine ethanolate (XIV) would be expected to have a spectrum similar to that of dihydrohortiamine (IX) and Fig. 1 shows that this is indeed the case.

The ultraviolet spectrum of the yellow hydrate of hortiamine is also dependent upon the nature of the solvent employed, as shown in Fig. 2. Its infrared spectrum in mineral oil mull shows carbonyl bands at 5.98 and 6.05 μ as well as NH bands at 2.95 and 3.10 μ. It is formulated as 2-(*o*-methylaminobenzoyl)-6-methoxy-1-oxo-1,2,3,4-tetrahydropyrid[3,4-b]indole (XV) and, thus, is the 6-methoxy derivative of the alkaloid rhetsinine.[17,18] The spectrum in acetonitrile corresponds predominantly to structure XV. A model compound, 2-benzoyl-6-methoxy-1-oxo-1,2,3,4-tetrahydropyrid-[3,4-b]indole (XVI), prepared through reaction of



XVI

(17) A. Chatterjee, S. Bose and C. Ghosh, *Tetrahedron*, **7**, 257 (1954).

(18) I. J. Pachter and G. Suld, *J. Org. Chem.*, **25**, 1680 (1960).

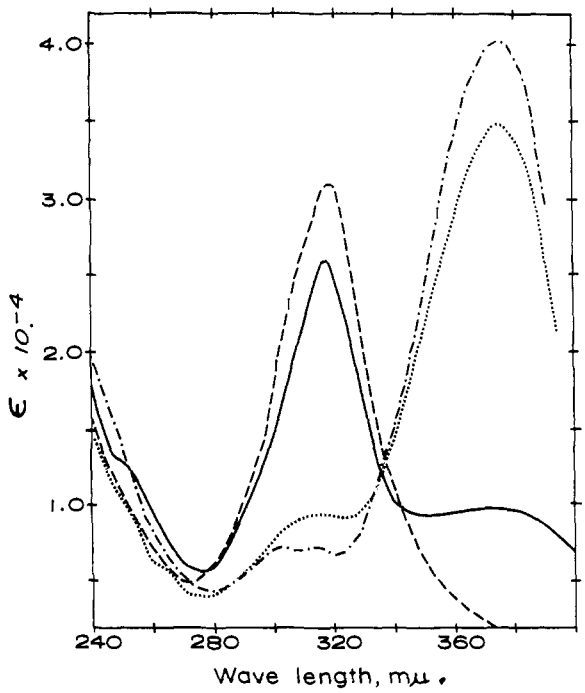


Fig. 2.—6-Methoxyrhetsinine (hortiamine hydrate) in acetonitrile, ———; in 1:1 acetonitrile–water, ...; hortiamine tartrate in acetonitrile, -·-·-·; 2-benzoyl-6-methoxy-1-oxo-1,2,3,4-tetrahydropyrid[3,4-b]indole (XVI) in acetonitrile, – – – –.

V with benzoyl chloride, is also yellow, has an ultraviolet spectrum (Fig. 2) similar to that of 6-methoxyrhetsinine and exhibits infrared bands at 3.1, 5.98 and 6.05 μ.

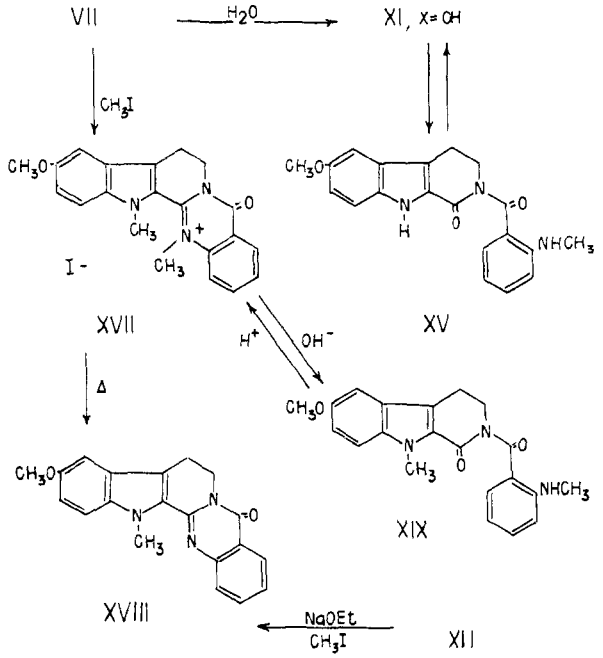
When an acetonitrile solution of 6-methoxyrhetsinine is diluted with an equal volume of water and is allowed to equilibrate for one hour, a mixture of molecules is formed in which the quaternary base XI(X = OH), related to the hortiamine salts, predominates. The relationship is illustrated in Fig. 2.

In 95% ethanol, hortiamine dissolves to produce an equilibrium mixture of solvated molecules (Fig. 1) in which the less polar structure XV predominates over the quaternary structure XI (X = OH). 6-Methoxyrhetsinine dissolves in 95% ethanol to produce an identical mixture.

The dehydration of 6-methoxyrhetsinine to produce hortiamine has not been paralleled yet by a similar dehydration of rhetsinine itself, although reddish-yellow crystals which changed to yellow[17] and a red solution which became yellow upon treatment with an aqueous solvent,[18] have been observed during work with rhetsinine.

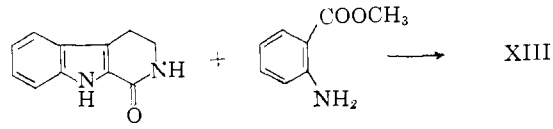
Hortiamine reacts with methyl iodide to form the quaternary salt XVII which, when heated to 270°, loses methyl iodide to form a colorless hortiamine isomer XVIII. Compound XVIII was also synthesized from XII through the action of methyl iodide and alkali.

When the methiodide XVII is treated with aqueous ammonia it is immediately transformed into yellow 6-methoxy-9-methylrhetsinine (XIX). Hortiamine salts, under the same conditions, produce



the red anhydro base VII. Compound XIX is readily recyclized by hydriodic acid to form XVII.

The synthesis of hortiamine appeared to be a simple task in view of the reported reaction of 1-oxo-1,2,3,4-tetrahydropyrid[3,4-b]indole (XX) with methyl anthranilate in phosphorus trichloride to produce rutecarpine (XIII).[19] It seemed most



XX

probable that substitution of V for XX and methyl N-methylanthranilate for methyl anthranilate in the reaction would lead to hortiamine. To our surprise, the attempted synthesis yielded only starting materials. Using the conditions already described,[19] we then attempted to repeat the synthesis of rutecarpine. Again only starting materials were obtained.

It is known that phosphorus trichloride is slowly oxidized by air into phosphorus oxychloride. It appeared possible that an equivalent of the latter is necessary for reaction. The hortiamine and rutecarpine syntheses were accordingly tried with phosphorus oxychloride present in phosphorus trichloride solvent and each was found to proceed as desired. Actually, for best yields phosphorus trichloride can be eliminated entirely. In dry toluene containing phosphorus oxychloride, the synthesis of hortiamine proceeds in 66% yield.[20] N-Methylanthranilic acid may be substituted for methyl N-methylanthranilate in the synthesis, but with a decrease in yield.

The remaining alkaloidal fraction of the plant, the tan weak bases whose yellow salts were hydro-

(19) Y. Asahina, R. H. F. Manske and R. Robinson, *J. Chem. Soc.*, 1708 (1927).

(20) The syntheses of various hortiamine-like compounds have been described by the senior author in U. S. Patent 2,866,788 (1958) and in ref. 18.

lyzed by hot water, was dissolved in benzene and subjected to chromatography on alumina. It was found to consist of two bases: rutecarpine (XIII) and a new alkaloid for which the name hortiacine is proposed.

Rutecarpine, obtained from the bark in 0.001% yield, was identified through comparison with an authentic sample.[21]

Hortiacine, m.p. 215–252°, $C_{19}H_{13}N_3O_2$, was obtained in 0.001% yield and was found to be identical with the previously derived pyrolysis product XII. The alkaloid was synthesized by a modification of the Asahina, Manske, Robinson procedure[19] using phosphorus oxychloride as the condensing agent. Hortiacine was found to undergo O-demethylation when heated with 48% hydrobromic acid to produce 10-hydroxyrutecarpine.

Experimental[22]

Isolation of Gross Crude Alkaloidal Fractions.—A 100-kg. sample of dried ground bark of *Hortia arborea* Engl. was continuously extracted with hot neutral ethanol until the marc was non-alkaloidal. The ethanolic solution was concentrated to 9 l. and stirred with 12 l. of 5% aqueous ammonia and 6 l. of chloroform until all of the material had dissolved.

The layers were separated and the ammoniacal solution was extracted with 3 additional 3-l. portions of chloroform. The combined chloroform solutions were concentrated to a volume of 3 l. and shaken with 500 ml. of 5% hydrochloric acid. A deep yellow solid separated and was removed by filtration and dried to give 37.5 g. of resinous mixed yellow hydrochlorides. The acid filtrate was reserved since it was found to contain water-soluble alkaloid salts.

After the first extraction with acid and removal of yellow solid, the chloroform solution was shaken with 4 additional 500-ml. portions of 5% hydrochloric acid. All acid solutions were combined, made basic with ammonia and extracted several times with chloroform. The chloroform solutions were dried over magnesium sulfate and evaporated to dryness to give crude alkaloids whose salts are soluble in dilute acid.

Dictamnine.—The dark colored alkaloid fraction soluble in dilute acid was boiled and triturated with portions of benzene until no additional alkaloid dissolved. The benzene extracts were combined, concentrated to 150 ml. and treated with hydrogen chloride gas. A semi-solid resin separated. The benzene layer was discarded and the residue was taken up in a small volume of boiling methanol. From the methanolic solution, there crystallized on cooling 3.0 g. of prisms. A second crystallization from methanol yielded 2.6 g. of hydrochloride.

The hydrochloride was shaken with dilute aqueous ammonia and chloroform. The chloroform layer was dried and evaporated to dryness. The crystalline residue was recrystallized twice from cyclohexane to give 1.9 g. of long colorless needles, m.p. 133–134°. A second crop of 160 mg. of prisms, m.p. 129–131°, was obtained upon concentration of the cyclohexane mother liquors. When the prisms were subjected to further purification, needles, identical with first crop material, resulted. The alkaloid was compared with an authentic specimen of dictamnine[8] and was found to be spectrally identical and to give no depression of melting point upon admixture.

Anal. Calcd. for $C_{12}H_9NO_2$: C, 72.8; H, 4.57; N, 7.08. Found: C, 72.61, 72.49; H, 4.55, 4.98; N, 7.21, 7.27.

The alkaloid formed a picrate m.p. 163–163.5° dec., when heated at the rate of 2°/min. A m.p. of 163° has been reported previously for dictamnine picrate.[7]

Nor-γ-fagarine.—After removal of crystalline dictamnine hydrochloride from the methanolic solution, the methanol was removed at room temperature and the non-crystalline residue was taken up in water. The alkaloids were liberated from their salts with ammonia, taken up in a small volume of benzene and allowed to stand at room temperature. After two weeks, rosettes of prisms began to form. Upon further standing, a mass of 4.8 g. of mixed crystalline alkaloids was obtained. The crystalline bases were heated with 50 ml. of fresh benzene. A small quantity of colorless material failed to dissolve. This was separated and recrystallized from methanol–benzene and then from ethanol to give 180 mg. of needles which begin to decompose at 258° and, when heated at a rate of 5°/min., melt at 263–265° dec. The compound was found to be identical with a sample of nor-γ-fagarine prepared from α-fagarine as described by Deulofeu and Bassi.[9]

Anal. Calcd. for $C_{12}H_9NO_3$: C, 66.97; H, 4.20; N, 6.52; OCH_3, 14.48. Found: C, 66.92; H, 5.06; N, 6.23; OCH_3, 14.39.

γ-Fagarine and Skimmianine.—The benzene solution remaining after removal of nor-γ-fagarine was chromatographed on grade F-20, 200 mesh Alcoa alumina. Two alkaloids were obtained. The first, eluted with benzene, melted to 141–142° and was found to be identical with an authentic sample of γ-fagarine[8] through a mixed melting point determination and through comparison of ultraviolet and infrared spectra. A yield of 1.7 g. was obtained.

The second alkaloid, obtained in a yield of 0.93 g., was partially eluted with benzene and more completely eluted with benzene containing 20% acetone. It melted at 175–176° and gave no depression of melting point upon admixture with an authentic specimen of skimmianine.[8] Its spectra were found to be identical with those of authentic skimmianine.

In addition to the separated alkaloids, an additional gram of mixed crystals was also obtained from the chromatogram. It was shown, by paper chromatography, to consist of γ-fagarine and skimmianine.

Separation of Yellow Salts into Weak and Strong Bases.—The 37.5 g. of dried crude yellow hydrochloride was stirred with hot ethanol to remove resinous material. The ethanolic slurry was cooled and the yellow solid filtered. Concentration of the dark ethanolic liquors yielded additional yellow product. The combined dried yellow solids weighed 31 g. This material was boiled with 2.5 l. of water until all of the deep yellow colored material had dissolved. A tan solid remained insoluble. The mixture was filtered while hot and the tan crude weak bases were dried to give 4.1 g. of product.

The hot aqueous filtrate was cooled and 20 g. of bright yellow salt crystallized. A second crop of 4 g. was obtained from the aqueous mother liquors.

Hortiamine and 6-Methoxyrhetsinine.—A 16.5-g. portion of the 24 g. of bright yellow hydrochloride, which crystallized from water after removal of the weakly basic alkaloids, was ground to a powder and shaken with 500 ml. of chloroform and 200 ml. of 10% aqueous ammonia. The layers were separated and the ammoniacal solution was shaken four times with 25-ml. portions of chloroform. The combined deeply colored chloroform solutions were concentrated at atmospheric pressure and diluted with 800 ml. of boiling benzene. Upon cooling, 4.2 g. of beautiful red-orange needles of hortiamine crystallized, m.p. 209° dec. The alkaloid was crystallized once again from benzene, but the melting point remained unchanged. Its ultraviolet spectrum in absolute acetonitrile shows λ_{max} 411, ϵ 47,300.

Anal. Calcd. for $C_{20}H_{17}N_3O_2$: mol. wt., 331.35; C, 72.49; H, 5.17; N, 12.68, 1 OCH_3, 9.37. Found: mol. wt. (by non-aqueous titration), 329; C, 72.11, 72.30; H, 5.29, 5.35; N, 12.79; OCH_3, 9.57; 9.83.

After removal of the red-orange crystals, the benzene–chloroform mother liquors were placed in a refrigerator overnight. Yellow prisms of 6-methoxyrhetsinine separated from solution. These were removed and the solution was concentrated to a small volume and allowed to stand again. An additional yield of yellow prisms was obtained. The prisms were combined and boiled continuously in benzene until they dissolved to give a deep red solution. The solution was concentrated to a small volume at atmospheric pressure and 9.5 g. of separated red-orange needles of hortiamine were collected to give a total yield of 13.7 g. from 16.5 g. of salt. This corresponds to a total yield from the bark of 0.020% of hortiamine.

A sample of hortiamine was dissolved in boiling benzene and allowed to stand. Yellow prisms of 6-methoxyrhetsinine

(21) We are grateful to Dr. Léo Marion of the National Research Council, Ottawa, Canada, for providing a sample of rutecarpine when we were unable to repeat the synthesis described in ref. 19.

(22) We are grateful to Dr. Walter E. Thompson and to Mrs. Doris Ralston of these laboratories for spectral and analytical data.

separated. The yellow product, upon heating, turns orange and then red and has the same melting point as hortiamine. Its ultraviolet spectrum in absolute acetonitrile shows λ_{max} 318, ϵ 26000, and λ_{max} 375, ϵ 9700. By analysis, the yellow prisms contain one mole of water more than hortiamine.

Anal. Calcd. for $C_{20}H_{19}N_3O_3$: C, 68.75; H, 5.48; N, 12.03. Found: C, 68.90; H, 5.53; N, 12.07, 11.91.

Hortiamine hydrochloride is formed when hydrogen chloride is bubbled into a benzene solution of hortiamine. Upon recrystallization from aqueous ethanol, the salt melts at 243° dec. Analysis suggests that the salt is hydrated. Its ultraviolet spectrum in acetonitrile shows λ_{max} 376, ϵ 40,300.

Anal. Calcd. for $C_{20}H_{17}N_3O_2 \cdot H_2O \cdot HCl$: C, 62.25; H, 5.23; N, 10.89; Cl, 9.19. Found: C, 61.95, 61.87; H, 5.61, 5.23; N, 11.02, 11.17; Cl, 9.21, 9.29.

O-Demethylation of Hortiamine.—To 5.0 g. of hortiamine was added 100 ml. of 48% hydrobromic acid. The mixture was heated under reflux for 8 hours. On cooling and dilution with an equal volume of water, a fine yellow solid separated. Upon filtration and recrystallization from aqueous ethanol containing a few drops of hydrobromic acid, there was obtained 5.0 g. (83.5% yield) of yellow needles, m.p. 266–267° dec. The product dissolves readily in dilute sodium hydroxide to give an intense magenta-colored solution which soon becomes pale yellow in color.

Anal. Calcd. for $C_{19}H_{15}N_3O_2 \cdot HBr$: C, 57.35; H, 4.05; N, 10.55. Found: C, 57.42; H, 4.30; N, 10.67.

Conversion of Hortiamine into Hortiacine.—A 200 - mg. sample of hortiamine hydrochloride hydrate was placed in a sublimation apparatus and heated at 230–270° at 0.5 mm. until no additional material sublimed. The sublimate was dissolved in ethyl acetate and filtered to remove some yellow insoluble matter. The solution was concentrated, diluted with cyclohexane, concentrated again and cooled to yield 125 mg. (69% yield) of hortiacine, m.p. 250°. The product was recrystallized from ethanol and long flat needles, m.p. 251.2–252° were obtained. There was no depression of melting point upon admixture with hortiacine derived from the plant.

Hydrogenation of Hortiamine. Preparation of 10-Methoxyevodiamine (IX).—To 5.0 g. of hortiamine was added 50 ml. of glacial acetic acid and 0.07 g. of platinum oxide. The mixture was shaken under 30 p.s.i. of hydrogen for 1 hour. Methoxyevodiamine crystallized from solution during the reduction. Filtration and recrystallization of the product from ethanol–chloroform yielded 4.0 g. of colorless needles, m.p. 249–251°.

Anal. Calcd. for $C_{20}H_{19}N_3O_2$: C, 72.05; H, 5.74. Found: C, 71.93, 72.18; H, 5.87, 6.03.

5-Methoxyisoevodiamine Chloride Hydrate.—A 5.0-g. sample of 10-methoxyevodiamine was heated under reflux for 2 hours with 100 ml. of 95% ethanol containing 5% hydrogen chloride. The solid dissolved and 5.2 g. of the product crystallized from solution. It was collected and recrystallized twice from water to give light yellow prisms, m.p. 238–239°.

Anal. Calcd. for $C_{20}H_{21}N_3O_3 \cdot HCl$: C, 61.93; H, 5.72. Found: C, 62.05; H, 5.57.

Alkaline Cleavage of Hortiamine.—To a solution of 3.0 g. of potassium hydroxide in 75 ml. of ethanol was added 1.65 g. of hortiamine. The mixture was heated under reflux for 2 hours and then concentrated under reduced pressure to a volume of 8 ml. The residue was diluted with 30 ml. of water and a crystalline solid separated. This was filtered and dried to give 765 mg. of prisms. Upon recrystallization from ethanol the compound melted at 281–282°. It was found to be identical with a synthetic specimen of **6-methoxy-1,2,3,4-tetrahydropyrid[3,4-b]indole**[14] through mixed melting point determination and comparisons of infrared and ultraviolet spectra.

Anal. Calcd. for $C_{12}H_{12}N_2O_2$: C, 66.65; H, 5.59; N, 12.96; OCH_3, 14.36. Found: C, 66.56; H, 5.79; N, 12.90; OCH_3, 14.35.

The basic filtrate was saturated with carbon dioxide and the resulting solution allowed to stand overnight. A small quantity of bright yellow needles, which soften at 165° and melt with decomposition and gas evolution at 168–169°, separated. The nature of this substance has not been determined.

After removal of the yellow substance, the filtrate was acidified with acetic acid and cooled in the refrigerator. A crystalline substance was obtained which was dried and sublimed at 100°(2 mm.). The sublimate was recrystallized from aqueous ethanol to give 195 mg. of plates of **N-methylanthranilic acid**, m.p. 177–179°. The acid was identified through mixed melting point and spectral comparisons with an authentic sample.

2-Benzoyl-6-methoxy-1-oxo-1,2,3,4-tetrahydropyrid[3,4-b]indole (XVI).—A mixture of 1.0 g. of 6-methoxy-1-oxo-1,2,3,4-tetrahydropyrid[3,4-b]indole (V) and 10 ml. of benzoyl chloride was heated under reflux for 2 hours. Excess benzoyl chloride was removed and the residue was poured into aqueous sodium carbonate solution. The resulting precipitate was recrystallized from ethyl acetate to give 0.8 g. of bright yellow prisms, m.p. 284–285.5°. The ultraviolet spectrum in acetonitrile showed λ_{max} 319, ϵ 31,000.

Anal. Calcd. for $C_{19}H_{16}N_2O_3$: C, 71.24; H, 5.03. Found: C, 71.13; H, 5.25.

Synthesis of Hortiamine.—To 9 l. of hot dry toluene was added 100 g. of finely powdered 6-methoxy-1-oxo-1,2,3,4-tetrahydropyrid[3,4-b]indole. The mixture was boiled with stirring and traces of water were azeotropically removed. After 30 minutes of heating under reflux to get as much of the starting compound into solution as possible, heating was suspended and 75 ml. of freshly distilled phosphorus oxychloride was added in one portion. Stirring without heating was maintained for 15 minutes, after which 125 ml. of methyl N-methylanthranilate was added in one portion. Heating with stirring was resumed and maintained for 3 hours. The mixture was allowed to stand overnight and the yellow product was collected by filtration. Without drying to remove the last traces of toluene, the product was recrystallized from 10 l. of boiling water. The bright yellow hydrochloride, on filtration and drying, weighed 128 g.

The yellow salt was ground to a powder and stirred with 1.5 l. of chloroform to form a slurry. The slurry was shaken vigorously with 100 ml. of concentrated aqueous ammonia. The chloroform solution, which had become intensely colored, was separated and, without delay, was concentrated by boiling off solvent at atmospheric pressure. When the solution was concentrated to about 800 ml., hot benzene was added and boiling started again. The solution was filtered at this point and then concentrated until most of the chloroform had been boiled off. Red-orange needles separated from solution during the concentration process. These were collected to give 64 g. of hortiamine, m.p. 208–210° dec., identical by mixed melting point and by comparisons of ultraviolet and infrared spectra with natural hortiamine.

Further boiling of the chloroform–benzene solution eventually yielded 36 g. of second crop hortiamine as red needles. The combined first and second crops represent a 66% yield of synthetic hortiamine.

Modified Asahina, Manske, Robinson Synthesis[19] **of Rutecarpine.**—A 5.0-g. sample of powdered 1-oxo-1,2,3,4-tetrahydropyrid[3,4-b]indole was heated under reflux with stirring in 150 ml. of dry toluene until the solid dissolved. Heating was discontinued and 5.0 g. of freshly distilled phosphorus oxychloride was added. The mixture was heated under reflux for 10 minutes and then 7.5 g. of methyl anthranilate was added. Heating under reflux was maintained for 2 hours while a bright yellow solid was formed. The mixture was cooled and the solid collected by filtration. The yellow solid was stirred with ammoniacal ice-water and the resulting crude rutecarpine was collected, dried and recrystallized from acetic acid–ethyl acetate three times. The product weighed 2.95 g. and melted at 257.5 to 258.5°. It did not depress the melting point of rutecarpine derived from *Hortia arborea*.

Anal. Calcd. for $C_{18}H_{13}N_3O$: C, 75.24; H, 4.56; N, 14.63. Found: C, 75.09; H, 4.52; N, 14.58.

Hortiamine Methiodide and 6-Methoxy-9-methylretsinine.—To 2.0 g. of hortiamine in 20 ml. of boiling chloroform was added 50 ml. of benzene. The deeply colored solution was heated under reflux for 5 minutes and 5 ml. of methyl iodide was added in one portion. Heating under reflux was maintained for 1 hour. The solution became pale orange in color and yellow prisms separated from solution. The mixture was cooled and filtered to give 2.0 g. of bright yellow crystals which, upon recrystallization from 70% aqueous ethanol, gave 1.8 g. of salt, m.p. 208–209°

dec. This material gives an orange-red color when shaken with aqueous ammonia and chloroform, indicating that it contains hortiamine hydroiodide in addition to hortiamine methiodide. This indication was confirmed through pyrolysis, described below.

The chloroform–benzene mother liquor, remaining after removal of the yellow salts, was evaporated to dryness and the residue was dissolved in ethanol. Upon concentration of the ethanolic solution, there was obtained 0.7 g. of light yellow crystalline product. The product, after three recrystallizations from ethanol, gave 0.45 g. of yellow prisms of 6-methoxy-9-methylrhetsinine, m.p. 168–169°.

Anal. Calcd. for $C_{21}H_{21}N_3O_3$: C, 69.40; H, 5.83; N, 11.56. Found: C, 69.18; H, 5.96; N, 11.59, 11.69.

A sample of 6-methoxy-9-methylrhetsinine was dissolved in boiling ethanol and treated with a few drops of hydriodic acid. Hortiamine methiodide crystallized from solution in almost quantitative yield. It was recrystallized once from 70% aqueous ethanol and melted at 209° with evolution of gas. This product gives no orange-red color when shaken with aqueous ammonia and chloroform.

Anal. Calcd. for $C_{21}H_{20}IN_3O_2$: C, 53.29; H, 4.26; N, 8.88. Found: C, 52.88; H, 4.45; N, 8.84, 8.89.

The reaction of 6-methoxy-9-methylrhetsinine with hydriodic acid to form hortiamine methiodide is reversible, for when the latter is treated with aqueous ammonia, the former is quantitatively regenerated.

Pyrolysis of Hortiamine Methiodide. 13-Methylhortiacine.—A 100-mg. sample of hortiamine methiodide was heated in a sublimation apparatus to 270°(1 mm.). The sample melted with gas evolution and then distilled. The solid distillate was dissolved in ethyl acetate. The solution, upon concentration, yielded 64 mg. (91% yield) of fine, long, almost colorless needles, m.p. 195–196°. The product was spectrally identical and gave no depression of melting point with a sample of 13-methylhortiacine prepared from hortiacine through methylation with sodium ethoxide and methyl iodide as described below.

Anal. Calcd. for $C_{20}H_{17}N_3O_2$: C, 72.49; H, 5.17. Found: C, 72.24; H, 5.43.

A 1.0-g. sample of the yellow crystals which separated from solution during the reaction of hortiamine with methyl iodide was heated to 300°(1 mm.) in a sublimation apparatus. The material melted with evolution of gas and distilled. The solid distillate was dissolved in boiling ethyl acetate. Upon cooling, there was obtained 0.25 g. of small needles, m.p. 250°, identical with hortiacine. Concentration of the ethyl acetate mother liquor yielded 0.4 g. of long needles, m.p. 188–190°. Upon recrystallization from ethyl acetate, the melting point was raised to 192–193° and the melting point was not depressed upon admixture with 13-methylhortiacine derived through methylation of hortiacine.

13-Methylhortiacine from Hortiacine.[23]—To a solution of 150 mg. of sodium in 50 ml. of absolute ethanol was added 1.0 g. of hortiacine suspended in 50 ml. of absolute ethanol. The suspension was heated under reflux on a steam-bath and 30 ml. of dimethylformamide was added to aid solution. The solution became bright yellow in color. A 0.4-ml. portion of methyl iodide in 2 ml. of ethanol was added. The color faded rapidly. Alternate additions of 150-mg. portions of sodium and 0.4-ml. portions of methyl iodide were made several times. The mixture was then poured into water and the precipitated solid filtered and dried. The solid was heated with ethyl acetate, filtered to remove less soluble unchanged hortiacine and concentrated. There was obtained 0.61 g. of needles, m.p. 179–185°. After several recrystallizations this material yielded 0.43 g. of pure 13-methylhortiacine, m.p. 193–194°.

Isolation of Rutecarpine and Hortiacine.—The 4.1 g. of tan solid that remained when the precipitated yellow salts were boiled with water was dissolved in benzene and passed through a short column of alumina to remove colored impurities. Elution with benzene containing 5% acetone yielded 2.5 g. of cream-colored solid which crystallized from acetone to give short needles, m.p. 238–241°. A chloroform solution of the material gave a deep yellow precipitate when shaken with 5% hydrochloric acid. The precipitate was readily hydrolyzed when boiled with water to regenerate the cream-colored solid. Paper chromatography showed that the solid was not homogeneous but consisted of two principal alkaloids.

A 1.0-g. sample of the cream-colored solid was dissolved in 500 ml. of benzene and passed into a benzene-wet column of grade F-20, 200 mesh Alcoa alumina which was packed 56 cm. high in a column 3.5 cm. in diameter. The alkaloids are strongly fluorescent and progress of the materials through the column was followed with ultraviolet light. The column was washed continously with benzene containing 5% acetone. Four bands developed. The first consisted of a trace of rapidly moving weakly fluorescent material. The second band exhibited a strong bluish fluorescence. The third band exhibited strong fluorescence of a yellowish cast. The fourth band, consisting of material strongly adsorbed on the top of the column, was not eluted.

The collection of fractions was begun when the band of bluish fluorescence reached the bottom of the column. It was eluted with 22 500-ml. portions of benzene containing 5% acetone. The first 8 portions yielded only traces of alkaloid. Portions 9 through 22 each yielded an alkaloid of m.p. 259–260°. The alkaloidal fractions were combined and recrystallized from ethyl acetate to give 255 mg. of fine, long, colorless needles, m.p. 259.5–260°. The melting point of the compound was not depressed upon admixture with an authentic sample of rutecarpine and its ultraviolet and infrared spectra were found to be identical with those of authentic rutecarpine.

Anal. Calcd. for $C_{18}H_{13}N_3O$: C, 75.24; H, 4.56; N, 14.63. Found: C, 75.07; H, 4.68; N, 14.52.

The band of yellowish fluorescence began to emerge in the 23rd 500-ml. portion of benzene containing 5% acetone. The 72 mg. of alkaloidal material of portion 23 was found to be a mixture, however. Portion 24 yielded 60 mg. of a new alkaloid, m.p. 251–252°. The eluting solvent was changed to benzene containing 15% acetone and 500-ml. portions, 25 through 30, each yielded pale yellow needles, m.p. 215–252°. The alkaloidal fractions 24 through 30 were combined and recrystallized from ethyl acetate to yield 265 mg. of pale yellow needles, m.p. 252–252.5°. The alkaloid was found to analyze for a substance of formula $C_{19}H_{15}N_3O_2$ containing one methoxyl group. It was named hortiacine.

Anal. Calcd. for $C_{19}H_{15}N_3O_2$: C, 71.91; H, 4.76; OCH_3, 9.78. Found: C, 71.63; H, 4.96; OCH_3, 10.00.

Portions 31 through 38 also yielded hortiacine, but of lesser purity. The alkaloid from portion 31 melted at 247–249°. The alkaloid from portion 38 melted at 233–236°, but the melting point was raised upon admixture with hortiacine.

The solvent was changed to pure acetone but little additional alkaloid could be eluted. Portions 39 and 40 yielded less than 20 mg. of residual solid.

Synthesis of Hortiacine.—To 27.0 g. of 6-methoxy-1-oxo-1,2,3,4-tetrahydropyrid[3,4-b]indole in 2.5 l. of dry, boiling toluene was added 32 g. of phosphorus oxychloride. The reaction mixture was stirred for 10 minutes and 32 ml. of methyl anthranilate was added. Heating under reflux was continued for 3 hours. The toluene layer was then decanted and the residue was treated with cold water to decompose excess phosphorus oxychloride. The remaining solid was thoroughly triturated with aqueous ammonia, dried and recrystallized from acetic acid-ethyl acetate to give 18.4 g. of fine needles, m.p. 250°. The product gave no depression of melting point upon admixture with natural hortiacine.

Demethylation of Hortiacine. 10-Hydroxyrutecarpine.—To 4.5 g. of hortiacine was added 45 ml. of 48% hydrobromic acid and the mixture was heated under reflux for 17 hours. It was then cooled, filtered and the yellow solid stirred with dilute sodium hydroxide solution. Nonphenolic materials were removed by filtration and the clear basic solution was acidified with acetic acid. A gray solid separated and was recrystallized twice from isopropyl alcohol to give 2.3 g. of 10-hydroxyrutecarpine, m.p. 286–288°.

Anal. Calcd. for $C_{18}H_{13}N_3O_2$: C, 71.27; H, 4.32. Found: C, 70.92; H, 4.47.

(23) This synthesis was performed by Miss Elinor Fisher of these laboratories.