137. *Curare Alkaloids. Part V. Alkaloids of some* Chondrodendron *Species and the Origin of* Radix Pareiræ Bravæ.

By Harold King.

The aim of this investigation was the determination of the botanical source of the substance known in pharmacy as *radix pareiræ bravæ*, since its alkaloids are related to the phenolic alkaloids of tube- and pot-curare. This object has been attained. When *pareira brava* yields *l*-bebeerine it comes from *Chondrodendron platyphyllum* and when it yields *d*-bebeerine from *Ch. microphyllum*. These two species are very similar in taxonomical and pharmacognostical characters. In addition *Ch. candicans* from British Guiana has been examined. All these species contain bebeerine (*d*- or *l*-) and *d-iso*chondrodendrine in widely varying proportions. A study of the properties of *d-iso*chondrodendrine, which is now available in quantity, has enabled probable structures to be assigned to protocuridine and *neo*protocuridine, isomeric phenolic alkaloids of pot-curare. From the leaves of *Ch. platyphyllum* a new alkaloid, *chondrofoline*, has been obtained. It is related to bebeerine and a probable structure is assigned. From a relatively large amount of *radix pareiræ bravæ* a new alkaloid, iso*coclaurine*, isomeric with coclaurine has been isolated and its structure determined. A classification of certain bisbenzyl*iso*quinoline alkaloids is given.

The phenolic alkaloids of tube- and pot-curare are derived from plants of the N.O. *Menispermaceæ* and in all probability from the genus *Chondrodendron*. The drug *radix pareiræ bravæ* bought on the English market has been shown to contain *d*-bebeerine, which is enantiomorphous with an alkaloid occurring in tube-curare and related to its active principle *d*-tubocurarine chloride. This relationship raises once more the question of the botanical origin of *pareira brava*, a subject which has long been a matter of dispute.

In 1694, Pomet, in his "Histoire Générale des Drogues," p. 69, avers that the Portuguese first brought *pareira brava* * from Mexico, the Indies and Brazil. Its botanical origin was unknown but the drug had a considerable reputation in the treatment of calculus and other bladder complaints. In 1648, Piso (*De Facultatibus simplicium*, **4**, 94) and Marcgrav (*Hist. Plantarum*, **1**, 25) both described and figured a plant with a wide distribution in the New World known as *Caapeba*, which also had a reputation in the treatment of those ailments for which *pareira brava* was used. An element of confusion thus arose which was

* The spelling *pareira* instead of the Portuguese *parreira*, meaning a vine, is probably the French or English rendering of the sound of the Portuguese word.

consolidated by the weight of authority of Linnaeus, who in 1753 gave the name *Cissampelos pareira* to *caapeba*, thus perpetuating for over a century the erroneous idea that *pareira brava* originated from *Cissampelos pareira*. During the past hundred years there has been a natural phase of chemical development, but the confusion which existed has not been resolved and in some respects has been made worse. Three of the contributory causes have been the adulteration of the market by roots purporting to be *pareira brava*, the lack of authoritative botanical identification of the materials examined, and the uncritical pronouncements of pharmacists of repute.

Hanbury (*Pharm. J.*, 1873, **4**, 81) made a bold attempt to solve the problem by securing specimens of the drug from Rio de Janeiro, Brazil, together with herbarium specimens, and came to the conclusion that *pareira brava* came from *Chondrodendron tomentosum* R. and P., a species which, as is now known, is not recorded for Brazil. In the monograph on the *Menispermaceæ* in Engler's *Pflanzenreich* (1910, **4**, 41) Diels mentioned these herbarium specimens but clearly favoured the view that *pareira brava* came from a Brazilian *Chondrodendron* and probably *Ch. platyphyllum* (St. Hil.) Miers. From a perusal of Hanbury's account of his researches into this question it seems probable that he regarded these two species as identical and unfortunately chose the older name *tomentosum*. More recently Krukhoff and Moldenke (*Brittonia*, 1938, **3**, 1) have critically surveyed the American *Menispermaceæ* and concluded that *Ch. platyphyllum* is the major source of the drug, but they do not exclude the possibility that an additional source might be *Ch. microphyllum* (Eichl.) Moldenke, also a native of southern Brazil.

The reliable modern chemistry of *pareira brava* may be said to start with the work of Scholtz (*Ber.*, 1896, **29**, 2054), who examined a commercial powder called *Bebeerinum purum* and isolated therefrom for the first time a crystalline lævo-bebirine, $[\alpha]_D$ − 298°. In 1899 Scholtz (*Arch. Pharm.*, **237**, 199) also isolated bebirine, now spelt bebeerine, from *radix pareiræ bravæ*. In 1906, however, he (*ibid.*, **244**, 555) obtained the alkaloid in the dextro-form, $[\alpha]_D$ + 297°, from pareira root and also a fraction claimed to be racemic bebeerine and the same constituents from the *Bebeerinum purum* of Merck. At a later date Scholtz and Koch (*ibid.*, 1914, **252**, 513) also examined a large quantity of *radix pareiræ bravæ* from a different commercial source and only found traces of bebeerine. As a result of these recorded variations and of their own experiences, Faltis and Neumann (*Monatsh.*, 1921, **42**, 325) came to the conclusion that, unless roots from closely allied species exist which cannot be differentiated by the pharmacognosist, then it may be that *Ch. platyphyllum* is subject to climatic and seasonal influences which determine the nature of the alkaloid.

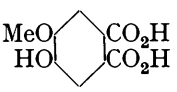
In the following pages an account is given of the alkaloids of *radix pareiræ bravæ* of the English market, of botanically identified *Ch. platyphyllum* collected in Brazil in the region of Rio de Janeiro and from Bahia, a thousand miles further north, and of *Ch. microphyllum*, also collected near Bahia. An account of the alkaloids of *Ch. candicans* (Rich *ex* D.C.) Sandwith, the only *Chondrodendron* of British Guiana, is also included, since it is said to be used by the Warrau Indians in some of their poisons. The root of *Ch. platyphyllum* from Rio contained *l*-bebeerine and *d-iso*chondrodendrine and the same alkaloids were found in the root of this species collected near Bahia. In the stems, however, from Bahia, only *l*-bebeerine was found but the leaves contained *l*-bebeerine, *d-iso*chondrodendrine and a new alkaloid to which the name l-*chondrofoline* has been given. On the other hand *Ch. microphyllum* from Bahia contained the optical enantiomorph *d*-bebeerine and *d-iso*chondrodendrine. *Radix pareiræ bravæ* of the English market contained *d*-bebeerine, *d-iso*chondrodendrine and in small amount a new alkaloid, l-iso*coclaurine*. The results are summarised in the following table, the more abundant alkaloid being placed first.

Species.	Locality.	Organ.	Alkaloids.
Ch. platyphyllum	Rio	Root	*d-iso*Chondrodendrine, *l*-bebeerine.
,, ,,	Bahia	Root	*l*-Bebeerine, *d-iso*chondrodendrine.
		Stems	*l*-Bebeerine.
		Leaves	*l*-Chondrofoline, *d-iso*chondrodendrine, *l*-bebeerine.
Ch. microphyllum	Bahia	Root	*d-iso*Chondrodendrine, *d*-bebeerine.
Ch. candicans	British Guiana	Stem	*d-iso*Chondrodendrine, *d*-bebeerine.
Pareira brava			*d*-Bebeerine, *d-iso*chondrodendrine, *l-iso*coclaurine.

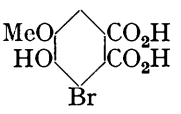
Chondrodendron platyphyllum and *microphyllum* are two species whose taxonomical characters are very close, but chemical examination shows a clear distinction. Since it is unlikely that native plant collectors could distinguish these species and there is no apparent difference in the pharmacognostical characters of their roots, it seems clear that *pareira brava* has originated in the past in these two species. The findings of Scholtz and Faltis thus receive a simple and adequate explanation. The occasional occurrence of racemic bebeerine or bebeerine of low rotation, if the observations of various authors bear the interpretation given to them, might be due to admixture of the two species.

A survey of the table shows that the relative proportions of the alkaloids in any one species may show considerable variation. This is brought out more forcibly by the quantitative figures given in the experimental section. The factors which bring about this variation are probably climatic and seasonal and there are also the inherent characters of the plants. *Radix pareiræ bravæ* of the English market contained large amounts of *d*-bebeerine but very little *d-iso*chondrodendrine. On the evidence available it must have come from *Ch. microphyllum*. The remote possibility must however be envisaged that there are two species included under the designation of *Ch. platyphyllum*, one giving *d*-bebeerine and the other *l*-bebeerine, which are indistinguishable so far taxonomically but distinguishable chemically. There is some support for this, since the physical properties of the percolate from *Ch. microphyllum* seemed to be strikingly different from those of *pareira brava*.

It was difficult at first to be sure of the identity of the *iso*chondrodendrine, since the possibilities of isomerism in this group of alkaloids are numerous, and since this alkaloid has not been well characterised in spite of the fact that its general structure is known; in addition the melting points of *iso*chondrodendrine and derivatives are nearly all above 300°. The fact that *iso*chondrodendrine gave a typical Millon reaction was also disconcerting, since the chemical structures on which this reaction depend and which were discussed in Part III (J., 1937, 1481) seemed to preclude a positive reaction with *iso*chondrodendrine. However, it was found and recorded in Part IV (J., 1939, 1163) that, although *O*-methylnor-*m*-hemipinic acid (I) does not give a Millon reaction, its bromo-derivative (II) does.



(I.)



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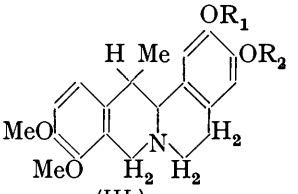
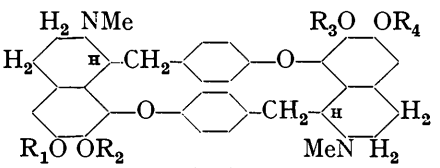


(III.)

Furthermore neither corybulbine (III; R_1 = Me, R_2 = H) nor *iso*corybulbine (III; R_1 = H, R_2 = Me) gives the reaction. It follows that *iso*chondrodendrine, which has two methoxy-groups and two phenolic groups, should have at least one phenolic group adjacent to the ether oxygen atoms (IV, R_2 or R_3 or both = H). The isomeric alkaloid *neo*protocuridine discovered in pot-curare (King, J., 1937, 1472) was optically inactive and gave no Millon reaction but on complete methylation and degradation gave a methine methiodide indistinguishable from inactive α-methyl*iso*chondrodendrinemethine methiodide obtained from *d-iso*chondrodendrine. Its constitution is therefore to be represented by a centro-



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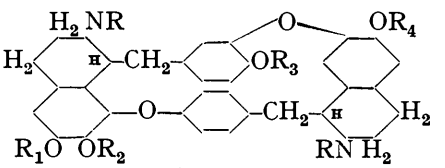


(V.)

symmetric form of (IV) in which R_1 and R_4 are H and R_2 and R_3 are Me. The availability of *iso*chondrodendrine as the main alkaloidal constituent of some of these chondrodendron

species has facilitated its characterisation and it has been found that its methylation product, *O*-methyl*iso*chondrodendrine methiodide, is in all probability identical with the methylation product of the other main phenolic alkaloid of pot-curare, protocuridine. The amount of *O*-methylprotocuridine methiodide available for comparison was small, but a close study of its properties along with those of *O*-methyl*iso*chondrodendrine methiodide failed to reveal any differences. The parent alkaloids *iso*chondrodendrine and protocuridine are, however, quite different but isomeric. Since both parent alkaloids gave the Millon reaction and both were optically active, they must be represented by (IV), in which neither has a catechol arrangement of phenolic groups and at least one phenolic group in both alkaloids must be at OR_2. In one of these alkaloids OR_2 and OR_3 will each be represented by OH and in the other only one of these can be OH.

The new alkaloid *chondrofoline*, which has only been encountered in the leaves of *Ch. platyphyllum*, has the formula $C_{33}H_{36}O_6N_2$, is phenolic and contains three methoxyl groups. On complete methylation it gave an amorphous *O*-methyl methiodide and methochloride closely resembling the corresponding products from bebeerine, and on degradation by a one-stage Hofmann reaction it gave crystalline *O*-methylchondrofolinemethine methiodide, which proved to be identical with the inactive *O*-methylbebeerinemethine methiodide B obtained in an analogous degradation of *d*-bebeerine or *d*-tubocurarine (King, J., 1935, 1381). A second crystalline product was obtained, *l*-*O*-methylchondrofolinemethine methiodide, which proved to be the enantiomorph of *d*-*O*-methylbebeerinemethine methiodide, also obtained both from *d*-bebeerine and from *d*-tubocurarine. Since chondrofoline does not give a Millon reaction and it only contains one phenolic group, its constitution is shown by (V) in which R = H, and the phenolic group is at OR_1 or OR_4.

A number of bisbenzyl*iso*quinoline alkaloids can now be classified as belonging to a bebeerine or an *iso*chondrodendrine type.

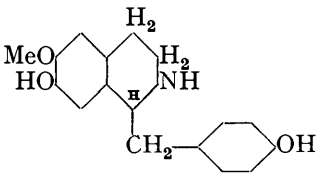
iso*Chondrodendrine Type* (IV).		*Bebeerine Type* (V).	
Sub-type a	*d-iso*chondrodendrine *O*-methyl*iso*chondrodendrine * *d*-protocuridine	Sub-type c	*d*-bebeerine *l*-bebeerine (*l*-curine) *l*-chondrofoline
Sub-type b	*i-neo*protocuridine	Sub-type d	*d*-tubocurarine

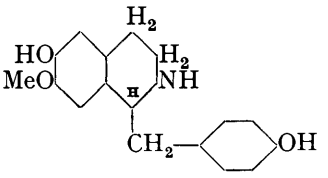
* Kondo, Tomita, and Uyeo, *Ber.*, 1937, **70**, 1890.

Completely methylated structures corresponding to the *iso*chondrodendrine type will occur in two sub-types a and b depending on whether the two centres of asymmetry are the same or opposite in sign (centro-symmetry). Examples of both sub-types are known. Similarly the bebeerine type on complete methylation will give rise to two sub-types c and d, both optically active, in which the two centres of asymmetry are the same or opposite in sign. In this case also examples of both sub-types are known.

*d-iso*Coclaurine found to a small extent in *radix pareiræ bravæ* has the formula $C_{17}H_{19}O_3N$, is phenolic and contains one methoxyl group. It is isomeric with coclaurine, an alkaloid of the Menisperm, *Cocculus laurifolius* D.C. On complete methylation it gave d-O-*dimethyl*-N-*methylcoclaurine methiodide*, which proved to be the optical enantiomorph of the corresponding product obtained by complete methylation of natural *l*-coclaurine (VI). Since *iso*coclaurine does not give the catechol reaction and since it is an isomeride of coclaurine, its constitution must be that shown by (VII).

(VI.)





(VII.)

I am indebted to Mr. N. Y. Sandwith, M.A., of the Herbarium, Kew, for much enlightenment on the botanical aspects of this communication.

Experimental.

Alkaloids of Chondrodendron platyphyllum *from Rio.*—Two pounds of the root of this plant together with the leaves were kindly presented to the writer by Sr. Carlos da Silva Araujo of Rio de Janeiro. It was identified locally as *Ch. platyphyllum* and this was confirmed by Mr. N. Y. Sandwith of the Herbarium, Kew. It is known locally as " abútua grande " or large abutua and rarely under the name " parreira brava " (reiz).

The powdered root (924 g.) was percolated with 1% tartaric acid solution until the alkaloid was exhausted. The solution was then concentrated to 1460 c.c. at 40—45°. A 10 c.c. sample was freed from fat, made alkaline with sodium bicarbonate, and extracted thrice with chloroform. Removal of the chloroform left 0·57 g. of crude alkaloids, corresponding to a yield of about 9%. When this was moistened with a little methyl alcohol, it gave a microcrystalline powder (0·3 g.), m. p. 310°, $[\alpha]_{5461}$ + 158° (for the base in N/10-hydrochloric acid), corresponding to a 4·7% yield of readily crystallisable bases.

The remaining 1450 c.c. of extract were cooled to 0° and to the solution solid sodium bicarbonate was added in excess. The precipitated solid was collected, and the filtrate extracted repeatedly with chloroform, emulsions being circumvented by filtration through a thin layer of kieselguhr. The chloroform was removed by distillation; the syrupy residue on dilution with a little methyl alcohol crystallised instantly (yield, 28·5 g.). The initial solid precipitated by sodium bicarbonate was extracted by boiling with chloroform and on removal of the solvent gave 3·3 g. of crystalline powder on moistening with methyl alcohol. The chloroform-extracted solid was finely powdered and extracted with ether (Soxhlet) for 14 days; a further 1·1 g. of crystalline alkaloid separated when the residue, on removal of the ether, was moistened with methyl alcohol.

The various methyl-alcoholic mother-liquors (A) were combined and kept. The total microcrystalline alkaloidal bases (32·9 g.) had $[\alpha]_{5461}$ + 158·7° in N/10-hydrochloric acid solution. They consisted mainly of *d-iso*chondrodendrine, a little *l*-bebeerine, and some more complex amorphous basic material, probably of high positive rotation, which was carried along with the crystalline bases. This was demonstrated in the following way. The crystalline bases were neutralised with warm N-sulphuric acid (106 c.c.); the solution on cooling to 0° gave a copious yield (39·8 g.) of *iso*chondrodendrine sulphate, crystallising in double pyramids containing 28% of water of crystallisation. On careful concentration of the mother-liquor a further 3·3 g. of the same alkaloidal sulphate were obtained. The mother-liquors were now basified with sodium bicarbonate in the presence of chloroform, deep brown, amorphous, insoluble alkaloidal material being removed by filtration through kieselguhr. The chloroform extracts were completely freed from solvent and gave 4 g. of syrupy bases, which on solution in warm methyl alcohol rapidly deposited *d-iso*chondrodendrine base (1·7 g.), m. p. 305°, as a very fine, crystalline powder. On concentration of the methyl-alcoholic mother-liquor *l*-bebeerine (0·7 g.), m. p. 215°, separated. The high dextro-rotation for the crude crystalline starting material cannot be due to the *d-iso*chondrodendrine or to the *l*-bebeerine and must be attributed to the chloroform-insoluble amorphous bases which separate whenever these alkaloids are regenerated from crude solutions of their salts.

The combined methyl-alcoholic mother-liquors (A) were neutralised with mineral acid, the methyl alcohol removed, and the aqueous solution precipitated with saturated sodium bicarbonate solution (100 c.c.). The vacuum-dried precipitate was finely powdered and extracted with ether (Soxhlet) and gave 14·8 g. of crude amorphous solid bases, which on boiling with methyl alcohol (50 c.c.) gave *l*-bebeerine (9·0 g.), m. p. 208°.

From 924 g. of root there were therefore isolated 43·1 g. of *d-iso*chondrodendrine sulphate, 1·7 g. of *iso*chondrodendrine base, and 9·7 g. of *l*-bebeerine base.

Alkaloids of Ch. platyphyllum *from Bahia.*—Through the kindness of Dr. Randolph T. Major of Merck and Co., Inc., of Rahway, New Jersey, I was able to examine a quantity of the root, stems and leaves of *pareira brava*, collected by Mr. S. S. Schindler, a local exporter of medicinal plants and sent *via* the American Consulate in Bahia, Brazil. The leaves were identified as *Ch. platyphyllum* by Mr. B. A. Krukoff of New York.

Stems. The powdered material (538 g.) was percolated with 1% tartaric acid, the solution concentrated, and the alkaloids extracted with chloroform after basification with sodium bicarbonate. The residue obtained on removal of the solvent was dissolved in methyl alcohol (50 c.c.); *l*-bebeerine (28·1 g.), m. p. 215°, slowly crystallised. The yield corresponds to a 5·2% yield of *l*-bebeerine, calculated on the weight of the original stem. The optical rotation was taken in N/10-hydrochloric acid, $[\alpha]_{5461}$ − 337·9° (calculated in terms of the base, c = 0·64).

The hydrochloride had m. p. 278° (efferv.) and this was not depressed by admixture with *d*- or *l*-bebeerine hydrochloride. A specimen of this base, recrystallised from boiling methyl alcohol (125 vols.), separated in prismatic needles, m. p. 215—216°. The specific rotation of the base, freed from methyl alcohol of crystallisation, was determined in N/10-hydrochloric acid: $[\alpha]_{5461}$ — 340°, whence $[\alpha]_{5461}$ — 339° for the ion (c = 0·44); $[\alpha]_D$ — 275·9° for the ion.

The main methyl-alcoholic mother-liquor was evaporated to dryness, and the residue extracted with ether (Soxhlet). A further 0·22 g. of *l*-bebeerine was thus obtained; there was no evidence for the presence of any *d-iso*chondrodendrine.

Root. The finely powdered material (720 g.) was worked up in the same way as the stem. The yield of crystalline alkaloids obtained on solution of the chloroform-soluble bases in methyl alcohol was 50·8 g., corresponding to a 7% yield on the root. The base had a slightly mixed appearance and had $[\alpha]_{5461}$ — 308°, calculated in terms of the base in N/10-hydrochloric acid. It was crystallised as hydrochloride and gave *l*-bebeerine hydrochloride (46·4 g.), m. p. 280° (efferv.). On concentration, the mother-liquors gave a mixture of the hydrochlorides of *l*-bebeerine and *d-iso*chondrodendrine. They were readily separated without filtration by dilution with a small volume of water and gentle warming, *d-iso*chondrodendrine hydrochloride (1·1 g.), m. p. 323° (decomp.), being undissolved. The mother-liquors on concentration now gave *l*-bebeerine hydrochloride (8·9 g.), m. p. 280° (decomp.). The bases from the mother-liquor were then regenerated and dissolved in hot methyl alcohol (50 c.c.); *d-iso*chondrodendrine (0·62 g.), m. p. 318°, soon separated and was collected while the solution was still warm. The filtrate then deposited *l*-bebeerine (1·36 g.), m. p. 209°.

The original methyl-alcoholic liquors which had given 50·8 g. of crystalline bases were evaporated to dryness, and the residue powdered and extracted with ether (Soxhlet). A small quantity of amorphous alkaloid separated, which on solution in the usual way in a little warm methyl alcohol gave *l*-bebeerine (0·14 g.) and *d-iso*chondrodendrine (30 mg.).

Leaves. These (300 g.) were dried at 37°, powdered, and extracted with cold 1% tartaric acid solution. The concentrated extract was basified with sodium bicarbonate, and the bases extracted with chloroform (yield, 3·4 g.). This alkaloidal material was a gum which gave a positive Millon reaction and in alcoholic solution a purple colour with ferric chloride, a reaction also shown by bebeerine base. All attempts to obtain crystals from it by the use of methyl alcohol or other solvents failed. It was therefore dried, finely powdered, and exhaustively extracted (Soxhlet) with dry ether. An amorphous solid (2·9 g.) separated in crusts in a manner suggestive of bebeerine, but, when moistened with a little methyl alcohol, did not crystallise. It was therefore neutralised with N-hydrochloric acid (8·5 c.c.), and the bases fractionally liberated to chloroform, by addition of nine 1 c.c. portions of N-sodium hydroxide. After removal of the chloroform from each fraction, the residue was dissolved in a little methyl alcohol and kept. Fraction 8, the most basic, gradually deposited a fine powder (40 mg.), m. p. 303°. It was recrystallised from methyl alcohol by extraction with boiling methyl alcohol in a micro-thimble and separated as a crystalline powder, which proved to be *d-iso*chondrodendrine, m. p. 316° (decomp.), undepressed by admixture with pure *d-iso*chondrodendrine. It also gave a typical Millon reaction and was very sensitive to warm dilute nitric acid.

Fractions 5 and 6 gradually deposited *l*-bebeerine, which crystallised from methyl alcohol (yield, 10 mg.) in prisms which readily became opaque through loss of methyl alcohol of crystallisation, m. p. 204°, mixed m. p. with *l*-bebeerine, 208°.

Isolation of l-*chondrofoline.* Fractions 2 and 3, representing much less basic material, slowly deposited well-formed glassy crystals (0·23 g.). Crystallised from methyl alcohol, it separated in triangular plates with some double pyramids (0·18 g.), m. p. about 135° (slow efferv.) (Found * for the air-dried base: loss at 95°, 6·0. $C_{35}H_{36}O_6N_2,2H_2O$ requires H_2O, 5·8%. Found for partially dried base: C, 70·7, 70·6; H, 6·8, 6·6; N, 4·7, 4·9; MeO, 14·6, 14·9. $C_{35}H_{36}O_6N_2,H_2O$ requires C, 70·2; H, 6·4; N, 4·7; 3MeO, 15·6%. Found for the dried base: *M*, Rast, 594, 576. $C_{35}H_{36}O_6N_2$ requires *M*, 580).

Chondrofoline is a phenolic alkaloid which does not give a Millon reaction; in methyl alcohol it gives a faint pink-purple colour on addition of a trace of ferric chloride. The hydrochloride and the sulphate were not obtained crystalline, but the *nitrate*, obtained by addition of ammonium nitrate to a solution of the base in dilute hydrochloric acid, separated in needles. On recrystallisation from water it separated as a felt of needles, m. p. 225° (decomp.) (Found for the salt dried at 105°: C, 57·5, 57·7; H, 6·1, 6·2; N, 7·6, 7·3; MeO, 11·6, 11·7.

$C_{35}H_{36}O_6N_2,2HNO_3,H_2O$

requires C, 58·0; H, 5·6; N, 7·7; 3 MeO, 12·8%). The specific rotation of the base dried at

* All analyses are micro.

95°, determined in N/10-hydrochloric acid, was $[\alpha]_{5461}^{20°}$ − 280·6° (calculated in terms of the base).

Exhaustive methylation of chondrofoline. Chondrofoline (117 mg.) was boiled in methyl-alcoholic solution with methyl iodide and potassium hydroxide (0·4 g.). After removal of the solvent and replacement by warm water the solution gelled on cooling. The product was converted into the chloride by freshly prepared silver chloride, the solution of the methochloride evaporated to dryness, and the non-crystalline methochloride extracted from inorganic salts by treatment with ethyl alcohol. The syrup left on removal of the alcohol was boiled with 20% sodium hydroxide solution (10 c.c.) for 2 hours, and the methine bases taken up in chloroform. After removal of the solvent the bases were boiled for several hours in methyl-alcoholic solution with methyl iodide. On concentration a crystalline solid separated which on one crystallisation from methyl alcohol gave small prisms or fine needles of *O*-methylbebeerinemethine methiodide B (43 mg.), m. p. 237°, not depressed by an authentic specimen. It gave the colour reaction with sulphuric acid characteristic of the methine methiodide B and not that of the methine methiodide A (King, J., 1935, 1381). From the mother-liquors a more soluble salt separated; on crystallisation from methyl alcohol it formed diamond-shaped plates (21 mg.), m. p. 190°. This salt had a lævorotation and was the lævo-enantiomorph of *d*-*O*-methylbebeerinemethine methiodide. A mixture of the two salts showed no depression of melting point and their colour reactions with sulphuric acid were identical.

Alkaloids of Chondrodendron microphyllum.—Through the good offices of the British Consul in Bahia a quantity of "*parreira brava*" roots was collected locally together with specimens of the leaves. The latter were identified through the kindness of Prof. Hochreutiner of Geneva as *Chondrodendron microphyllum*. The root (845 g.) was powdered and percolated with 1% tartaric acid solution in the usual way. The percolate was very dark coloured and percolation was very slow. In this respect it differed from *pareira brava*. The concentrated solution was made alkaline with sodium bicarbonate, and the chloroform-soluble alkaloids collected, dissolved in methyl alcohol, and kept at 0°; yield, 28·1 g. of crystalline bases, corresponding to a 3·3% yield on the original root. This was neutralised with N-sulphuric acid (93·3 c.c.), and the solution concentrated slightly over sulphuric acid in a vacuum. *iso*Chondrodendrine sulphate (18·3 g.) separated in the characteristic double pyramids. The base isolated from the aqueous mother-liquor was dissolved in hot methyl alcohol (50 c.c.). There was a rapid crystallisation of 16·7 g. of mixed bases melting over a range upwards from 213°. This crop was crystallised as hydrochloride and gave *d*-bebeerine hydrochloride (15·4 g.), $[\alpha]_{5461}$ + 318° for the basic ion. The mother-liquor was regenerated to base, which on solution in boiling methyl alcohol (30 c.c.) crystallised very rapidly in microscopic crystals of *d*-*iso*chondrodendrine (1·22 g.), m. p. 323—325°. The still warm filtrate deposited *d*-bebeerine (1·52 g.), m. p. 212°, followed on concentration by a further crop of *d*-*iso*chondrodendrine (0·2 g.). The methyl-alcoholic mother-liquors from which 16·7 g. of mixed bases had been obtained, on concentration and prolonged standing, gave a further 60 mg. of *iso*chondrodendrine base, m. p. 313°. From 845 g. of root there were thus obtained 18·3 g. of *d*-*iso*chondrodendrine sulphate, 1·42 g. of *iso*chondrodendrine base, 15·4 g. of *d*-bebeerine hydrochloride, and 1·5 g. of *d*-bebeerine base.

Alkaloids of Ch. candicans (*Rich* ex *D.C.*) *Sandwith*.—Through the valuable co-operation of Mr. B. N. Wood, Curator of Forests, British Guiana, and the Senior Forestry Officer, Mr. T. A. W. Davis, an abundant supply of this *chondrodendron*, known locally as "granny's backbone," was obtained. Its identity was kindly confirmed by Mr. N. Y. Sandwith of the Herbarium, Kew.

The stems were first cut up into amenable size and then fed into a drug mill and powdered to pass a 12-mesh sieve. A 1500 g. sample was then percolated with 1% tartaric acid, and the solution concentrated to 1200 c.c. This was worked up in 100 c.c. portions by adding excess of saturated sodium bicarbonate solution in the presence of a liberal supply of chloroform. After thorough mixing, the emulsified solutions were filtered through a thin layer of kieselguhr under reduced pressure to remove non-chloroform soluble material. After being well washed with chloroform, the filtrate was extracted six times with chloroform, no further difficulty with emulsions being encountered. The combined chloroform extracts corresponding to 1500 g. of stem were evaporated to dryness (yield, 32·7 g.) and the residue dissolved in hot methyl alcohol. Rapid crystallisation took place and on keeping at 0° at least two different crystalline bases had separated; yield 20·1 g., m. p. 282—289°, $[\alpha]_{5461}$ + 197° for the base in N/10-hydrochloric acid solution. On evaporation of the solution from the rotation determination the hydrochloride crystallised in plates (characteristic of *iso*chondrodendrine hydrochloride).

The separation of the main alkaloids was carried out as follows. The crude crystalline

alkaloids (19·8 g.) were neutralised with warm N-sulphuric acid (63 c.c.) and readily gave *d-iso*chondrodendrine sulphate (17·72 g. after recrystallisation) in two crops. The first mother-liquors, which would not crystallise further as sulphate, were regenerated to base by means of chloroform and bicarbonate, an insoluble amorphous product being removed by filtration. The syrupy base left on removal of the chloroform was dissolved in warm methyl alcohol (50 c.c.) and gave *iso*chondrodendrine base (1·0 g.) and on concentration *d*-bebeerine (3·14 g.), followed by a further small crop of *d-iso*chondrodendrine (0·48 g.). The residual base left in this methyl-alcoholic mother-liquor and the base left in the aqueous recrystallisation liquors of the *iso*chondrodendrine sulphate were recovered, powdered, and extracted (Soxhlet) with ether. An amorphous base separated in the ether and when crystallised from methyl alcohol gave *d*-bebeerine (0·94 g.).

The original methyl-alcoholic mother-liquor which had deposited 20·1 g. of mixed alkaloids was evaporated to dryness, and the amorphous solid powdered and extracted with ether (Soxhlet). In this way a further 0·85 g. of crude *d*-bebeerine was obtained. From 1500 g. of stem there were thus obtained 17·7 g. of hydrated *d-iso*chondrodendrine sulphate, 1·48 g. of *iso*chondrodendrine base, and 4·93 g. of *d*-bebeerine.

Alkaloids of Radix Pareiræ Bravæ *of Commerce.—Isolation of* d-*bebeerine*, d-iso*chondrodendrine, and* d-iso*coclaurine*. 56 Pounds of this material were bought on the London market through Messrs. Allen and Hanbury. Its authenticity was kindly confirmed by Mr. T. E. Wallis, Reader in Pharmacognosy in the University of London and Curator of the Pharmaceutical Society's Museum. One-half of this material was ground to pass a 12-mesh sieve and was then percolated with 1% tartaric acid solution in the usual way. The concentrated extract was precipitated at 0° with excess of saturated sodium carbonate solution, and the solid collected, dried in a vacuum, powdered, and extracted with dry ether (Soxhlet) for many weeks to exhaust the ether-soluble bases. At intervals the undissolved solid was dried and reground and extraction continued. On removal of the ether the cream-coloured amorphous powder containing fats was neutralised with hydrochloric acid, and filtered through a thin layer of kieselguhr, which removed most of the fat. The filtrate was extracted once or twice with ether to remove the remaining fat, concentrated under reduced pressure at 50°, and allowed to crystallise. The *d*-bebeerine hydrochloride so obtained was recrystallised from water to constant rotation, $[\alpha]_{5461}^{20°}$ + 294° (*c* = 0·7) for the dried salt, whence $[\alpha]_{5461}$ + 329° for the ion. This is not the maximum value for pure *d*-bebeerine, since from the stems of *Ch. platyphyllum* which did not contain any *iso*chondrodendrine a *l*-bebeerine was obtained having $[\alpha]_{5461}$ − 339° for the ion in N/10-hydrochloric acid. The slightly lower rotation recorded above for the *d*-bebeerine hydrochloride is probably due to the presence of *d-iso*chondrodendrine hydrochloride, for, from the final crystallisation liquors of a large amount of *d*-bebeerine hydrochloride, *d-iso*chondrodendrine hydrochloride was isolated and characterised as the sulphate.

As an alternative to crystallisation as the hydrochloride, the fat-free solution of the hydrochloride, freed from ether by warming, was poured into excess of dilute aqueous ammonia. The amorphous base was collected, dried, and added to 5 parts of hot methyl alcohol. (With care the whole of the amorphous base can be quickly got into solution before the crystalline bebeerine base begins to separate.) When recrystallised from methyl alcohol, the base had m. p. 215° and $[\alpha]_{5461}^{20°}$ + 345·7° (*c* = 0·4) for the ion when determined in N-hydrochloric acid solution. When once obtained pure, bebeerine will crystallise from other solvents; from benzene it separates in fine needles, from ethyl alcohol in very small needles, from propyl alcohol in tufts of needles. It also crystallises from aqueous acetone, but methyl alcohol has advantages over all other solvents. In methyl-alcoholic solution the base gives a port-wine colour on addition of a trace of aqueous ferric chloride. *iso*Chondrodendrine gives no colour. On addition of ammonium nitrate to a solution of a soluble salt of bebeerine or *iso*chondrodendrine sparingly soluble salts are precipitated. *iso*Chondrodendrine nitrate crystallises well in tablets, but bebeerine nitrate separates in amorphous globules. Both alkaloids are very sensitive to warm 3N-nitric acid. Attempts to prepare a crystalline sulphate from bebeerine in aqueous solution have failed.

Isolation of d-iso*Coclaurine*. From the final mother-liquors of the crystallisation of *d*-bebeerine hydrochloride a small crop of uniform plate-like crystals of d-iso*coclaurine hydrochloride* separated on long standing. These (0·7 g.) were dissolved in boiling water (10 c.c.) and separated in clusters of plates or narrow leaflets (0·45 g.), m. p. 175—176° (Found: C, 60·0, 60·3; H, 6·2, 6·4; N, 4·1, 4·2; Cl, 9·9, 9·8; MeO, 9·9, 9·8; H_2O, 2·3. $C_{17}H_{17}O_3N,HCl,H_2O$ requires C, 60·4; H, 6·0; N, 4·1; Cl, 10·5; MeO, 9·2; H_2O, 5·3%). The specific rotation was determined in water (*c* = 0·28 for the anhydrous salt), $[\alpha]_{5461}^{20°}$ + 23·9°. The free base, iso-

coclaurine, was liberated by sodium bicarbonate and was very sparingly soluble in chloroform. It crystallised from methyl alcohol in clusters of plates, m. p. 216—217°, almost the same m. p. as that of *d*-bebeerine but depressed to 195° by admixture with *d*-bebeerine (Found: C, 71·9, 71·7; H, 6·6, 6·7; N, 5·3, 5·5; *M*, 321. $C_{17}H_{17}O_3N$ requires C, 72·1; H, 6·1; N, 4·9%; *M*, 283). The amount of material available for the molecular weight by Rast's method in camphor was very small, but it sufficed to distinguish between a benzyl*iso*quinoline and a bisbenzyl*iso*quinoline. A methyl-alcoholic solution of the base gave with a trace of ferric chloride a coloration similar to that given by bebeerine but redder in shade. *iso*Coclaurine gave a typical Millon reaction, as does coclaurine. The nitrate was sparingly soluble and crystallised in rods; the sulphate was not obtained crystalline. O-*Methyliso*coclaurine methiodide was prepared by boiling *iso*coclaurine (50 mg.) in methyl alcohol containing potassium hydroxide with methyl iodide for several hours. On removal of most of the solvent and dilution with water the methiodide crystallised (70 mg.). It was recrystallised from water (2·5 c.c.) and showed a tendency to separate as an oil but eventually it separated in glassy tablets (50 mg.), m. p. about 113°, but on drying at 100° it melted about 137° but not sharply (Found: C, 50·0, 50·2; H, 6·6, 6·3; N, 2·8, 2·7; H_2O, 8·0; MeO, 18·0, 17·9. $C_{21}H_{26}O_3NI,2H_2O$ requires C, 50·1; H, 6·0; N, 2·8; H_2O, 7·2; 2MeO, 18·5%). Through the generosity of Prof. H. Kondo a specimen of *l*-coclaurine hydrochloride was available. A portion of this was methylated similarly and gave *O*-methylcoclaurine methiodide which showed all the properties of the above *O*-methyl*iso*coclaurine methiodide. It melted at the same temperature, separated in glassy tablets which were hydrated and showed on crystallisation a great tendency to separate as an oil. It is probably the *l*-enantiomorph, since the parent alkaloid was *l*-coclaurine.

Residual alkaloids. The residual alkaloidal mass left after exhaustive extraction with ether in a Soxhlet still contained crystalline alkaloidal bases. These may be the crystalline constituents of the amorphous chondrodine, bebeerine-B and β-bebeerine of earlier workers. They have not yet been examined in detail.

Characterisation of d-iso*Chondrodendrine.*—The most characteristic salt is the sulphate; this crystallises from water in colourless, glassy, double pyramids which when freshly prepared contain $15H_2O$. On prolonged exposure to air they effloresce and may lose as much as $8H_2O$. This salt is best crystallised from its own weight of warm water; it melts anhydrous at 291—292° (efferv.) (Found for two different samples of the freshly prepared salt: loss at 100°, 27·4, 28·7. Found after prolonged air-drying for many weeks: loss at 100°, 15·9. Calc. for $15H_2O$, 28·1; for $7H_2O$, 15·4%. Found for dried solid: C, 61·9, 61·9; H, 6·0, 6·1; N, 4·2, 4·1; MeO, 9·1. Calc.: C, 62·4; H, 5·8; N, 4·1; 2MeO, 9·0%). The rotation was determined in water with the hydrated salt, a separate sample being used for determining the water content, since the salt colours at 95°. For anhydrous salt, $[\alpha]_{5461}$ + 115·6° (c = 0·5). If Biot's law holds, this corresponds to $[\alpha]_D$ + 99·7°. Faltis and Neumann (*Monatsh.*, 1921, **42**, 321) record figures of $[\alpha]_D$ + 90·1° and 94·8° for commercial samples of the sulphate and describe hydrates containing 33·5 and 17·3% of water; they do not, however, give a m. p. for the dried salt. Scholtz (*Arch. Pharm.*, 1912, **250**, 684) found that the sulphate lost 31—32% of water and described it as crystallising with 16 to 18 molecules of water of crystallisation. *iso*Chondrodendrine hydrochloride was obtained by Scholtz (*ibid.*, 1913, **251**, 136) by addition of hydrochloric acid to a solution of the sulphate and was described as needles with no definite m. p. Faltis and Neumann describe it only as having $[\alpha]_J^{16°}$ + 140° in water. I find that it usually crystallises in plates, rarely in needles, m. p. 333° (decomp.), even when prepared by Scholtz's method. It is sparingly soluble in water and when dissolved in the minimum of boiling water will not readily separate without concentration of the solution (Found: C, 64·0; H, 6·1; N, 4·1. Calc.: C, 65·3; H, 6·0; N, 4·2%).

*iso*Chondrodendrine crystallises from methyl alcohol, in which it is extremely sparingly soluble, in microscopic needles, m. p. 316° (decomp.). Scholtz (*ibid.*, 1913, **251**, 136) describes the base obtained by adding water to the pyridine solution as small needles, m. p. 297°; Faltis (*Monatsh.*, 1912, **33**, 873) describes it as needles, m. p. 290°. It gives a typical Millon reaction in cold solution, as does bebeerine. *iso*Chondrodendrine methiodide was prepared by boiling the base in methyl alcohol with methyl iodide. It crystallised from methyl alcohol in short prisms, m. p. 287° (decomp.); from water, however, in microscopic double square pyramids (Found: C, 44·9; H, 6·0; N, 2·8; I, 25·4; H_2O, 13·6. Calc. for $C_{38}H_{44}O_6N_2I_2,8H_2O$: C, 44·6; H, 5·9; N, 2·7; I, 24·8; $8H_2O$, 14·1%). In water $[\alpha]_{5461}^{20°}$ + 64·3° (c = 0·17), whence for the ion + 90·2°. Scholtz (*Arch. Pharm.*, 1913, **251**, 136) describes the methiodide as large, hydrated prismatic crystals, m. p. 275° (efferv.). *O*-Methyl*iso*chondrodendrine methiodide was prepared by boiling *iso*chondrodendrine base (3 g.) in 0·5N-methyl-alcoholic potash (25 c.c.)

with methyl iodide (5 c.c.), an extra one-half of these quantities of methylating agents being added after one hour's boiling. The iodide separated almost quantitatively from the boiling solution in microscopic needles (4·29 g.), m. p. 312° (decomp.) (Found: C, 52·3; H, 5·5; N, 3·6. Calc.: C, 52·9; H, 5·3; N, 3·0%). It was anhydrous and had $[\alpha]_{5461}^{20°}$ + 1·5° (c = 0·39 in water). Scholtz and Koch (*ibid.*, 1914, **252**, 513) describe this salt as crystallising from water in needles, m. p. 294°; Faltis and Neumann give $[\alpha]_{J}^{16°}$ − 7° in 50% alcohol (c = 3·0).

The whole of the iodide (4·2 g.) was converted into the chloride, which was extremely soluble in water and crystallised in silky needles. Without isolation of the solid salt the volume was made up to 18 c.c., whence c is approximately 18 and $[\alpha]_{5461}$ + 57°. This figure is only a very approximate one, but it confirms the dextrorotation of these salts in aqueous medium. The solution was diluted to 30 c.c., treated with 6·0 g. of sodium hydroxide, and boiled for 90 minutes. The solid base which separated was taken up in chloroform, the latter evaporated, and the residual base neutralised with N-hydrochloric acid (9·0 c.c.). α-O-*Methyl*iso*chondrodendrine-methine hydrochloride* (2·26 g.) separated as a very sparingly soluble salt in tablets, m. p. 299° (decomp.), and was optically inactive; a further 80 mg. were obtained from the mother-liquor. This salt gave with sulphuric acid a cherry-red colour which turned to indigo-blue on heating (Found: C, 63·5; 63·7, 64·0; H, 7·2, 7·0, 6·8; N, 3·9. $C_{40}H_{46}O_6N_2,2HCl,2H_2O$ requires C, 63·2; H, 6·9; N, 3·7%). The base, regenerated through chloroform, sodium bicarbonate being used for liberation, crystallised readily and separated from absolute ethyl alcohol in stout crystals containing some rods, m. p. 206—207°. Faltis and Neumann give m. p. 204—205°.

The aqueous mother-liquors of the α-methine hydrochloride were strongly dextrorotatory (vol. 20 c.c., l 1 dcm., α_{5461} + 11·7°) and contained the β-methine of Faltis and Neumann. Also in agreement with their observations, the crude base isolated as a gum did not give the indigo-blue reaction with hot sulphuric acid.

iso*Chondrodendrine and Protocuridine.*—These bases are isomeric and both give the Millon reaction. Identity is precluded, since the bases and hydrochlorides are quite distinct. On complete methylation protocuridine gave *O*-methylprotocuridine methiodide, which formed needles, m. p. 318° (decomp.) (King, J., 1937, 1479). The *O*-methyl*iso*chondrodendrine methiodide described above crystallises in needles, m. p. 312° (decomp.). A careful comparison of these salts was made; unfortunately there was insufficient of the protocuridine derivative for conversion into another crystalline salt. Both methiodides crystallised from water or methyl alcohol in needles of the same appearance and they melted alone or in admixture at 314° (decomp.). Their solubilities also were similar. An aqueous solution of either gave no precipitate with ammonium nitrate and immediate amorphous precipitates with aqueous sodium picrate and perchloric acid. As a control, *O*-methylneoprotocuridine methiodide under parallel conditions behaved quite differently, giving crystalline precipitates with all three reagents.

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