The constitution of pilocarpine / by Hooper Albert Dickinson Jowett.

LXXIX.—*The Constitution of Pilocarpine. Part I.*

By Hooper Albert Dickinson Jowett, D.Sc.

In a previous communication (this vol., 494), it was shown that the constitutional formula previously proposed for pilocarpine by Hardy and Calmels must be abandoned, and an account was given of certain preliminary experiments on the constitution of isopilocarpine. Since then, Pinner and Kohlhammer (*Ber.*, 1900, **33**, 1424) have confirmed these conclusions, and have described several compounds of pilocarpine with bromine, as well as a new crystalline product, bromocarpinic acid, $C_{10}H_{15}O_4N_2Br$. The present paper contains a full account of the isolation and identification of the products formed by the oxidation of isopilocarpine with permanganate, and of the reactions of the base with soda lime, fused caustic potash, and methyl iodide.

In attacking the problem of the constitution of pilocarpine, two difficulties are encountered, first, the remarkable stability of isopilocarpine towards many reagents, for example, caustic potash or nitric acid, and secondly, the high price of the alkaloid, which renders it necessary to work with small quantities of material. Although it is not yet possible to propose a formula for the alkaloid, yet, from the results recorded in this paper, the existence of certain groups in the molecule can be shown to be highly probable.

From the fact that isopilocarpine only with difficulty forms salts with metallic hydroxides, such as sodium, barium, and copper hydroxides, as well as from its behaviour towards hot alkali, it is legitimate to conclude that it contains a lactone group. In many reactions, the nitrogen is eliminated as ammonia and methylamine, and when isopilocarpine methiodide is treated with caustic potash, methylamine alone is formed. Di-alkyl derivatives of isopilocarpine could not be prepared, thus confirming Chastaing's observation (*Compt. rend.*, 1885, **101**, 507), and since methyl iodide may be supposed to act as an acid towards a base, it must be assumed that of the two nitrogen atoms, the one which confers the basic character on isopilocarpine is evolved as ammonia, whilst the other is split off as methylamine, and does not act as basic nitrogen. Herzig and Meyer (*Monats.*, 1898, **19**, 56) have stated that one methyl group is attached to a nitrogen atom, and since methylisopilocarpine cannot be further methylated, it must be inferred that the nitrogen atoms occur as $:NH$ and $:N\cdot CH_3$, the former being that to which the basic character of isopilocarpine is due.

Further, the facts, that isopilocarpine is stable towards acids or alkalis and at high temperatures, and that when methylisopilocarpine

methiodide is decomposed by potash, methylamine and not dimethylamine is formed, show that these groups are attached to the rest of the molecule by a much stronger affinity than is the case in tropine, in which, by similar reactions, the nitrogen atom is eliminated, after successive methylations, as trimethylamine by simple heating with water. By distillation of isopilocarpine with soda-lime, a picoline, probably 3-methylpyridine, is formed in addition to ammonia and methylamine, but only in very small quantity. As the preparation of a sufficient quantity of the methylpyridine for its complete identification would have necessitated the use of large quantities of valuable material, and as its production in such small quantity, by so drastic a reaction, would not afford much clue to the constitution of the alkaloid, the investigation of this reaction was not further pursued.

By oxidation with permanganate, in addition to acetic acid, an acid, $C_7H_{10}O_4$, has been isolated, which proved on titration to be lactonic. It was not identical with any acid of this formula previously described, but, from the fact that the corresponding unsaturated dibasic acid yielded isobutyric acid on ultimate oxidation, the most probable formula would appear to be

$$(CH_3)_2CH\cdot\underset{O\text{—}CO}{CH}\cdot\underset{}{CH}\cdot CO_2H.$$

This would necessitate the existence of the complex

$$(CH_3)_2CH\cdot\underset{O\text{—}CO}{CH}\cdot\underset{}{CH}\cdot C$$

in isopilocarpine.

The formation of isobutyric acid by fusion of isopilocarpine with caustic potash, which is proved in this paper, and the lactonic nature of the alkaloid would thus be explained.

The results of the present investigation therefore indicate the existence of the following groups in pilocarpine and isopilocarpine:

(i) $(CH_3)_2CH\cdot\underset{O\text{—}CO}{CH}\cdot\underset{}{CH}\cdot C$. (ii) $:NCH_3$. (iii) $:NH$.

Experimental.

Distillation of Isopilocarpine with Soda Lime.

Four grams of isopilocarpine nitrate were treated with 50 grams of soda-lime as previously described (*loc. cit.*, 494), and the ammonium salt separated from the hydrochlorides soluble in absolute alcohol. The latter were dissolved in water, excess of auric chloride added, and the crystalline precipitate recrystallised from hot dilute hydrochloric acid. The filtrate from the precipitated aurichloride contained methylamine, which was isolated as the platinichloride and analysed.

0·0358 fused on heating and gave 0·0148 Pt. Pt = 41·34.
$(CH_3 \cdot NH_2)_2, H_2PtCl_6$ requires Pt = 41·3 per cent.

The aurichloride, amounting to less than 0·2 gram, melted at 185—187° (corr.), contained Au = 45·71 per cent. (methylpyridine aurichloride requires Au = 45·64 per cent.), and yielded a crystalline picrate which melted at about 130°, and a crystalline platinichloride melting indistinctly at 200°. When oxidised with permanganate, an acid was formed which gave a precipitate with copper acetate, but no coloration with ferrous sulphate; the quantity obtained was too small to admit of identification. These properties agree best with those of 3-methylpyridine.

The bases formed by distillation with soda-lime, therefore, are ammonia and small amounts of methylamine and (probably 3-) methylpyridine.

Interaction of Isopilocarpine with Methyl Iodide.

As isopilocarpine under certain conditions loses ammonia, and as it forms a methiodide, experiments were undertaken similar to those carried out on tropine (Merling, *Ber.*, 1891, **24**, 3108) to see whether, by successive methylation, the hydrogen atoms attached to the nitrogen could be replaced by methyl, and possibly some of the bonds broken. Pure isopilocarpine methiodide was treated with the theoretical amount of moist silver oxide and filtered, a solution of isopilocarpine methyl-hydroxide being obtained, which gave no precipitate on the addition of picric acid or platinic chloride. After boiling with these reagents and cooling, crystalline salts separated, however, and the same result was obtained if the solution of the methyl hydroxide was previously boiled with dilute hydrochloric acid. On boiling, therefore, isopilocarpine methyl-hydroxide loses water, becoming converted into methyl-isopilocarpine, which is capable of forming salts.

Methylisopilocarpine Picrate crystallises in beautiful, orange needles from hot water or alcohol, being sparingly soluble in these solvents at the ordinary temperature. The crystals melt sharply at 136° (corr.) to a clear liquid, and the melting point is not altered by further recrystallisation.

Methylisopilocarpine Platinichloride is obtained in well shaped, orange cubes which melt sharply at 218° (corr.) to a clear liquid without decomposition. It can be recrystallised from hot acidified water. On analysis:

0·1846 gave 0·0422 Pt. Pt. = 22·87.
0·2494 „ 0·0572 Pt. Pt. = 22·93.
$[C_{11}H_{15}(CH_3)O_2N_2]_2, H_2PtCl_6$ requires Pt = 22·87 per cent.

The hydrochloride was obtained by boiling the methyl-hydroxide with dilute hydrochloric acid, but neither this salt nor the nitrate has yet been crystallised. The base is soluble in water but insoluble in chloroform, thus differing from isopilocarpine, which is miscible with this solvent.

Methylisopilocarpine was heated with excess of methyl iodide, and the latter removed by distillation; the syrupy residue was then treated with moist silver oxide and filtered. After boiling with dilute hydrochloric acid, it yielded a crystalline picrate and platinichloride melting at 129° and 213° respectively, or a few degrees lower in each case than the corresponding salts of methylisopilocarpine. On analysis, it was found that further methylation had not taken place and that the base was unaltered methylisopilocarpine.

0·225 platinichloride gave 0·0514 Pt. Pt = 22·84.

$[C_{11}H_{15}(CH_3)O_2N_2]_2,H_2PtCl_6$ requires Pt = 22·87 per cent.

Further heating with methyl iodide and subsequent decomposition produced a similar result, the platinichloride melting at 212° and containing Pt = 22·54 per cent. It follows therefore that by the action of methyl iodide only monomethylisopilocarpine can be produced.

The methiodide prepared as above, was treated with 20 per cent. caustic potash solution in a sealed tube for 4 hours at 150°, and the volatile bases collected and converted into the hydrochloride. The salt was entirely soluble in absolute alcohol and was precipitated in four fractions with platinic chloride. These platinichlorides were analysed with the following results:

Fraction 1.	0·2226	gave	0·0916	Pt.	Pt = 41·15.
„ 2.	0·3258	„	0·134	Pt.	Pt = 41·13.
„ 3.	0·1788	„	0·074	Pt.	Pt = 41·38.
„ 4.	0·1654	„	0·0688	Pt.	Pt = 41·59.

$(CH_3\cdot NH_2)_2,H_2PtCl_6$ requires Pt = 41·3 per cent.

The base was therefore homogeneous and was methylamine.

The explanation of these results would seem to be that there exists in isopilocarpine the group :NH, which reacts with methyl iodide, forming $R{:}NH,CH_3I$; this with moist silver oxide yields $R{:}NH,CH_3\cdot OH$, which on boiling with dilute acids loses water and forms $R{:}N\cdot CH_3$.

The methylated base can react with methyl iodide, forming $R{:}NCH_3,CH_3I$, but on further treatment, the methyl iodide is eliminated, leaving the original base $R{:}NCH_3$. With caustic potash, the nitrogen of this complex is eliminated as methylamine, together with the same base formed from the other nitrogen group.

The non-basic product of the action of potash on methylisopilocarpine is under investigation.

Nitrous acid appears to have no action on isopilocarpine.

Oxidation of Isopilocarpine with Permanganate.

In the preliminary account of this reaction (*loc. cit.*), it was shown that the bases formed were ammonia and methylamine, and that two oils had been isolated, one having the properties of a lactone. Further investigation has shown that, in addition to a small quantity of acetic acid, an almost theoretical yield of a new acid, $C_7H_{10}O_4$, is obtained. Experiments were made with varying amounts of permanganate, but the best results were obtained when 6 molecular proportions were used, the yield then amounting to 70 per cent. of the theoretical. The reaction may be expressed by the following equation :

$$C_{11}H_{16}O_2N_2 + 7O + H_2O = NH_3 + CH_3{\cdot}NH_2 + C_7H_{10}O_4 + 3CO_2.$$

The details of one experiment may be given. Fifty-four grams of pure isopilocarpine nitrate, dissolved in a litre of water, were oxidised at 80° by the gradual addition of 188 grams of permanganate dissolved in 5 litres of water, the whole being kept thoroughly stirred by a turbine. The manganese dioxide was removed by filtration and thoroughly washed with hot water, the colourless filtrate and washings evaporated to a low bulk, made alkaline with caustic soda, and then distilled until the distillate was no longer alkaline. The residue was neutralised with hydrochloric acid, evaporated to dryness, mixed with sand, thoroughly dried, and then extracted with hot absolute alcohol in a Soxhlet apparatus. The alcoholic liquid, after saturation with hydrogen chloride, was allowed to stand, then boiled in a reflux apparatus for 2 hours, and distilled on the water-bath.

The distillate had the distinct odour of ethyl acetate, so the first fractions were collected separately. The residue, after removal of the alcohol by distillation, was thrown into water, the acid liquid extracted several times with ether, the ethereal liquid washed with water until free from acid, dried over calcium chloride, and distilled to remove the ether.

The first fraction distilled from the alcoholic solution was hydrolysed with caustic potash, and the alcohol removed by evaporation ; the residue was then dissolved in water, acidified with dilute sulphuric acid, and distilled. The crystalline barium salt obtained from the acid distillate was converted into the crystalline silver salt by treatment with silver nitrate solution, and on analysis the following result was obtained :

0·0584 gave 0·0376 Ag. Ag = 64·4.

$C_2H_3O_2Ag$ requires Ag = 64·67 per cent.

The acid was therefore *acetic* acid, and examination of the other portions of the alcoholic distillate failed to reveal the presence of any other acid.

The ethereal residue was fractionated first in a vacuum and finally under atmospheric pressure, the following fractions being obtained:

1. Boiling at 280—299°. Yield = 5 per cent.
2. „ „ 299° „ = 90 „
3. „ „ 200—210° under 10 mm. pressure. Yield = 5 per cent.

The liquid boiling at 299° was therefore pure, and on further fractionation was found to distil almost completely at this temperature. It was quite free from nitrogen. Analyses were made of a number of specimens, with the following results:

1. 0·1608 gave 0·34 CO_2 and 0·1106 H_2O. C = 57·83; H = 7·65.
2. 0·1822 „ 0·393 CO_2 „ 0·1266 H_2O. C = 58·83; H = 7·72.
3. 0·22 „ 0·4712 CO_2 „ 0·153 H_2O. C = 58·41; H = 7·73.
4. 0·239 „ 0·511 CO_2 „ 0·164 H_2O. C = 58·32; H = 7·62.

$C_9H_{14}O_4$ requires C = 58·07; H = 7·53 per cent.

The specific rotation of the pure liquid was determined with the following result:

$\alpha_{15°} = +22°$; $l = 50$ mm.; d 15°/15° = 1·1053; $[\alpha]_D^{15°} = +39·8°$.

The liquid was insoluble in water, but soluble in ether or alcohol, and had a peculiar, but not unpleasant, smell. On hydrolysis, it was found to be the ethyl ester of a dibasic or lactonic acid, as with phenolphthalein as indicator it required 2 molecular proportions of alkali to effect neutralisation.

Acid $C_7H_{10}O_4$, *formed by Oxidation.*

The acid was formed from the ester by hydrolysing either with caustic potash or aqueous hydrochloric acid. It is a thick, slightly yellow oil which does not solidify at −21°, boils at 210—220° under 10 mm. pressure, and is freely soluble in water, ethyl alcohol, or benzene. All attempts to crystallise it failed. On analysis:

0·1826 gave 0·3514 CO_2 and 0·1094 H_2O. C = 52·48; H = 6·65.

$C_7H_{10}O_4$ requires C = 53·16; H = 6·33 per cent.

When titrated in the cold with $N/10$ alkali, using phenolphthalein as indicator, 0·1678 required 10·7 c.c. $N/10$ alkali. Calculated 10·6 c.c. When titrated by boiling, first with excess of alkali, and then titrating back with acid, while still hot, 0·2234 required 26·00 c.c. $N/10$ alkali. Calculated 28·2 c.c.

The acid is therefore lactonic, and the formula may be written

$$CO_2H \cdot \dot{C}_4H_9 \cdot CO \cdot \dot{O}.$$

A neutral solution of the potassium salt gave white, gelatinous precipitates with lead acetate and silver nitrate, the precipitate with the latter undergoing slow reduction. It gave no reaction with barium or calcium chloride, or copper acetate.

Formation of the Unsaturated Diethyl Ester.

A preliminary experiment having shown that phosphorus pentabromide reacted with the ethyl ester, the whole of the ester at disposal was treated in this way. Twelve grams of the ester and 60 grams of phosphorus pentabromide were placed in a reflux apparatus, and, after the pasty mass had gradually liquefied, the whole was gently heated. Ethyl bromide was first given off, and then hydrogen bromide. As the object of the reaction was to obtain, not the bromo-derivative, but the unsaturated ester, the heating was continued until no more hydrogen bromide was evolved. The product was then gradually added to an excess of absolute alcohol, warmed to remove ethyl bromide, poured into ice-cold water, and the aqueous liquid extracted several times with ether. The ethereal solution, after washing with water, was dried over calcium chloride and distilled. In this way, 16·6 grams of crude product was obtained, which was fractionated in a vacuum. Considerable difficulty was experienced during distillation owing to frothing of the liquid, but ultimately three fractions were obtained, boiling respectively at 165—170°, 170—195°, and 195—200° under 20 mm. pressure. There were thus at least two substances present, so the first and third fractions were examined.

The *first fraction* (b. p. 165—170°) contained a large amount of bromine, but also instantly decolorised cold alkaline permanganate solution. It consisted chiefly of the bromoethyl ester, with a small quantity of the unsaturated compound. On analysis, the following results were obtained :

0·1702 gave 0·293 CO_2 and 0·097 H_2O. C = 46·94 ; H = 6·34.
$C_5H_9Br(CO_2C_2H_5)_2$ requires C = 44·74 ; H = 6·44 per cent.

The *third fraction* (b. p. 195—200°) contained bromine and readily decolorised cold alkaline permanganate. It consisted chiefly of the unsaturated ethyl ester, and on analysis gave the following result :

0·2934 gave 0·0654 AgBr. Br = 9·5.
$C_{11}H_{19}BrO_4$ requires Br = 27·1 per cent.

In order to complete the debromination, the whole of the liquid was heated with two molecular proportions of diethylaniline in a reflux apparatus on a sand-bath for 13 hours, when, on cooling, crystals of diethylaniline hydrobromide separated. The coloured liquid was then poured into excess of dilute hydrochloric acid and extracted with

ether. The ethereal solution was washed successively with dilute acid, dilute sodium carbonate solution, and water, dried over calcium chloride, and the ether removed by distillation. The residue was distilled in a vacuum, and on fractionation the greater part came over at 155° under 10 mm. pressure, a small quantity of a liquid of higher boiling point being apparently present.

Hydrolysis of the Unsaturated Ester and Oxidation of the Acid.

As the quantity of the ester was insufficient to admit of satisfactory fractionation, the unsaturated acid was isolated, purified as far as possible, and then oxidised by Crossley and Le Sueur's method (Trans., 1899, **75**, 161), in the hope that identification of the products of oxidation would give a clue to the constitution of the acid.

The ester boiling at 155° under 10 mm. pressure was dissolved in methyl alcohol and treated with an equal weight of caustic potash in methyl alcoholic solution. On the addition of alkali, a red colour was produced, changing to brown. The alcoholic liquid was heated on a water-bath for 2½ hours in a reflux apparatus, the alcohol removed by evaporation, and the residue acidified and extracted with ether. The ethereal solution, after washing with water, was dried over calcium chloride, and allowed to evaporate spontaneously. No crystals separated, but a light coloured oil was obtained, having the odour of a fatty acid. The oil distilled between 180° and 200° under 10 mm. pressure, and dissolved in sodium carbonate solution with effervescence; this solution at once decolorised permanganate at the ordinary temperature, thus affording proof of its unsaturated character (von Baeyer, *Annalen*, 1888, **245**, 146). On oxidation with cold permanganate, the acid required 98 c.c. *N*/10 solution (calculated 110 c.c.). After removal of the manganese dioxide, the filtrate was oxidised by chromic acid, distilled with steam and the volatile acid, which smelt strongly of a butyric acid, converted into the silver salt. On analysis, the following result was obtained:

0·126 gave 0·111 CO_2; 0·037 H_2O; and 0·069 Ag. C = 24·05; H = 3·26; Ag = 54·76.

$C_4H_7O_2Ag$ requires C = 24·61; H = 3·59; Ag = 55·38 per cent.

The neutral solution gave a precipitate with calcium chloride which dissolved on warming, and separated on cooling.

The acid, therefore, is isobutyric acid; the amount is small, but additional indirect proof of its identity is furnished by the fact that isobutyric acid is formed by fusion of pilocarpine with potash.

The reactions with phosphorus pentabromide and diethylaniline seem to prove conclusively the lactonic nature of the acid $C_7H_{10}O_4$, and since it does not agree with any of the known lactonic acids of

this composition, the formation of isobutyric acid would leave only one formula possible, namely,

$$(CH_3)_2CH\cdot\underset{O}{CH}\cdot\underset{CO}{CH}\cdot CO_2H.$$

The stability of the compound is remarkable, if it is the lactone of a β-hydroxymalonic acid; the formation of isobutyric acid, however, does not admit of the possibility of its being a γ-lactone, for if that were the case, acetone should be formed on oxidation, whereas careful search failed to detect it.

The reactions described in the preceding paragraphs may be represented as follows:

$$(CH_3)_2CH\cdot\underset{O}{CH}\cdot\underset{CO}{CH}\cdot CO_2Et \rightarrow (CH_3)_2CH\cdot CHBr\cdot CH(CO_2Et)_2$$

$$\rightarrow (CH_3)_2CH\cdot CH:C(CO_2Et)_2 \rightarrow (CH_3)_2CH\cdot CH(OH)\cdot C(OH)(CO_2H)_2$$

$$\rightarrow (CH_3)_2CH\cdot CO_2H + CO_2.$$

Experiments were made with pilocarpine to obtain the bromoethyl ester, by treatment with phosphorus pentabromide, but the basic character of pilocarpine interfered with the reaction, and a negative result was obtained.

Fusion of Isopilocarpine with Caustic Potash.

Chastaing (*Compt. rend.*, 1882, 94, 223) has described the fusion of pilocarpine with caustic potash, and stated that the products of the reaction were methylamine, carbon dioxide, butyric acid, and traces of acetic acid, and that the reaction may be represented by the equation

$$2C_{11}H_{16}O_2N_2 + 2H_2O = 2CO_2 + 4C_4H_8O_2 + 4CH_3\cdot NH_2 + O_2,$$

The analytical numbers quoted for the platinichloride * prove, however, that he was working with a mixture of ammonia and methylamine, and no details are given of the identification of butyric or acetic acids.

In the preliminary account of this reaction (*loc. cit.*, 496), the bases formed were proved to be ammonia and methylamine, and the acids yielded a silver salt, which contained a higher percentage of silver than required for silver butyrate. The reaction has been further investigated. Five grams of pure isopilocarpine were fused with 50 grams of caustic potash, and the fused mass, after cooling, dissolved in water acidified with sulphuric acid, and distilled with

* Found Pt = 42·5 per cent.
$(NH_3)_2, H_2PtCl_6$ requires Pt = 43·9 and $(CH_3\cdot NH_2)_2, H_2PtCl_6$ requires Pt = 41·3 per cent.

steam. The acid liquid was extracted with ether, the ethereal solution dried over calcium chloride, and distilled.

The residual liquid, which had a strong, rancid smell, was distilled at atmospheric pressure, when a small fraction was collected below 120°, but the greater portion came over between 120° and 155°, leaving no residue. On adding a little water to the higher fraction, oily drops separated, which dissolved on the addition of more water. From the aqueous liquid, a crystalline barium salt was prepared; this was extracted with boiling alcohol, in which it was very slightly soluble.

(i) The portion dissolved by alcohol gave a white, silver salt, which was not reduced on standing. On analysis the following result was obtained:

0·0388 gave 0·0214 Ag. Ag = 55·2.

(ii) The residue after extraction with alcohol gave a crystalline silver salt, which was recrystallised from hot water. On analysis, the following result was obtained:

0·0592 gave 0·0328 Ag. Ag = 55·4.

$C_4H_7O_2Ag$ requires Ag = 55·38 per cent.

The calcium salt was soluble in warm water, but separated again on cooling. The analytical numbers, boiling point, immiscibility with water, and solubility of the calcium salt, prove conclusively that the acid formed is isobutyric acid.

The portion boiling below 120° gave a small amount of an amorphous barium salt, insoluble in alcohol, and an amorphous silver salt (Ag = 57·4 per cent.), which underwent reduction on standing. No trace of acetic acid could be found.

Further investigation of the subject is proceeding on different lines, and it is hoped, either by the use of larger quantities of material, or by synthetical methods, to bring forward further proof of the correctness, or otherwise, of the formula now proposed for the acid $C_7H_{10}O_4$.

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LXII.—*The Constitution of Pilocarpine. Part II.*

By Hooper Albert Dickinson Jowett, D.Sc.

In former papers (this Journal, 1900, 77, 494, 851), it was shown that the nitrogen atoms occur in *iso*pilocarpine as :NH and :NCH_3, and that when oxidised with permanganate, an acid of the formula $C_7H_{10}O_4$ is produced. The constitutional formula,

$$(CH_3)_2CH\cdot\underset{O\text{—}}{CH}\cdot\underset{CO}{CH}\cdot CO_2H\ ,$$

was provisionally assigned to this acid, but as it is at present under investigation, and as frequent reference has to be made to it in this paper, the name of *pilopic acid* is proposed for it. The present paper deals with experiments on pilocarpine and *iso*pilocarpine, chiefly on the latter, as it is the more stable of the two alkaloids, and it was thought that the nature of the isomerism could be more conveniently studied when the constitution of the more stable isomeride had been determined. By the action of bromine on *iso*pilocarpine under varying conditions, several new products have been obtained. *Dibromo*iso-

pilocarpine, $C_{11}H_{14}O_2N_2Br_2$, is obtained as the first and principal product of the interaction of bromine and *iso*pilocarpine at the ordinary temperature. When the reaction is carried out in acetic acid solution, small quantities of an acid, probably *isopilocarpinic acid*, $C_{11}H_{16}O_4N_2$, are formed, together with a very small quantity of *monobromoisopilocarpine*, $C_{11}H_{15}O_2N_2Br$. Dibromo*iso*pilocarpine is a very feeble base, does not react with methyl iodide, and on reduction yields *iso*pilocarpine quantitatively, identical in all respects with the parent base. On oxidation with permanganate, it yields methylamine, a little ammonia, pilopic acid, and a new crystalline acid, *pilopinic acid*, $C_8H_{11}O_4N$. On further oxidation, the latter acid yields ammonia and pilopic acid.

When bromine acts on *iso*pilocarpine in aqueous solution at 100° in a sealed tube, two chief products are formed, *dibromoisopilocarpinic acid*, $C_{11}H_{14}O_4N_2Br_2$, and *monobromoisopilocarpinic acid*, $C_{11}H_{15}O_4N_2Br$, the former being a well-defined, crystalline acid, whilst the latter has only been obtained as an impure oil. Both these acids, on reduction with zinc and glacial acetic acid, yield the same product, *isopilocarpinolactone*, $C_{11}H_{14}O_4N_2$. This substance, obtained in well-defined crystals, is a neutral lactone, and with barium hydroxide yields the barium salt of hydroxy*iso*pilocarpinic acid. It possesses one remarkable character, namely, it is non-basic; it can be recrystallised from hot dilute acids, and does not unite with methyl iodide even at 100°. When dibromo*iso*pilocarpinic acid is reduced in alcoholic solution with sodium amalgam, the molecule is broken down and pilopic acid is formed. Dibromopilocarpine has been further studied, and it has been found, contrary to the statements of Pinner and Kohlhammer (*Ber.*, 1900, **33**, 1424), who first prepared this substance, that, like its isomeride, dibromo*iso*pilocarpine, it is very feebly basic and does not unite with methyl iodide. On reduction, it yields pilocarpine quantitatively, identical in all respects with the parent base. Experiments made to prepare bromocarpinic acid, first described by the above-mentioned authors, proved unsuccessful, and the amorphous acids, obtained by the action of bromine on pilocarpine at 100° in aqueous solution and in a sealed tube, yielded, on reduction, an amorphous acid which was certainly not carpinic acid, but probably pilocarpinic acid, $C_{11}H_{16}O_4N_2$.

In addition to the description, mode of preparation, &c., of the above-mentioned new substances, a number of experiments are recorded, in many cases with negative results, but which have an important bearing on the constitution of the alkaloid.

It is shown that *iso*pilocarpine is unaffected by reducing agents such as sodium in boiling amyl alcoholic solution or concentrated hydriodic acid at 127°, and does not exhibit the characteristic

behaviour of an unsaturated compound towards bromine or a saturated solution of hydrogen bromide in glacial acetic acid. The base is also remarkably stable towards chromic acid; action takes place only on prolonged heating, and no crystalline substance has been isolated from the product. Finally, although a full discussion of these results is reserved for a future communication, some of the more important theoretical points of interest are alluded to at the conclusion of this paper.

Action of Bromine on iso*Pilocarpine.*

(1) *In Acetic Acid Solution.*—When bromine acts upon *iso*pilocarpine at the ordinary temperature, dibromo*iso*pilocarpine, $C_{11}H_{14}O_2N_2Br_2$, is the chief product of the reaction; the experiment is best carried out in an acetic acid solution in a manner similar to that described by Pinner and Kohlhammer (*loc. cit.*) for the preparation of dibromopilocarpine. When solutions of bromine and *iso*pilocarpine in 80 per cent. acetic acid are mixed, an oily precipitate is formed which, however, quickly dissolves, and, on standing, the crystalline dibromo-*iso*pilocarpine perbromide separates in brownish needles. By the addition of water to the mother liquor, a further crop of crystals is obtained, but in most cases it was noted that the yield was only 60 per cent. of the theoretical, so the mother liquor from the crystals was further examined to discover the cause of the deficiency. The mother liquors, from which excess of bromine had been removed by long standing, either in the air or over caustic alkali, were extracted several times with ether, the ether then removed by distillation, and the residue boiled with water until all the acetic acid had been driven off. By this method, an oily acid residue was obtained, readily soluble in ether, alcohol, or benzene, but only sparingly so in water. After long standing in a vacuum over sulphuric acid, a solid varnish was obtained which could not be crystallised. It was therefore converted into the barium salt by adding water and boiling with excess of barium carbonate, filtering, and evaporating the solution to a low bulk. The barium salt was then precipitated with absolute alcohol, and was obtained as a microcrystalline powder which contained no bromine, and was analysed, with the following results:

0·0926 gave 0·0358 $BaSO_4$. Ba = 22·7.
0·131 „ 10 c.c. moist nitrogen at 20° and 766 mm. N = 8·6.
$(C_{11}H_{15}O_4N_2)_2Ba$ requires Ba = 22·3; N = 9·1 per cent.

The acid liquid, after extraction with ether, was rendered alkaline with ammonia and shaken several times with chloroform. After removing the chloroform by distillation, a small quantity of a neutral

crystalline substance was obtained. It was purified by repeated crystallisation from alcohol until of constant melting point, and then melted sharply at 164° (corr.). The quantity obtained was too small to admit of a complete analysis, but it was found to contain bromine and nitrogen. On analysis, the following results were obtained:

0·0276 gave 0·046 CO_2 and 0·0132 H_2O. C = 45·4; H = 5·3.
$C_{11}H_{15}O_2N_2Br$ requires C = 45·9; H = 5·2 per cent.

From these results, it would appear almost certain that this substance is *monobromoisopilocarpine.*

The alkaline liquid, after extraction with chloroform, was acidified with sulphuric acid, and then boiled to remove all acetic and hydrobromic acids. It was neutralised with caustic potash, evaporated to dryness, and extracted with absolute alcohol. A slight excess of sulphuric acid was next added to the alcoholic extract, and the precipitated potassium sulphate removed by filtration. Water was added to the alcoholic filtrate, the alcohol removed by evaporation, the aqueous solution boiled with excess of barium carbonate, and the barium salt separated from the filtrate by evaporation and precipitation with alcohol. This barium salt contained no bromine, and on analysis furnished the following results:

0·113 gave 0·1648 CO_2 and 0·05 H_2O. C = 39·8; H = 4·9.
0·0942 „ 7 c.c. moist nitrogen at 20° and 756 mm. N = 8·3
0·1072 „ 0·0418 $BaSO_4$. Ba = 22·93.
0·0956 „ 0·0364 $BaSO_4$. Ba = 22·40.
$(C_{11}H_{15}O_4N_2)_2Ba$ requires C = 42·9; H = 4·9; Ba = 22·3; N = 9·1 per cent.

The low percentage of carbon found might be due to the difficulty of burning a barium salt in an open tube, and to the presence of slight impurity in the material burnt. The yield of these bye-products was so small, and the difficulty of isolating the pure acid so great, that the investigation was not further pursued on these lines. Although the acid could not be obtained in crystalline form and its character determined, yet the results of the analyses indicate that only one acid, *iso*pilocarpinic acid, $C_{11}H_{16}O_4N_2$, is present. Further, the method of preparation, and the close agreement of the analytical numbers of three of the constituents with that required by theory, justify the assumption that the acid is sufficiently pure to assign the above formula to it.

The action of bromine on *iso*pilocarpine in acetic acid solution is therefore one mainly of substitution, with the formation of dibromo-*iso*pilocarpine and a very small quantity of monobromo*iso*pilocarpine, but at the same time a small proportion of the *iso*pilocarpine is oxidised to *iso*pilocarpinic acid.

(2) *In Chloroform Solution.*—When chloroform solutions of bromine and *iso*pilocarpine are mixed, a thick, red oil separates, which does not readily crystallise. By the addition of ammonia to this oil, decolorisation takes place, and a white, crystalline substance is obtained which, on examination, proved to be dibromo*iso*pilocarpine.

Even when a small amount of bromine was added to *iso*pilocarpine, dibromo*iso*pilocarpine was obtained, and monobromo*iso*pilocarpine could not be isolated by this method.

(3) *In Aqueous Solution.*—When bromine water is added to an aqueous solution of *iso*pilocarpine, the colour is instantly discharged, and dibromo*iso*pilocarpine separates as a white, crystalline precipitate. A good yield of the latter substance can be obtained by this method, but it is not so convenient as that previously described. When excess of bromine was added, and the solution boiled to remove bromine, no crystals separated on cooling, but after extracting the acid liquid with ether and removing the ether by distillation, an oily acid was obtained which did not crystallise even after long standing.

The barium salt, prepared in the usual manner with barium carbonate, was found on analysis to contain 24·8 per cent. of barium and 14·45 per cent. of bromine. These figures do not agree with those required for any likely formula for the acid, so the substance was probably a mixture.

(4) *In Aqueous Solution at* 100° *under pressure.*—When bromine is added to an aqueous solution of *iso*pilocarpine, and the mixture heated in a sealed tube at 90—140° for some hours, the dibromo*iso*pilocarpine first produced is oxidised to several acids. The reaction is best carried out as follows. 3·5 grams of *iso*pilocarpine were dissolved in 40 c.c. of water, 2·5 grams of bromine added, and the mixture heated in a sealed tube for 6 hours at 100°. The contents of the tube were then evaporated on the water-bath to remove bromine, and on cooling, crystals of *dibromo*iso*pilocarpinic acid*, $C_{11}H_{14}O_4N_2Br_2$, separated. The acid liquor, after filtration, was then extracted several times with ether, and, after distillation, the ethereal extract yielded a syrupy acid residue consisting chiefly of *monobromo*iso*pilocarpinic acid*, $C_{11}H_{15}O_4N_2Br$.

The yields of these acids were found to depend on the temperature employed, and the following results were obtained:

At 100°, 65 per cent. $C_{11}H_{14}O_4N_2Br_2$ and 71 per cent. ether extract.
At 130°, 16 „ „ „ „ 70 „ „ „

The best yield of dibromo*iso*pilocarpinic acid was obtained by heating for 6 hours at 100°.

The residual liquid, after extraction with ether, was made alkaline with caustic soda and distilled. The volatile bases obtained were

identified as *ammonia* and *methylamine*, and were present in equal proportions. The soluble hydrochloride gave a platinichloride containing Pt = 41·1 per cent. $(CH_3 \cdot NH_2)_2H_2PtCl_6$ requires Pt = 41·3 per cent. The alkaline liquid was next boiled with excess of acid, made alkaline with ammonia, and extracted with chloroform. The chloroform extract yielded a small quantity of dibromo*iso*pilocarpine. The residual liquid was neutralised, evaporated to dryness, the residue extracted with absolute alcohol, and the alcoholic extract saturated with hydrogen chloride. The alcoholic liquid was then distilled, the residue thrown into water, and the aqueous solution extracted several times with ether.

The ethereal liquid was washed with water, dried over calcium chloride, and distilled, when an oil was left of which part distilled at 280—300°, and gave no reaction for nitrogen, but a decided one for bromine. This oil was probably the *ethyl ester* of *bromopilopic acid.* The residual oil was almost solid, and contained both nitrogen and bromine, and probably consisted largely of the *ethyl ester* of *bromopilopinic acid.*

The main products of this reaction are therefore the two bromo*iso*pilocarpinic acids, but a portion of the molecule suffers further oxidation with the splitting off of ammonia and methylamine, and the formation of bromopilopinic and bromopilopic acids, these acids being formed, however, only in small amount.

*Dibromo*iso*pilocarpine*, $C_{11}H_{14}O_2N_2Br_2$.

This substance may be obtained in various ways, but was most economically and conveniently prepared as follows. Forty-six grams of pure *iso*pilocarpine were dissolved in 200 c.c. of glacial acetic acid and to this 72 grams of bromine dissolved in 72 c.c. of glacial acetic acid were gradually added. The precipitate which forms at first was redissolved, and, on standing, crystals of the perbromide separated. One hundred c.c. of water were added, and the mixture allowed to stand for 24 hours. The crystals were then filtered off, washed first with 30 per cent. acetic acid, and then with water, and drained on the filter pump. The crystals of the perbromide were suspended in excess of strong aqueous ammonia, when nitrogen was evolved, and a white, crystalline mass separated. The crystals were filtered off, washed well with cold water, and then recrystallised from hot alcohol until pure. The yield varied from 60 to 70 per cent. of the theoretical.

*Dibromo*iso*pilocarpine perbromide*, $C_{11}H_{14}O_2N_2Br_2,HBr_3$, crystallises from glacial acetic acid in long, yellowish-brown needles which melt at 165° (corr.). The crystals readily lose bromine either on exposure to the air or in solution, and the salt cannot be recrystallised without losing

bromine. A specimen recrystallised once from glacial acetic acid was analysed, with the following result:

0·113 gave 0·152 AgBr. Br = 57·2.

$C_{11}H_{14}O_2N_2Br_2 \cdot HBr_3$ requires Br = 65·9 per cent.

When treated with sodium thiosulphate, the crystals give the calculated yield of dibromo*iso*pilocarpine.

*Dibromo*iso*pilocarpine*, $C_{11}H_{14}O_2N_2Br_2$, crystallised in well-defined, rectangular, anhydrous prisms which, after frequent recrystallisation from alcohol, melted at 135° (corr.). This melting point was not altered on further recrystallisation. The crystals are almost insoluble in water, alcohol, or ether, but are more soluble in hot alcohol, acetone, or strong acids. A 6 per cent. solution in acetone was optically inactive. On analysis, the following results were obtained:

0·1736 gave 0·2316 CO_2 and 0·0608 H_2O. C = 36·4; H = 3·88.

0·2254 „ 15·8 c.c. moist nitrogen at 18° and 766 mm. N = 7·98.

0·2286 „ 0·2326 AgBr. Br = 43·30.

$C_{11}H_{14}O_2N_2Br_2$ requires C = 36·07; H = 3·82; N = 7·65; Br = 43·71 per cent.

This substance is only very feebly basic, it does not react with methyl iodide, and when boiled with excess of dilute acids is recovered unchanged. It dissolves in strong acids, but on dilution the original substance, and not the salt, is precipitated. The perbromide can only be obtained by adding excess of bromine to a solution of the substance in strong hydrobromic acid.

*Reduction of Dibromo*iso*pilocarpine.*

When dibromo*iso*pilocarpine is reduced either by sodium in amyl alcoholic solution or by zinc and glacial acetic acid, *iso*pilocarpine is formed quantitatively.

(1) *By Sodium in Amyl Alcohol Solution.*—One part of *iso*pilocarpine was dissolved in 20 parts of amyl alcohol (boiling at 135—140°) in a reflux apparatus, and, when the solution was boiling vigorously, one part of sodium was added in two portions. When action had ceased, the alcoholic solution was extracted several times with dilute sulphuric acid, and the acid liquid boiled to remove amyl alcohol and to decompose the sodium salt of *iso*pilocarpine. The acid liquid was then made alkaline with ammonia, extracted several times with chloroform, and the chloroform removed by distillation. The residue obtained was a colourless, thick oil, soluble in water, and alkaline to litmus. It was converted into the nitrate in the usual way, and, after one recrystallisation, this salt melted at 159° (corr.), and when mixed with an equal weight of pure *iso*pilocarpine nitrate melted at the same temperature.

A determination of the specific rotation gave the following result:

$\alpha_D^{18°} = +1°$; $l = 1$ dcm.; $c = 2{\cdot}584$; $[\alpha]_D^{18°} = +38{\cdot}7°$.

For *iso*pilocarpine nitrate $[\alpha]_D = +35{\cdot}7°$.

On analysis:

0·112 gave 0·1986 CO_2 and 0·064 H_2O. C = 48·4; H = 6·3.

$C_{11}H_{16}O_2N_2,HNO_3$ requires C = 48·6; H = 6·3 per cent.

The picrate, prepared in the usual way, melted at 161° (corr.); the platinichloride, which melted at 227°, was analysed, with the following result:

0·0228 gave 0·0054 Pt. Pt = 23·68.

$(C_{11}H_{16}O_2N_2)_2,H_2PtCl_6$ requires Pt = 23·58 per cent.

(2) *By Zinc and Glacial Acetic Acid.*—This is the most convenient method for carrying out the reduction. Dibromo*iso*pilocarpine was dissolved in a convenient quantity of glacial acetic acid, zinc dust added, and the mixture kept at 60—70° for 12—24 hours. The solution was diluted with water, filtered, the filtrate made alkaline with ammonia, and extracted with chloroform. After removing the chloroform by distillation, a residue was obtained which yielded a crystalline nitrate melting at 159° (corr.) and a crystalline picrate melting at 161° (corr.).

The physical constants and analyses just given prove that the base formed by the reduction of dibromo*iso*pilocarpine is identical in all respects with the original base, *iso*pilocarpine.

Oxidation of Dibromoisopilocarpine with Permanganate.

When dibromo*iso*pilocarpine is oxidised with a limited quantity of permanganate, the bromine is eliminated as hydrogen bromide, and, in addition, two acids are formed, *pilopic acid*, $C_7H_{10}O_4$, previously obtained by the oxidation of *iso*pilocarpine with permanganate, and a new acid, *pilopinic acid*, $C_8H_{11}O_4N$, together with methylamine and a small quantity of ammonia. Pilopinic acid is readily oxidised to ammonia and pilopic acid, and hence is only obtained in small quantity, the best yield being 20 per cent. of the theoretical, an equal amount of pilopic acid being formed at the same time. Experiments were made with varying amounts of permanganate, and under different conditions, but the best results were obtained with 3 molecular proportions of permanganate. The formation of pilopinic acid may be expressed by the following equation:

$$C_{11}H_{14}O_2N_2Br_2 + 4O + 2H_2O = C_8H_{11}O_4N + NH_2{\cdot}CH_3 + 2CO_2 + 2HBr.$$

The reaction was carried out as follows. To 36 grams of potassium permanganate dissolved in 600 c.c. of water, 24 grams of finely

powdered dibromo*iso*pilocarpine were added, the mixture well shaken, raised to a temperature of about 80°, and then allowed to stand overnight. The manganese dioxide was filtered off, and the filtrate, which was slightly coloured, decolorised by the addition of a few drops of sulphuric acid, when a little bromine was set free. The liquid was now made alkaline with caustic soda, distilled, and the alkaline distillate collected in hydrochloric acid. This acid solution was evaporated to dryness, when a crystalline residue was obtained which was almost completely soluble in absolute alcohol. The insoluble portion was stable in the air, and was ammonium chloride. The greater portion was soluble in absolute alcohol, and was converted into the platinichloride and analysed, with the following result:

0·092 gave 0·0382 Pt. Pt = 41·5.

$(CH_3{\cdot}NH_2)_2,H_2PtCl_6$ requires Pt = 41·3 per cent.

The relative proportion of the bases thus formed is the opposite of that obtained by the oxidation of *iso*pilocarpine when the ammonia forms the larger portion. The alkaline liquid remaining after distillation was neutralised with sulphuric acid, evaporated to dryness, the residue mixed with sand, dried, extracted with hot absolute alcohol, and esterified in a similar manner to the ester obtained from *iso*pilocarpine by oxidation. In this way, 5·3 grams of a very thick oil were obtained. It was distilled under 10 mm. pressure, when about half came over at 180°, leaving a thick oil which distilled at about 250°.

The first fraction was distilled under the ordinary pressure, and after three fractionations the greater part was obtained as an oil which distilled as a pale, straw-coloured, mobile liquid at 290—300°. On analysis, the following results were obtained:

0·1632 gave 0·3487 CO_2 and 0·114 H_2O. C = 58·27; H = 7·76.

$C_9H_{14}O_4$ requires C = 58·07; H = 7·53 per cent.

The ester of lower boiling point was therefore the monoethyl ester of pilopic acid.

The fraction of higher boiling point would not distil under the ordinary pressure, but after refractionation under 10 mm. pressure distilled with difficulty and fairly constant at 262°. On analysis, the following results were obtained:

0·2628 gave 0·5402 CO_2 and 0·1674 H_2O. C = 56·1; H = 7·1.

0·1954 „ 0·4014 CO_2 „ 0·1266 H_2O. C = 56·1; H = 7·2.

0·182 „ 11 c.c. moist nitrogen at 18° and 763 mm. N = 6·86.

$C_{10}H_{15}O_4N$ requires C = 56·3; H = 7·0; N = 6·6 per cent.

This ester is a very pale straw-coloured, thick oil, insoluble in water but soluble in alcohol or ether. It is non-basic, and yields no platinum salt.

Pilopinic Acid, $C_8H_{11}O_4N$.

This acid, obtained by the hydrolysis of its ethyl ester, can be crystallised with considerable difficulty, but is generally obtained as a colourless syrup.

It is best prepared as follows. The pure distilled ester is boiled in a reflux apparatus for several hours with 30 per cent. aqueous sulphuric acid, then transferred to a beaker, boiled with a little animal charcoal, and filtered. The acid solution is saturated with ammonium sulphate and extracted with pure ether, the ether extract washed with water, and then evaporated. On long standing in a vacuous desiccator, crystals are sometimes obtained, but these appear to be mixed with an oily substance, which is sometimes present in sufficient amount to prevent the acid from crystallising. The crystals are drained on a porous tile, and recrystallised from ethereal solution by precipitation with light petroleum. The acid crystallises in pearly plates which melt at 98° and are soapy to the touch. It is soluble in most of the ordinary organic solvents except light petroleum, and this fact renders its separation from the oil, which accompanies it in slight traces, a most difficult matter. The pure crystalline acid was analysed, with the following result:

0·106 gave 0·203 CO_2 and 0·0594 H_2O. C = 52·2; H = 6·2.
$C_8H_{11}O_4N$ requires C = 51·9; H = 5·9 per cent.

The crystals melt readily to an oil, which does not distil undecomposed, even in a vacuum.

Titrated with decinormal alkali, with phenolphthalein as indicator:

0·2138 required, in the cold, 11·7 c.c. for neutralisation, and when boiled with excess of alkali and titrated back with acid, 21·8 c.c.
$C_8H_{11}O_4N$ requires 11·5 c.c. and 23·1 c.c. respectively.

Neither the acid nor the ethyl ester appears to be basic, or forms a salt with platinic chloride.

The acid is lævorotatory, a determination of its specific rotation in dilute alcoholic solution giving the following result:

$$\alpha_D^{16^\circ} = -0{\cdot}4^\circ;\ l = 0{\cdot}5 \text{ dcm.};\ c = 5{\cdot}9;\ [\alpha]_D^{16^\circ} = -13{\cdot}6^\circ.$$

Some of the oily acid which had not been crystallised was converted into the barium salt by boiling with excess of barium carbonate, filtering, evaporating the aqueous solution to a low bulk, and precipitating with absolute alcohol. In this way, the barium salt was obtained as a microcrystalline powder, very soluble in water, but only sparingly so in alcohol. On analysis, the following results were obtained:

0·1948 gave 8·6 c.c. moist nitrogen at 20° and 756 mm. N = 4·9.
0·1064 „ 0·0466 $BaSO_4$. Ba = 25·8.
$(C_8H_{10}O_4N)_2Ba$ requires N = 5·5; Ba = 27·1 per cent.

The results obtained for both nitrogen and barium are low, no doubt owing to slight impurity, but prove conclusively the lactonic monobasic nature of pilopinic acid.

Oxidation of Pilopinic Acid.

The most important point in connection with the study of pilopinic acid was the identification of the nitrogenous product of oxidation. The formation of pilopic acid, together with pilopinic acid, during the oxidation of dibromo*iso*pilocarpine proves almost conclusively that pilopinic acid, on further oxidation, yields pilopic acid, but it was of the utmost importance to determine whether pilopinic acid contained the :NH or the :NCH_3 group present in *iso*pilocarpine.

The whole of the acid and ester at disposal was therefore oxidised with potassium permanganate in excess, the precipitated manganese dioxide removed by filtration, and the filtrate rendered strongly alkaline with caustic soda and distilled. The volatile alkali was converted into the hydrochloride, and yielded a crystalline residue, insoluble in absolute alcohol. The platinichloride prepared from it did not fuse on heating, but gave off white fumes. The only base formed by the oxidation of pilopinic acid is therefore *ammonia*. The oxidation of pilopinic acid may be expressed by the equation:

$$C_8H_{11}O_4N + O + H_2O = C_7H_{10}O_4 + NH_3 + CO_2.$$

*Dibromo*iso*pilocarpinic acid*, $C_{11}H_{14}O_4N_2Br_2$.

The mode of preparation of this acid has already been described. The crude product was purified by repeated recrystallisation from hot 90 per cent. alcohol until of constant melting point. It crystallises in well-defined, rectangular prisms, which melt with effervescence and decomposition at 235° (corr.). The crystals are fairly soluble in cold alcohol, more so in hot alcohol, but almost insoluble in water or ether. They are anhydrous, but owing to the ease with which they lose bromine when heated they must be dried in a vacuum. On analysis, the following results were obtained:

0·1362 gave 0·1612 CO_2 and 0·0462 H_2O. C = 32·3; H = 3·7.
0·1522 „ 0·1810 CO_2 „ 0·0502 H_2O. C = 32·4; H = 3·7.
0·2284 „ 0·2718 CO_2 „ 0·0746 H_2O. C = 32·5; H = 3·6.
0·2042 „ 13·8 c.c. moist nitrogen at 16° and 755 mm. N = 7·6.
0·1538 „ 0·1442 AgBr. Br = 39·9.
$C_{11}H_{14}O_4N_2Br_2$ requires C = 33·16; H = 3·52; N = 7·1; Br = 40·2 per cent.

The persistently low figures obtained for the carbon in these analyses cannot be explained, although it is possible that the large amount of bromine present may have some effect on the combustion. The acid is dextrorotatory, a determination of its specific rotation in alcoholic solution giving the following result:

$$\alpha_D^{16^\circ} = +1{\cdot}6^\circ;\ l = 1 \text{ dcm.};\ c = 6{\cdot}544;\ [\alpha]_D^{16^\circ} = +24{\cdot}4^\circ.$$

The determination of the basicity of this acid proved unexpectedly to be a most difficult problem. The first attempts were made by titration with cold and hot caustic alkali in the usual manner, and results were obtained indicating a tetrabasic acid. On examining the solution, it was found that the acid had been decomposed and sodium bromide formed.

Attempts to prepare the barium salt were unsuccessful, as even with barium carbonate the acid was decomposed.

The only way by which an approximate titration could be made was as follows:

0·2 gram of acid was dissolved in alcohol and 5·5 c.c. of decinormal sodium carbonate were added; the liquid was found to be distinctly acid to litmus. Five c.c. of alkali were then added, and as the liquid was now distinctly alkaline, it was boiled and rapidly titrated back with decinormal acid; the total amount of decinormal alkali required was thus found to be 7·0 c.c. $C_{11}H_{14}O_4N_2Br_2$, if monobasic, requires 5·0 c.c.

The acid is therefore certainly not dibasic, and, as it acts towards sodium carbonate as a monobasic acid, the excess of alkali used may be due either to slight decomposition of the acid, or to the effect of the lactonic grouping originally present in *iso*pilocarpine.

Attempts to make the silver and ammonium salts were unsuccessful. The basicity was finally determined as follows. About 0·5 gram of acid was dissolved in slight excess of ammonia and then left in a partly exhausted desiccator over sulphuric acid for 18 hours. The solution, which was perfectly neutral to litmus, was precipitated with platinic chloride, the mixture evaporated to a low bulk on the water-bath, alcohol added, and the platinum salt ignited and weighed.

0·4838 acid gave 0·1192 Pt. Pt = 24·63.

$C_{11}H_{14}O_4N_2Br_2$ requires Pt = 24·74 per cent.

Dibromo*iso*pilocarpinic acid is therefore monobasic.

Action of Reducing Agents on Dibromoisopilocarpinic Acid.

(1) *With Sodium in Amyl Alcohol Solution.*—This method having given good results with dibromo*iso*pilocarpine, was first tried. One

gram of acid was dissolved in 20 c.c. of amyl alcohol (boiling point, about 135°) and 2 grams of sodium added in two portions to the boiling solution, which was kept in vigorous ebullition in a reflux apparatus until all the sodium had dissolved.

The alcoholic solution, which was slightly coloured, was extracted by shaking with water, the sodium salt decomposed by boiling with acid, and the acidified aqueous liquid extracted with ether. The ethereal residue, which was very small, crystallised on standing. The crystals melted at 189° (corr.), were insoluble in water, decomposed sodium carbonate solution with effervescence, and were acid to test paper. The quantity obtained was too small to admit of further examination, and attempts made to increase the yield were unsuccessful.

(2) *With Sodium Amalgam in Alcoholic Solution.*—Three grams of the acid were dissolved in alcohol, sodium amalgam was added in considerable excess, and the mixture allowed to stand for several hours. Water was then added, the mixture warmed and filtered. The alkaline liquid was then strongly acidified with sulphuric acid and repeatedly extracted with ether. The ethereal solution was washed with water and distilled to a low bulk, when, on allowing the remainder of the ether to evaporate spontaneously, an oily residue was obtained which after a time crystallised. The crystals were freed from the rancid-smelling oil accompanying them by draining on a porous tile, and were recrystallised from ethereal solution by the addition of light petroleum. After several recrystallisations, they melted at 103° (corr.), and the acid on titration gave the following results:

0·093 gram required for neutralisation in the cold, with phenolphthalein as indicator, 5·7 c.c. decinormal alkali, and after boiling with excess of alkali and titrating back with acid, 10·9 c.c. $C_7H_{10}O_4$ requires 5·9 c.c. and 11·8 c.c. respectively.

The crystalline acid contained no nitrogen, and on analysis afforded the following results:

0·1006 gave 0·1978 CO_2 and 0·0608 H_2O. C = 53·6 ; H = 6·7.
$C_7H_{10}O_4$ requires C = 53·2 ; H = 6·3 per cent.

The acid is therefore *pilopic acid*, identical with that obtained from *iso*pilocarpine by oxidation with permanganate. It may be here stated that the acid, $C_7H_{10}O_4$, now termed *pilopic acid*, obtained from *iso*pilocarpine and previously described as an oil, has since been obtained in crystals which melt at 104° (corr.), and may be readily recrystallised from hot benzene solution.

(3) *With Tin and Hydrochloric Acid.*—The reduction carried out by these reagents according to the usual methods afforded a negative result.

(4) *With Zinc and Glacial Acetic Acid.*—The reduction of dibromo-*iso*pilocarpinic acid is best carried out by the following method. The crystalline acid is dissolved in a large quantity (about 20 times its weight) of glacial acetic acid, small quantities of zinc dust are added from time to time, and the mixture kept at 60—70° for about 24 hours. The mixture is then diluted with water, filtered, and the filtrate saturated with hydrogen sulphide. The zinc sulphide is filtered off and the filtrate distilled in a vacuum, with frequent additions of water to remove the greater portion of the acetic and hydrobromic acids. The liquid, after evaporating down to a low bulk, is then left overnight in a vacuous desiccator over caustic potash. The residual varnish is dissolved in hot water, boiled with a little animal charcoal if necessary, and filtered; on cooling, crystals are obtained. These are not crystals of *iso*pilocarpinic acid, as was expected, but of the *lactone of hydroxyisopilocarpinic acid,* $C_{11}H_{14}O_4N_2,H_2O$. The yield is almost quantitative.

Monobromoisopilocarpinic acid, $C_{11}H_{15}O_4N_2Br$.

It has already been stated (p. 504) that by the action of bromine on *iso*pilocarpine in a sealed tube, in addition to dibromo*iso*pilocarpinic acid, a syrupy acid is formed to the extent of about 70 per cent. at 130°. Although, unfortunately, all attempts to obtain this acid in the pure state have failed, yet the identification of its reduction product (by zinc and glacial acetic acid) with that obtained from dibromo-*iso*pilocarpinic acid, together with analyses of partially purified material, renders it possible to state the composition of the amorphous acid with a considerable degree of certainty. The crude ether extract was known to contain, in addition to the main constituent, small quantities of dibromo*iso*pilocarpinic, bromopilopinic, and bromopilopic acids. All attempts to crystallise it having failed, the ether extract was dissolved in strong hydrochloric acid and then precipitated by the addition of water. In this way, it was freed from most of the bromopilopinic and bromopilopic acids. The partially purified acid was then further purified by extraction of the dried residue with anhydrous ether, in which the dibromo*iso*pilocarpinic acid is insoluble. It was thus obtained as a brittle varnish which readily became oily. Analysis of this product showed that it was still impure.

Found, N = 7·0; Br = 31·8.

$C_{11}H_{15}O_4N_2Br$ requires N = 8·7; Br = 25·0 per cent.

The amorphous acid was therefore dissolved in a slight excess of ammonia, the solution allowed to stand overnight over sulphuric acid in a partially exhausted desiccator, and the neutral solution then

precipitated in five fractions with silver nitrate solution. The middle fraction was analysed, with the following results:

0·2128 gave 0·09 AgBr. Ag = 24·3.
0·0954 „ 0·0408 AgBr. Ag = 24·5.
0·1252 „ by Carius' method, 0·0448 AgBr. Br = 15·2.
0·2164 „ 15·2 c.c. moist nitrogen at 17° and 760 mm. N = 8·0.
$C_{11}H_{14}O_4N_2BrAg$ requires Ag = 25·3; Br = 18·7; N = 6·5 per cent.

Although the results of the analyses show that the silver salt was not pure, they point to the correctness of the formula given for this acid, and as the acid on reduction with zinc and glacial acetic acid yields *iso*pilocarpinolactone, $C_{11}H_{14}O_4N_2$, there seems to be conclusive evidence that the main constituent of the ether extract is monobromo-*iso*pilocarpinic acid.

Reduction of Monobromoisopilocarpinic Acid with Zinc and Glacial Acetic Acid.

The reduction of the ether extract of the mother liquors from which the dibromo*iso*pilocarpinic acid had been separated was carried out in precisely the same manner as that of dibromo*iso*pilocarpinic acid (see p. 593). The crystals thus obtained were recrystallised once from water and melted at 83° (corr.), and, when mixed with an equal quantity of the crystals obtained from dibromo*iso*pilocarpinic acid, melted at the same temperature.

The air-dried crystals were analysed, with the following results:

0·1368 gave 0·2568 CO_2 and 0·0774 H_2O. C = 51·2; H = 6·28.
$C_{11}H_{14}O_4N_2,H_2O$ requires C = 51·5; H = 6·25 per cent.

A determination of its specific rotation in alcoholic solution afforded the following result:

$$\alpha_D^{16^\circ} = -1·13^\circ;\ l = 1 \text{ dcm.};\ c = 2·18;\ [\alpha]_D^{16^\circ} = -51·9^\circ.$$

These numbers are identical with those given by the substance obtained from dibromo*iso*pilocarpinic acid by reduction, and conclusively prove the identity of the reduction products of the two acids.

iso*Pilocarpinolactone*, $C_{11}H_{14}O_4N_2,H_2O$.

This substance, obtained as above stated by the reduction of either dibromo- or monobromo-*iso*pilocarpinic acid with zinc and glacial acetic acid, is readily purified by recrystallisation from hot water. When recrystallised until of constant melting point, it was obtained as well-defined, rectangular prisms, which soften at about 80° and melt to a clear liquid at 83° (corr.). The crystals are readily soluble

in hot water, alcohol, acetone, glacial acetic acid, chloroform, or hot benzene, somewhat sparingly so in cold water or benzene and ether, and are almost insoluble in light petroleum.

The substance is lævorotatory, and a determination of the specific rotation in alcoholic solution gave the following result:

$$\alpha_D^{16°} = -2{\cdot}1°\ ;\ l = 1\ \text{dcm.}\ ;\ c = 4{\cdot}0464\ ;\ [\alpha]_D^{16°} = -51{\cdot}9°.$$

The crystals contain 1 mol. of water of crystallisation, which is lost even at 60—70°, and the melting point previously given appears to be that of the anhydrous substance, because, after heating the crystals at 100—120° till of constant weight, the residue after powdering melts at 83°. The anhydrous residue was dissolved in dry ether, and on spontaneous evaporation in the air a varnish was left which quickly became crystalline, but the crystals on examination were found to consist of the hydrated lactone. The hydrated lactone was therefore used for the physical constants, analyses, and subsequent experiments, since it is most readily obtained.

The air dried, powdered crystals were analysed with the following results:

0·164, at 100—120°, lost 0·0114 H_2O. $H_2O = 6{\cdot}95$.
0·1838 gave 0·346 CO_2 and 0·105 H_2O. $C = 51{\cdot}34$; $H = 6{\cdot}34$.
0·1018 ,, 0·1906 CO_2 ,, 0·0564 H_2O. $C = 51{\cdot}1$; $H = 6{\cdot}15$.
0·132 ,, 13·3 c.c. moist nitrogen at 18° and 750 mm. $N = 11{\cdot}2$.
$C_{11}H_{14}O_4N_2,H_2O$ requires $H_2O = 7{\cdot}03$; $C = 51{\cdot}5$; $H = 6{\cdot}25$; $N = 11{\cdot}0$ per cent.

The reduction product, therefore, has not the formula expected, namely, $C_{11}H_{16}O_4N_2$, derived from $C_{11}H_{14}O_4N_2Br_2$ by the replacement of 2 atoms of bromine by 2 hydrogen atoms. Further examination of this substance shows that it has somewhat remarkable properties. The aqueous solution is but very feebly acid to litmus, and although the crystals dissolve in sodium carbonate solution, they do so without effervescence.

Attempts were made to titrate the substance with caustic alkali and with sodium carbonate, but when phenolphthalein was used as the indicator it was noted that the change of colour was so gradual that it was not possible to obtain a satisfactory determination, and in this respect the substance differed markedly from pilopinic and pilopic acids, which are readily titrated.

(I) With decinormal alkali:

0·2038, dissolved in alcohol, required 9·8 c.c. of $N/10$ alkali, cold or hot, for neutralisation. $C_{11}H_{14}O_4N_2,H_2O$ (monobasic) would require 8·0 c.c.

(II) With decinormal sodium carbonate:

0·1676, dissolved in hot water, required 3·7 c.c. $N/10$ sodium carbonate to produce a permanent red tint when the liquid was boiled. $C_{11}H_{14}O_4N_2,H_2O$ (monobasic) would require 6·5 c.c.

The lactone was dissolved in hot water and boiled with excess of barium carbonate; after filtration, the substance crystallised out unchanged on cooling.

The *barium* salt of *hydroxy*iso*pilocarpinic acid* was prepared by boiling the lactone with excess of barium hydroxide solution, removing the excess of barium with carbon dioxide, filtering off the barium carbonate, and evaporating the filtrate to a low bulk. The barium salt was readily soluble in water, but separated from the concentrated solution as a microcrystalline powder; it was analysed, with the following result:

0·1986, dried at 130° until of constant weight, lost 0·0204 H_2O and gave 0·0686 $BaSO_4$. $H_2O = 10·3$; $Ba = 20·3$.
$[C_{11}H_{14}(OH)O_4N_2]_2Ba,4H_2O$ requires $H_2O = 10·0$; $Ba = 19·1$ per cent.

The high percentage of barium found is due to the presence of a small amount of barium carbonate formed during evaporation.

The *silver* salt was prepared from the barium salt by adding silver nitrate solution to a concentrated aqueous solution of the barium salt. The amorphous precipitate was well washed with water, collected, dried on a porous tile, and analysed, with the following result:

0·0434 gave 0·0132 Ag. $Ag = 30·4$.
$C_{11}H_{14}(OH)O_4N_2Ag$ requires $Ag = 30·0$ per cent.

This substance is therefore the lactone of hydroxy*iso*pilocarpinic acid, and its formation may be explained as follows.

Only one of the bromine atoms suffers reduction, and the resulting monobromo-acid is decomposed on boiling with dilute acid with the formation of the hydroxy-acid, which loses water and gives the lactone. The reactions of dibromo*iso*pilocarpinic acid previously described indicate that the bromine is readily removed, and render this explanation probable.

*iso*Pilocarpinolactone is also non-basic; it may be recrystallised from hot acidified water, and when heated with methyl iodide in alcoholic solution is recovered unchanged. Even when heated with excess of methyl iodide in alcoholic solution in a sealed tube at 100° for 2 hours, no reaction took place, and the lactone was recovered unchanged. This non-basic character of *iso*pilocarpinolactone has, as will be shown later, a most important bearing on the constitution of the alkaloid.

Dibromopilocarpine, $C_{11}H_{14}O_2N_2Br_2$.

Chastaing (*Compt. rend.*, 1883, **97**, 1435) first described an amorphous product produced by the bromination of pilocarpine, but Pinner and Kohlhammer (*loc. cit.*) have prepared the crystalline dibromopilocarpine, and have described it somewhat briefly. According to these authors, the properties of dibromopilocarpine differ in some important respects from those of the isomeric dibromo*iso*pilocarpine described in another part of this paper. It was therefore considered necessary to prepare this substance and examine it in a more complete manner. The dibromopilocarpine was prepared according to the method given by these authors, and a yield of 71 per cent. of the theoretical obtained. This agrees fairly well with the yield of the isomeric dibromo*iso*pilocarpine (p. 585), but in this case the mother liquors were not further examined.

Dibromopilocarpine, prepared from pure pilocarpine nitrate (m. p. 176°), as first obtained, melted at 84°, and after frequent recrystallisation from 50 per cent. alcohol, separated in fine, colourless prisms which melted sharply at 95° (corr.). This melting point was not altered on further recrystallisation of the compound, so that the temperature given by Pinner and Kohlhammer, namely, 79°, is too low. Further, it was found that the substance could be readily recrystallised from hot 50 per cent. alcohol, whilst the above authors state that the crystals separate after long standing.

The crystals were air-dried and analysed, with the following results :

0·1436 gave 0·1904 CO_2 and 0·0498 H_2O. C = 36·2 ; H = 3·85.
0·1676 „ 0·2236 CO_2 „ 0·0578 H_2O. C = 36·4 ; H = 3·83.
0·1734 „ 0·1776 AgBr. Br = 43·59.
$C_{11}H_{14}O_2N_2Br_2$ requires C = 36·07 ; H = 3·82 ; Br = 43·71 per cent.

Dibromopilocarpine is dextrotatory like the parent base pilocarpine, and a determination of its specific rotation in alcoholic solution gave the following result :

$$\alpha_D^{15^\circ} = +1{\cdot}5^\circ ; \; l = 1 \text{ dcm.} ; \; c = 3{\cdot}444 ; \; [\alpha]_D^{15^\circ} = +43{\cdot}6^\circ.$$

Like its isomeride dibromo*iso*pilocarpine, dibromopilocarpine is very feebly basic, although Pinner and Kohlhammer state that its salts are all readily soluble. This important point was proved by the following experiments. Dibromopilocarpine was dissolved in excess of hot dilute sulphuric acid ; on cooling, crystals separated which were proved, by the melting point and absence of any reaction with barium chloride, to be unchanged dibromopilocarpine. Further, the substance was recovered unchanged after boiling with methyl iodide. The two isomeric bromo-derivatives have, therefore, similar properties.

Reduction of Dibromopilocarpine.

This was carried out with zinc and glacial acetic acid, as described for dibromo*iso*pilocarpine, the use of sodium and amyl alcohol being inadmissible in this case, since pilocarpine is converted into *iso*pilocarpine by this reagent.

The dibromopilocarpine gave a quantitative yield of a basic, colourless oil, soluble in water, which, with nitric acid, yielded a crystalline nitrate melting at 176°. The salt was once recrystallised from hot alcohol, and then melted at 177° (corr.).

A determination of its specific rotation in aqueous solution gave the following result:

$$\alpha_D^{18°} = +1{\cdot}8°;\ l = 1 \text{ dcm.};\ c = 2{\cdot}184;\ [\alpha]_D^{18°} = +82{\cdot}4°.$$

Pilocarpine nitrate melts at 177—178° (corr.) and has $[\alpha]_D = +82{\cdot}9°$. By the reduction of dibromopilocarpine there is formed quantitatively *pilocarpine*, identical in all respects with the parent base.

Bromocarpinic acid, $C_{10}H_{15}O_4N_2Br$.

This acid is stated by Pinner and Kohlhammer to be formed, together with another acid not yet examined, by the action of bromine on pilocarpine at 100° in a sealed tube, and to be a dibasic acid. It possesses, therefore, especial interest for two reasons, first, because it proves that the isomerism of pilocarpine and *iso*pilocarpine is structural and not stereoisomeric, since *iso*pilocarpine, by a similar reaction, yields dibromo*iso*pilocarpinic and monobromo*iso*pilocarpinic acids, and, secondly, because it completes the series of acids capable of formation between the parent base and pilopic acid by successive elimination of the three carbon atoms and the groups :NH and :NCH_3.

In view of the importance, therefore, of this compound, it was thought advisable to repeat the experiments.

Two experiments were conducted under the conditions detailed by Pinner and Kohlhammer, but in neither case could any crystalline product be isolated. This was the more remarkable as parallel experiments were proceeding at the same time on *iso*pilocarpine, and the crystalline dibromo*iso*pilocarpinic acid was readily obtained. The acid liquors from the bromination of pilocarpine were therefore extracted with ether, the ethereal extract distilled, and the residue reduced with zinc and glacial acetic acid in the usual manner. An oily substance was thus obtained which was insoluble in cold water, but fairly soluble in boiling water, and sparingly so in ether. Attempts to crystallise it from hot water and other solvents having failed, the oil was boiled twice with water, separated, and then taken up with ether. The

ethereal solution was washed many times with water to remove all acids soluble in water, and, on removing the ether by distillation, a residue was left which could not be crystallised. It was therefore dried in a vacuum over sulphuric acid, and titrated with decinormal sodium carbonate and with decinormal caustic alkali, with the following results :

1. 0·4256 required 17·8 c.c. $N/10$ sodium carbonate for neutralisation. $C_{10}H_{16}O_4N_2$ requires 37·3 c.c. ; $C_{11}H_{16}O_4N_2$ requires 17·7 c.c.
2. 0·1856 required 8·0 c.c. $N/10$ sodium hydroxide cold and 10·4 c.c. hot. $C_{10}H_{16}O_4N_2$ requires 16·3 c.c. ; $C_{11}H_{16}O_4N_2$ requires 7·7 c.c.

The end reaction in the second titration was not sharp, particularly when the solution was hot. The amorphous residue, which contained no bromine, was boiled with excess of barium carbonate, and the barium salt separated in the usual way by precipitation with alcohol. On analysis, the following result was obtained :

0·1764 gave 0·0664 $BaSO_4$. Ba = 22·14.

$C_{10}H_{14}O_4N_2Ba$ requires Ba = 37·7 per cent.
$(C_{11}H_{15}O_4N_2)_2Ba$ „ Ba = 22·3 „

The results of these experiments prove conclusively that in this case bromocarpinic acid is not formed, and they indicate with a considerable degree of probability that the action of bromine on pilocarpine under these conditions is analogous to the action on *iso*pilocarpine, namely, the formation of mono- or dibromo-pilocarpinic acid, and subsequent reduction of these acids to pilocarpinic acid. This view is supported by the experimental data, by analogy with *iso*pilocarpine, and by the fact that by similar reactions with *iso*pilocarpine a homogeneous product is formed, although, fortunately, in this case it was crystalline. It is hardly possible that such an amount of impurity could be present, if the acid be carpinic acid, as to account for the analytical results, and to produce the coincidence of these results with those required for pilocarpinic acid. It is possible that the acid formed under the conditions of these experiments is identical with that observed but not examined by Pinner and Kohlhammer.

Experiments on the Oxidation of iso*Pilocarpine.*

Experiments were made with various oxidising agents under varying conditions to see if any intermediate products of oxidation could be isolated. Although these experiments gave a negative result, they are placed on record, as they have an important bearing on the constitution of the alkaloid.

(1) *With Alkaline Permanganate.*—When *iso*pilocarpine is dissolved in sodium carbonate solution, and titrated with potassium perman-

ganate at 0°, oxidation takes place very quickly, the colour of the first few drops being discharged instantly and the reaction then proceeds quickly, but not instantaneously, each atomic proportion of oxygen requiring about 5 minutes for complete absorption. In this way, 5 atomic proportions of oxygen are quickly absorbed, but after that the reaction appears to proceed more slowly. To *iso*pilocarpine dissolved in alkali, potassium permanganate representing 5 atomic proportions of oxygen was added. The colour was quickly discharged, but on working up the volatile bases obtained it was found that the ammonia and methylamine had been produced in about equal proportion, so that it was not possible to prepare the pilopinic acid in this manner.

(2) *With Acid Permanganate.*—*iso*Pilocarpine in aqueous sulphuric acid solution was titrated with normal potassium permanganate solution at 15°, but oxidation proceeded very slowly indeed, the average time taken for the complete absorption of an atomic proportion of oxygen being 2 hours. No point was reached at which the action seemed to cease, and the results indicated that the oxidation was proceeding to some simple final product.

(3) *With Potassium Dichromate.*—*iso*Pilocarpine nitrate was treated with an equal weight of potassium dichromate in sulphuric acid solution for 12 days at 15°, and then for several hours at 80°, but no reaction took place, and the *iso*pilocarpine was recovered unchanged.

(4) *With Chromic Acid.*—When treated with an equal weight of chromic acid in aqueous solution for several days at the ordinary temperature, *iso*pilocarpine is recovered unchanged.

Pinner and Kohlhammer (*Ber.*, 1900, **33**, 2363) have stated that by the action of chromic acid on pilocarpine in sulphuric acid solution at the temperature of a water-bath, pilocarpoic acid, $C_{11}H_{16}O_5N_2$, is formed, and this acid with barium hydroxide yields a barium salt, $C_{11}H_{14}O_5N_2Ba$, which was only obtained in the amorphous condition. Their analyses, however, do not agree very well for the above formula, and a homogeneous product was hardly to be expected from such a reaction. Similar experiments have been carried out on *iso*pilocarpine with a negative result. No crystalline acid could be obtained from the products of the reaction, and the barium salts formed were of such an uninviting character that it was not considered worth while to analyse them. When the acid was regenerated from the barium salt, a rancid odour, recalling that of butyric acid, was noticed, so it was probable that the oxidation had been more complex than indicated by the authors above mentioned. The stability of both pilocarpine and *iso*pilocarpine towards strong nitric acid has already been noticed.

Action of Various Reagents on iso*Pilocarpine.*

The behaviour of *iso*pilocarpine towards bromine and alkaline permanganate suggested the possibility of the existence of a double bond in the part of the molecule other than that giving rise to pilopic acid, and as this point was of considerable importance, experiments were made to settle it. The reduction of dibromo*iso*pilocarpine to *iso*pilocarpine by various reducing agents had proved conclusively that bromine acts as a substituting and not as an additive agent, and experiments made by mixing chloroform solutions of *iso*pilocarpine and bromine showed that under no conditions was a dibromo-additive compound formed. Attempts were therefore made to reduce *iso*pilocarpine, but without success. Even when heated with four times its weight of fuming hydriodic acid at 127° in a reflux apparatus for 4 hours, the base was recovered unchanged. Sodium in boiling amyl alcohol or zinc and glacial acetic acid, even when acting on the base in a nascent condition, have been shown to be without action on *iso*pilocarpine (p. 586). Finally, the base was dissolved in glacial acetic acid saturated with hydrogen bromide, and allowed to stand many days, but no reaction took place other than the formation of *iso*pilocarpine hydrobromide. These experiments show conclusively that *iso*pilocarpine contains no ordinary double bond.

Discussion of Results.

The full discussion of the results recorded in this paper is reserved for a future communication, but a brief allusion may be made to one important point which arises out of the data here recorded. Two of the most important characters of *iso*pilocarpine which are not easy to explain are the non-basic nature of the methylamine residue and the peculiar behaviour of the carboxyl residue, which approximates most to a lactone, although the salt formed with barium hydroxide is decomposed by carbonic acid.

The results recorded in this paper give, however, a clue to the reason for this behaviour.

When an acidic group, for example, bromine or carboxyl, is introduced into *iso*pilocarpine, the ammonia residue loses its basicity, or at least to a very great extent. This happens in the case of dibromo*iso*pilocarpine, where the molecular structure remains unchanged, and also in *iso*pilocarpinolactone, where the molecular structure is probably altered. This "intramolecular neutralisation" of a basic nitrogen atom by carboxyl or bromine is unusual, as in many substances, for example, ecgonine or the pyridinecarboxylic acids, which contain an

acidic and basic element or groups, the dual property of acting as a base and an acid remains intact.

The fact is, however, that the introduction of these acidic groups into the molecule of *iso*pilocarpine does destroy (or nearly so) the basic character. This affords an explanation of the fact that the methylamine residue in *iso*pilocarpine is non-basic, for there already exists in the alkaloid a carboxyl residue, and these two groups may neutralise each other within the molecule and therefore leave a substance in which only the other nitrogen atom exerts a basic action. It is also conceivable that this intramolecular neutralisation may so modify the lactonic character as to cause the alkaloid to behave in the peculiar manner already indicated. This appears to be a much more probable explanation than the alternative one, which is that the nitrogen of the methylamine residue is pentad, and hence cannot react with methyl iodide, &c.

The further investigation of this subject is proceeding, particularly in reference to the constitution of pilopic acid, and it is hoped to communicate the results to the Society at a later date.

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CXLI.—*The Constitution of Pilocarpine. Part III.*

By Hooper Albert Dickinson Jowett.

In previous papers (Trans., 1900, **77**, 851, this vol., 580), it was shown that by the oxidation of *iso*pilocarpine with permanganate a small quantity of acetic acid is formed together with a crystalline lactonic acid, $C_7H_{10}O_4$, to which the name *pilopic acid* was assigned.

The present paper deals with a fuller examination of the products of oxidation of the alkaloid and with the constitution of pilopic acid.

By the oxidation of larger quantities of *iso*pilocarpine, it has been found that in addition to the acids above mentioned, small quantities of *propionic acid* and of a new acid, *homopilopic acid*, $C_8H_{12}O_4$, homologous with pilopic acid, are also obtained.

Although the correctness of the formula for pilopic acid has been questioned by Pinner and Kohlhammer (*Ber.*, 1900, **33**, 1424, 2357; 1901, **34**, 727), its accuracy has been confirmed, not only by analyses of the ethyl and methyl esters and of the crystalline acid and various derivatives, but particularly by the determination of the molecular weight of the methyl ester. The pure crystalline acid has been characterised and the anilide and the strychnine salt prepared in crystalline form and examined. The lactonic nature of pilopic acid has been proved by the preparation of the barium and silver salts and of the crystalline diamide of the corresponding hydroxy-acid, the behaviour of the acid in the last case recalling that of *iso*hexolactone, which with ammonia yields γ-hydroxy*iso*hexoamide. The reaction may be expressed by the following equation:

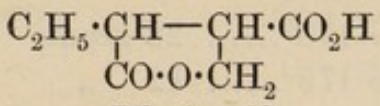
$$CO_2Et\cdot C_5H_9\begin{cases}CO\\ |\\ O\end{cases} + 2NH_3 = OH\cdot C_5H_9(CO\cdot NH_2)_2 + Et\cdot OH.$$

Homopilopic acid has not been crystallised, but the diamide of the hydroxy-acid has been obtained in well-defined crystals and the lactonic nature of the acid proved by the preparation of the barium salt of the hydroxy-acid. It is also shown that there are grounds for regarding Pinner and Kohlhammer's piluvic acid as a mixture of pilopic and homopilopic acids.

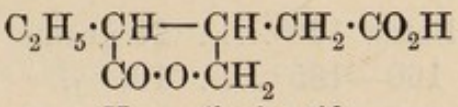
In a previous paper (Trans., 1900, **77**, 858), it was shown that from ethyl pilopate, by treatment with phosphorus pentabromide, &c., a small quantity of *iso*butyric acid was obtained. Repetition of this experiment with a larger quantity of material and the isolation and examination of the intermediate products of the reaction have shown that the quantity of *iso*butyric acid formed is very small and is produced by secondary reactions. As the constitutional formula previously proposed for pilopic acid was founded on the production of *iso*butyric acid as the main product of the reaction, the deductions are invalid and the formula must therefore be abandoned. By fusion of pilopic acid with potassium hydroxide at a high temperature, normal butyric acid is formed, whilst at a low temperature most of the acid is not attacked, but a small portion is converted into the isomeric unsaturated acid.

When homopilopic acid is fused with potassium hydroxide at a medium temperature, *α-ethyltricarballylic* acid is produced, and this acid has been identified beyond question by the formation of certain characteristic derivatives (see following paper).

Finally, it is shown that in all probability the constitution of pilopic and homopilopic acids may be represented by the following formulæ:



Pilopic acid.



Homopilopic acid.

Experimental.

Oxidation of iso*Pilocarpine and formation of Propionic Acid.*

About 1 kilo. of *iso*pilocarpine nitrate was oxidised with permanganate in the manner previously described, with this modification—that all the volatile acids formed were removed by steam distillation of the acid liquid previous to concentration and subsequent extraction with alcohol. The aqueous solution of the volatile acids, which was free from any rancid odour, was neutralised and evaporated to a low bulk, acidified with sulphuric acid, and extracted with ether. After removal of the ether by evaporation, the residue, which smelt strongly of acetic acid, was distilled. The first fraction, which came over below 120°, consisted chiefly of acetic acid and was not further examined. The remainder of the liquid distilled completely between 120° and 140°, the greater portion coming over at 136°. The distillate was converted into the crystalline barium salt, from which the silver salt was obtained in three fractions by precipitation with aqueous silver nitrate. Each fraction was analysed with the following results:

Fraction 1. 0·2706 gave 0·1616 Ag. Ag = 59·7.
„ 2. 0·1116 „ 0·0666 Ag. Ag = 59·7.
„ 3. 0·061 „ 0·0366 Ag. Ag = 60·0.
$C_3H_5O_2Ag$ requires Ag = 59·7 per cent.

In order to complete the proof of the identity of the acid, it was converted into the anilide, which melted at 103°, and on analysis furnished the following result:

0·1542 gave 13·6 c.c. nitrogen at 15° and 753 mm. N = 10·0.
$C_9H_{11}ON$ requires N = 9·4 per cent.

The only volatile acids formed during the oxidation of *iso*pilocarpine with permanganate are therefore *acetic* and *propionic* acids.

Fractional Distillation of the Ethyl Esters.

In order to determine the composition of the ethyl ester obtained by the oxidation of *iso*pilocarpine, it was submitted to careful fractionation.

One hundred and thirty grams of the crude ester were twice

refractionated under 10 mm. pressure and the following fractions obtained.

Below 160°.....	20 grams	170—175°.....	22 grams
160—165°.....	10 „	Above 175°.....	12 „
165—170°.....	52 „		

The fractionation in a vacuum was found to be unsatisfactory, as any slight variation in the pressure, which is almost unavoidable, affected the distilling point to a considerable extent.

An attempt was made to fractionate the ester under the ordinary pressure, using the rod and disc fractionating apparatus (Young, Trans., 1899, 75, 689) and a metal bath. This method, however, was only partially successful, owing to the risk of fracture of the flask by the high boiling liquid and to the loss of valuable material by decomposition in each distillation. If a small quantity, for example, 20 grams, of the ester be rapidly distilled from an ordinary distilling flask, it appears to boil quite constantly at 299° with very little decomposition, and a similar result is obtained by distilling it in a vacuum. As the result of three fractionations, the whole of the liquid was separated into three main fractions, which were analysed with the following results:

Fraction 1. B. p. 290—300° (or 164—166° under 10 mm. pressure).
0·2126 gave 0·4544 CO_2 and 0·1446 H_2O. C = 58·3; H = 7·6.
Fraction 2. B. p. 300—310°.
0·2102 gave 0·4544 CO_2 and 0·152 H_2O. C = 59·0; H = 8·0.
Fraction 3. B. p. 310—312° (or 210° under 10 mm. pressure).
0·1538 gave 0·338 CO_2 and 0·1126 H_2O. C = 59·9; H = 8·1.

$C_9H_{14}O_4$ requires C = 58·1; H = 7·5 per cent.
$C_{10}H_{16}O_4$ „ C = 60·0; H = 8·0 „
$C_{12}H_{20}O_5$* „ C = 59·0; H = 8·2 „

The greater portion of the ester was contained in fractions 1 and 2.

From these results, it would appear that two homologous esters are present, the first and third fractions each consisting of an almost pure ester and the second fraction being a mixture. This will account for the high figures persistently obtained for the carbon, as it is almost impossible to procure the lower fraction quite free from the higher homologue.

The first fraction on hydrolysis readily yields the crystalline pilopic acid, but the second and third fractions only yield an oil which could not be crystallised.

By distillation of the second fraction in a vacuum, it was possible to

* Pinner and Kohlhammer's formula.

effect a separation into three fractions, similar to those previously described, thus proving it to be a mixture of the two homologous esters.

Pilopic Acid, $C_7H_{10}O_4$.

This acid, which was crystallised with considerable difficulty, was prepared as follows. The ethyl ester boiling at 290—300° was hydrolysed either by aqueous potassium hydroxide or by 40 per cent. sulphuric acid, and the acid liquid saturated with ammonium sulphate and extracted by ether. The ethereal solution of the acid was next extracted with sodium carbonate solution, and the alkaline liquid subsequently acidified with sulphuric acid, saturated with ammonium sulphate, and extracted by ether. The ethereal extract was washed with water, dried over calcium chloride, and distilled. The syrupy residue, after standing for several days in a vacuous desiccator over sulphuric acid, with frequent stirring, became pasty and almost solid. It was spread on a porous tile, and the crystalline crust thus obtained recrystallised several times from hot benzene. After several recrystallisations, it separated in silky plates melting constantly at 104° (corr.). The acid can also be crystallised from water, in which, however, it is very soluble. On analysis:

0·09 gave 0·1748 CO_2 and 0·0526 H_2O. C = 53·0; H = 6·5.
0·1528 „ 0·297 CO_2 „ 0·0904 H_2O. C = 53·0; H = 6·57.
$C_7H_{10}O_4$ requires C = 53·2; H = 6·3 per cent.

The acid is dextrorotatory, and a determination of the specific rotation in aqueous solution gave the following result:

$$a_D^{15°} = +1·2°;\ l = 1 \text{ dcm.};\ c = 3·324;\ [a]_D^{15°} = +36·1°.$$

When excess of alkali is added to the acid solution, the specific rotation diminishes, a property also shown by pilocarpine and *iso*pilocarpine. A determination of the specific rotation in alkaline solution gave the following result:

$$a_D^{17°} = +0·3°;\ l = 1 \text{ dcm.};\ c = 9·5;\ [a]_D^{17°} = +3·2°.$$

The *methyl ester*, prepared in the usual way by means of sulphuric acid and methyl alcohol, is a colourless liquid boiling at 155—160° under 10 mm., and at 275° under 757 mm. pressure. On analysis:

0·1534 gave 0·315 CO_2 and 0·099 H_2O. C = 56·0; H = 7·2.
$C_8H_{12}O_4$ requires C = 55·8; H = 7·0 per cent.

In order to prove conclusively the correctness of the formula $C_7H_{10}O_4$, proposed for pilopic acid, the molecular weight of the methyl ester was determined. Although the percentages of carbon and hydrogen required for the formulæ proposed by the author and by

Pinner and Kohlhammer are not very dissimilar, there is a considerable difference in the molecular weights of the corresponding ester. The molecular weight was determined by the depression of the freezing point of benzene and of glacial acetic acid.

Solvent.	Wt. of ester.	Wt. of solvent.	Depression.	Mol. wt.
Benzene	0·3322	13·98	0·65°	179
Glacial acetic acid	0·3964	15·762	0·59	166

Molecular weight of $C_8H_{12}O_4$ = 172
" " $C_{10}H_{16}O_5$ = 216 (P. and K. formula)

These results, in addition to the analytical data recorded in this paper, conclusively prove the correctness of the formula $C_7H_{10}O_4$ previously ascribed to pilopic acid.

The *barium* salt of pilopic acid was prepared by digesting an aqueous solution of the acid with excess of barium carbonate, filtering, evaporating the filtrate to a low bulk, and precipitating with alcohol. The microcrystalline salt, dried at 120°, was analysed with the following result:

0·1406 gave 0·0722 $BaSO_4$. Ba = 30·2
$(C_7H_9O_4)_2Ba$ requires Ba = 30·4 per cent.

The *anilide*, $C_7H_9O{\cdot}NH{\cdot}C_6H_5$, was prepared by boiling the acid with three times its weight of aniline in a reflux apparatus for 24 hours. The liquid was poured into excess of dilute hydrochloric acid and the acid liquid extracted with ether. The ethereal extract was washed four times with dilute acid, finally with water, and dried over calcium chloride. After removal of the ether by distillation, the residue was placed in a vacuous desiccator over sulphuric acid and frequently stirred. In a short time, the oil became almost solid; the mass was spread on a porous tile and the dry powder then recrystallised from hot ether until of constant melting point. It was thus obtained in white, flat, pearly plates melting sharply at 110° (corr.). On analysis:

0·1124 gave 6·4 c.c. nitrogen at 22° and 764 mm. N = 6·3.
$C_{13}H_{15}O_3N$ requires N = 6·0 per cent.

The *strychnine* salt was prepared by boiling the aqueous solution of the acid with excess of strychnine and filtering. The filtrate was then evaporated in a vacuous desiccator over sulphuric acid and a very hygroscopic crystalline mass obtained, which was dissolved in a little hot alcohol. On standing, after the addition of ether, a quantity of strychnine separated, which was removed, and more ether added to the mother liquor. After long standing in a stoppered bottle, rosettes of crystals separated. These were dried and found to be very soluble in

water or alcohol and to melt at 120° (corr.); the aqueous solution was acid to litmus. On analysis:

0·186 gave 7·6 nitrogen at 22° and 764 mm. N = 4·5.
0·1606 „ 0·0832 anhydrous strychnine = 51·8.
$C_{21}H_{22}O_2N_2(C_7H_{10}O_4)_2$ requires N = 4·3; strychnine = 51·4 per cent.

This was therefore the acid salt of strychnine, formed by the separation of strychnine from the normal salt which was apparently first produced.

The *diamide* of the *hydroxy-acid* was prepared by mixing equal volumes of the methyl or ethyl ester of pilopic acid and strong aqueous ammonia. After standing for some hours with frequent shaking, the oil disappeared and the liquid solidified to a mass of crystals which was drained on a porous tile and recrystallised from hot alcohol until of constant melting point. The amide was sparingly soluble in cold water or alcohol, fairly so in hot water or alcohol, and almost insoluble in ether, benzene, or chloroform. The crystals melted at 160° (corr.), and on analysis yielded the following results:

0·0934 gave 0·167 CO_2 and 0·069 H_2O. C = 48·7; H = 8·2.
0·1206 „ 17·0 c.c. nitrogen at 14° and 764 mm. N = 16·4.
$C_7H_{14}O_3N_2$ requires C = 48·3; H = 8·1; N = 16·1 per cent.

Lactonic character of Pilopic Acid.—The correctness of the formula for pilopic acid having been demonstrated, experiments were next undertaken to obtain further proof of its lactonic character. Two grams of the acid were boiled with excess of baryta water for an hour, the solution saturated with carbon dioxide, and filtered. The filtrate was evaporated to a low bulk, acidified with the requisite quantity of sulphuric acid, and extracted with ether. The ethereal solution was washed with water, the ether spontaneously evaporated, and the residue placed overnight in a desiccator over sulphuric acid. It was then titrated with normal alkali, using phenolphthalein as indicator, with the following results:

0·52 required in the cold, 3·4 c.c. for neutralisation, and, when boiled with excess of alkali and titrated back with acid, 6·5 c.c.; this amount of an acid, $C_7H_{10}O_4$, requires 3·3 c.c. and 6·6 c.c. respectively. The substance was therefore the lactonic acid.

The *barium* salt of the *hydroxy-acid* was prepared by adding alcohol to a concentrated aqueous solution of the salt, prepared as just described. The precipitate was filtered off and dried on a porous tile. On analysis, the air-dried salt yielded the following result:

0·1404 gave 0·0982 $BaSO_4$. Ba = 41·1.
0·258 at 150° lost 0·0112 H_2O. H_2O = 4·4.
$C_7H_{10}O_5Ba,H_2O$ requires Ba = 41·6; H_2O = 5·5 per cent.

The specific rotation of the barium salt was determined in aqueous solution with the following result.

$$\alpha_D^{15°} = +0{\cdot}216°;\ l = 1 \text{ dcm.};\ c = 3{\cdot}512;\ [\alpha]_D^{15°} = +6{\cdot}1°.$$

The *silver* salt of the *hydroxy-acid*, prepared from the barium salt by interaction with silver nitrate, was a gelatinous precipitate, which, after drying on a porous tile, yielded, on analysis, the following result:

0·256 gave 0·1404 Ag. Ag = 55·0.

$C_7H_{10}O_5Ag_2$ requires Ag = 55·4 per cent.

Action of Ammonia on the Ethyl Esters (Middle Fraction).

The middle fraction of the ethyl ester obtained from the oxidation of *iso*pilocarpine (p. 1334) was shaken with an equal volume of strong ammonia, and a crystalline amide obtained melting at 151°. This was recrystallised many times from hot alcohol and from water, and a small quantity of an amide melting at 200° separated. As this proved to be identical with the amide obtained from the third fraction of the ester, it was set aside. The crystals from the mother liquors were recrystallised several times from alcohol and from water, and a product obtained melting at 161°, but analysis showed that it was not pure, so that it was not possible to separate the homologous amides completely by this method. In spite of all attempts to separate this middle fraction into its constituents, it could not be purified further than into two portions, one chiefly ethyl pilopate, and the other containing only a small quantity of this ester.

Homopilopic Acid, $C_8H_{12}O_4$.

The third fraction of the ethyl esters obtained by the oxidation of *iso*pilocarpine (p. 1334) gave, on analysis, results agreeing with those required for an ester of the formula $C_{10}H_{16}O_4$. On treatment with ammonia, a crystalline amide was obtained which melted at 199°. This *amide*, together with the portion melting at 200° from the second fraction of the ester, was recrystallised from water and from hot alcohol until of constant melting point. It was readily crystallised from hot water, and separated in well-defined prisms melting sharply at 208° (corr.) On analysis:

0·158 gave 0·2938 CO_2 and 0·1246 H_2O. C = 50·7; H = 8·7.

0·1172 „ 15 c.c. nitrogen at 20° and 769 mm. N = 14·6.

$C_8H_{16}O_3N_2$ requires C = 51·1; H = 8·5; N = 14·9 per cent.

As in the case of the pilopic ester, the diamide of the hydroxy-acid is formed in this reaction.

The *acid*, for which the name *homopilopic acid* is proposed, was obtained from the pure amide by heating with 20 per cent. hydrochloric acid in a sealed tube at 120°. The acid was extracted with ether, the ethereal solution washed with water, dried over calcium chloride, and distilled. The residual oil, after remaining in a vacuous desiccator over sulphuric acid for some days, showed no signs of crystallisation, and was therefore distilled in a vacuum. It boiled at 235—237° under 20 mm. pressure. On analysis:

0·141 gave 0·2864 CO_2 and 0·092 H_2O. C = 55·4; H = 7·2.
$C_8H_{12}O_4$ requires C = 55·8; H = 7·0 per cent.

The acid is dextrorotatory, and a determination of its specific rotation in aqueous solution gave the following result:

$\alpha_D^{21°} = +1·6°$; $l = 1$ dcm.; $c = 3·524$; $[\alpha]_D^{21°} = +45·4°$.

With excess of alkali, the specific rotation is diminished:

$\alpha_D^{21°} = +0·16°$; $l = 1$ dcm.; $c = 2·82$; $[\alpha]_D^{21°} = +5·9°$.

The acid was titrated with decinormal alkali, using phenolphthalein as indicator, with the following results:

0·185 required in the cold, 10·5 c.c. for neutralisation, and, when boiled with excess of alkali and titrated back with acid, 21·4 c.c.; this amount of an acid, $C_8H_{12}O_4$, requires 10·75 c.c. and 21·5 c.c. respectively.

The barium salts of the lactonic and hydroxy-acids were prepared in a manner similar to the corresponding salts of pilopic acid.

The *barium* salt of the *lactonic acid* is a very hygroscopic powder, which, dried at 150°, yielded the following result on analysis:

0·1614 gave 0·079 $BaSO_4$. Ba = 28·8.
$(C_8H_{11}O_4)_2Ba$ requires Ba = 28·6 per cent.

The *barium* salt of the *hydroxy-acid*, which is microcrystalline and stable in the air, contains 1 mol. of water of crystallisation. On analysis of the air-dried salt:

0·6094 at 150° lost 0·0314 H_2O. $H_2O = 5·1$.
0·3542 gave 0·2414 $BaSO_4$. Ba = 40·1.
$C_8H_{12}O_5Ba,H_2O$ requires Ba = 40·0; $H_2O = 5·3$ per cent.

Pinner and Kohlhammer's Piluvic Acid.

It having been proved that pilopic acid has not the same formula as piluvic acid, the question arises whether homopilopic and piluvic acids may not be identical. Although it is not possible to decide this question definitely, inasmuch as no physical constants have been recorded for piluvic acid by means of which it may be identified, and as no proof

has yet been adduced that the acid and its derivatives were pure products, yet there are a number of facts which suggest the possibility of the identity of these acids, or, at any rate, that piluvic acid consisted largely of homopilopic acid.

Forty grams of *iso*pilocarpine were oxidised with 127 grams of permanganate in the cold, and the ethyl ester isolated by the usual method. On distillation under 20 mm. pressure, three fractions were obtained boiling at 160—170°, 170—190°, and 190—240° respectively. The lowest fraction, distilled under 760 mm. pressure, gave a liquid boiling at 295°, which was analysed with the following results:

0·1996 gave 0·4126 CO_2 and 0·1308 H_2O. C = 57·6; H = 7·3.
$C_9H_{14}O_4$ requires C = 58·1; H = 7·5 per cent.

The liquid was therefore ethyl pilopate, so that, under the conditions of this oxidation, pilopic acid was formed.

Experiments were made, using larger quantities of permanganate than previously employed, in order to obtain pilopic acid free from homopilopic acid, but this purpose was not accomplished, although the amount of volatile acids appeared to increase. It is more probable, therefore, that in the oxidation of *iso*pilocarpine the molecule is attacked at two points, namely, at contiguous carbon atoms, with the formation of pilopic and homopilopic acids, rather than that homopilopic acid is first produced and then oxidised to pilopic acid. It is of course possible that both factors come into play when a large excess of permanganate is used. Pinner and Kohlhammer may therefore have been dealing with a mixture of pilopic and homopilopic acids, and their analytical results are in harmony with such a suggestion. Moreover, there is an experiment recorded by them which supports this view, namely, that in which an attempt was made to purify the amyl ester by distillation. In this case, the analytical results did not agree with any formula, and the authors admit the ester was impure; it was, however, prepared from the barium salt of the acid by a simple reaction, such as would lead to the formation of a product requiring only a single fractionation to obtain it in a state of purity. The fact that the ester was not pure throws doubt on the purity of the barium salt, although it is on the analysis of this salt, and of the acid derived from it, that the proof of the formula, $C_8H_{12}O_5$, given by Pinner and Kohlhammer for piluvic acid is partly based.

Experiments on the Constitution of Pilopic and Homopilopic Acids.

For the purposes of these experiments, the syrupy acid was used, analysis having shown that only a small proportion of homopilopic acid was present. Twenty-four grams of ethyl pilopate were treated with

phosphorus pentabromide, as already described (Trans., 1900, 77, 858), and 20 grams of an ester boiling at 180—190° under 10 mm. pressure obtained. This, when treated with diethylaniline, yielded 13 grams of a product which, when fractionated, first in a vacuum and then under the ordinary pressure, could be divided into two fractions: (1) 270—290°, a limpid oil; (2) 290—300°, a thicker oil, showing a tendency to deposit a waxy substance.

The first fraction proved to be ethyl pilopate, and by hydrolysis the crystalline pilopic acid was obtained from it. On analysis of the ester:

0·0946 gave 0·2016 CO_2 and 0·0672 H_2O. C = 58·1; H = 7·8.
$C_9H_{14}O_4$ requires C = 58·1; H = 7·5 per cent.

The second fraction, on analysis, yielded the following result:

0·1014 gave 0·2262 CO_2 and 0·0714 H_2O. C = 60·8; H = 7·8.
$C_{11}H_{18}O_4$ requires C = 61·7; H = 8·4 per cent.

It was not therefore the expected diethyl ester of the unsaturated acid. On hydrolysis, a syrupy acid was obtained which did not crystallise even when left in a vacuum over sulphuric acid at 0° for several days.

This acid yielded an amorphous silver salt containing 53·1 per cent. Ag ($C_7H_8O_4Ag_2$ requires Ag = 58·1 per cent.). It was oxidised with permanganate at 0°, and an oily acid with a rancid odour was isolated from the products of the reaction. The silver salt prepared from this acid contained 51·4 per cent. Ag, but the quantity obtained was too small to purify, and as it was evident that this amount of acid did not represent the main reaction, the experiment was abandoned.

Action of Hydriodic Acid on Pilopic Acid.—The acid was heated with three times its weight of fuming hydriodic acid in a reflux apparatus for several hours, but the pilopic acid was recovered unchanged. When heated in a sealed tube at 180°, no crystalline product was obtained, but only a small quantity of an oil having the odour of petroleum.

A quantity of the acid was heated to 210° in a sulphuric acid bath, but no change took place, no gas or water being given off.

Fusion of Pilopic Acid with Potassium Hydroxide.

Fusion at a High Temperature.—Five grams of the syrupy acid were mixed with 25 grams of potassium hydroxide and a few drops of water, and the mixture fused for a short time. The fused mass was dissolved in water, acidified with sulphuric acid, and distilled with steam. The distillate, which had a rancid smell, was extracted with ether, the ethereal solution dried over calcium chloride, and distilled.

The residue distilled completely at 110—160°, and the distillate was miscible with water. The barium salt was prepared and analysed:

0·2672 gave 0·2014 $BaSO_4$. Ba = 44·3.
$(C_4H_7O_2)_2Ba$ requires Ba = 44·1 per cent.

The calcium salt was also prepared, and obtained as white, pearly plates, which were dried on a porous tile, and, on analysis, yielded the following result:

0·127, air-dried, lost 0·0112 H_2O at 150°. H_2O = 8·8.
$(C_4H_7O_2)_2Ca,H_2O$ requires H_2O = 7·8 per cent.

Calcium *iso*butyrate crystallises with 4 mols. of water of crystallisation. Further proof of the identity of this salt with calcium *n*-butyrate was afforded by making a saturated aqueous solution of the salt at 0°, and placing the solution in warm water, when crystals separated which redissolved on cooling to 0°.

The silver salt was prepared, and, after recrystallising once from hot water, furnished the following result on analysis:

0·136 gave 0·0752 Ag. Ag = 55·3.
$C_4H_7O_2Ag$ requires Ag = 55·4 per cent.

The volatile acid formed is therefore *normal butyric acid.* No other acid could be isolated from the residue left after distillation.

Fusion at a Low Temperature.—Ten grams of acid were fused with 30 grams of potassium hydroxide and 5 c.c. of water, and kept gently simmering for some time. The fused mass was dissolved in water, acidified with sulphuric acid, and distilled with steam, but no volatile acid was obtained. The acid liquid was extracted with ether, the ethereal solution washed with water, dried over calcium chloride, and distilled. The residue weighed 6 grams, and on standing in a vacuum for some time, deposited a small quantity of crystals, which were separated, and drained on a porous tile. The non-crystalline residue was found to be unchanged pilopic acid. The residual liquid, after extraction with ether, was neutralised, evaporated to dryness, and extracted with alcohol. The alcohol was removed by distillation, the residue dissolved in a little water, and precipitated with lead acetate. The acid, regenerated from the lead salt by means of sulphuretted hydrogen, gave crystals mixed with unchanged pilopic acid, and when recrystallised from hot water came out in scales. When pure, it melted at 190° (corr.), losing water, and forming, apparently, the anhydride. On analysis:

0·0258 gave 0·05 CO_2 and 0·016 H_2O. C = 52·8; H = 6·9.
$C_7H_{10}O_4$ requires C = 53·2; H = 6·3 per cent.

The acid was almost insoluble in ether, and sparingly soluble in cold water; it decolorised permanganate solution.

The silver salt was obtained as a granular precipitate on adding silver nitrate to a solution of the ammonium salt of the acid. After washing, it was dried on a porous tile and analysed, with the following result:

0·116 gave 0·0674 Ag, 0·094 CO_2, and 0·0238 H_2O. Ag = 58·1; C = 22·1; H = 2·3.

$C_7H_8O_4Ag_2$ requires Ag = 58·1; C = 22·6; H = 2·1 per cent.

From a consideration of the properties of this acid, it would appear probable that it is an unsaturated acid closely allied to ethylitaconic acid, and that it is produced from the lactonic acid by a similar reaction to that generally brought about by sodium ethoxide.

By fusion at a low temperature, therefore, a small quantity of the isomeric unsaturated acid is formed, but the greater portion of the lactonic acid is recovered unchanged.

Fusion of Homopilopic Acid with Potassium Hydroxide and Formation of α-Ethyltricarballylic Acid.

A preliminary experiment having shown that a crystalline acid is formed by the fusion of homopilopic acid with potassium hydroxide at a moderate temperature, the whole of the syrupy homopilopic acid at disposal was fused with three times its weight of the alkali and a little water for half an hour; the mass was then dissolved in water, acidified with sulphuric acid, and distilled with steam. Only a trace of a volatile acid was obtained.

The acid liquid remaining after distillation was extracted with ether, the ethereal solution washed with water, dried over calcium chloride, distilled to a low bulk, and then poured into excess of benzene. On standing, crystals separated, which were filtered off and dried on a porous tile. From the acid liquid, after extraction with ether, more acid can be obtained through the lead salt, as previously described.

In this way about 3 to 4 grams of a crystalline acid were obtained. This *acid* melted indefinitely at about 117°, but after washing with hot benzene, fused at 145°. It was recrystallised from ether and from water until the melting point was constant, and was then obtained in hard prisms melting sharply at 157° (corr.). The acid is readily soluble in water or alcohol, and moderately so in ether, but on evaporating the ethereal solution an oil is obtained which gradually solidifies; it is insoluble in benzene or light petroleum. A 6 per cent. aqueous solution was optically inactive. On analysis:

0·1338 gave 0·23 CO_2 and 0·073 H_2O. C = 46·9; H = 6·1.
0·0922 „ 0·159 CO_2 „ 0·05 H_2O. C = 47·0; H = 6·0.
$C_8H_{12}O_6$ requires C = 47·1; H = 5·9 per cent.

The analytical results, melting point, and behaviour towards solvents agree with those recorded for *α-ethyltricarballylic acid*, and a mixture of equal parts of the synthetical acid and that obtained by fusion melted at 157° (corr.).

Further evidence as to its identity with α-ethyltricarballylic acid was afforded by an examination of the following derivatives, which agreed in all respects with those recorded in the following paper (p. 1346) for this acid.

The *anhydro-acid*, prepared by heating the acid with excess of acetyl chloride in a reflux apparatus for 2 hours, could only be obtained as an oil soluble in benzene and yielding with aniline an amorphous compound.

The *silver* salt was prepared by adding silver nitrate to an aqueous solution of the ammonium salt. The flocculent precipitate, insoluble in water, was well washed and dried, first on a porous tile, and then at 100°. On analysis:

0·1248 gave 0·0816 CO_2, 0·0246 H_2O, and 0·0768 Ag. C = 17·8; H = 2·2; Ag = 61·6.
0·1354 gave 0·0836 Ag. Ag = 61·7.
$C_8H_9O_6Ag_3$ requires C = 18·3; H = 1·7; Ag = 61·7 per cent.

The *calcium* salt was prepared by digesting the aqueous solution of the acid with an excess of calcium carbonate and filtering. The filtrate, when heated to 100°, became a firm jelly which liquefied on cooling. This behaviour has been shown to be very characteristic of calcium α-ethyltricarballylate. The solution of the calcium salt was evaporated to a low bulk in a vacuum and precipitated with alcohol. The precipitate was dried, first on a porous tile and then between blotting paper. On analysis, the air-dried salt furnished the following results:

0·1284 at 150° lost 0·0306 H_2O, and on ignition gave 0·031 CaO.
H_2O = 23·8; Ca = 17·3.
$(C_8H_9O_6)_2Ca_3,9H_2O$ requires H_2O = 23·7; Ca = 17·5 per cent.

The *copper* salt, prepared in the usual way with copper acetate, was a bluish-green precipitate insoluble in hot water. After drying on a porous tile and then at 120°, it furnished the following results on analysis:

0 0636 gave 0·0256 CuO. Cu = 32·1.
$(C_8H_9O_6)_2Cu_3$ requires Cu = 32·2 per cent.

Constitutional Formulæ of Pilopic and Homopilopic Acids.

The identification of α-ethyltricarballylic acid as a product of the fusion of homopilopic acid with potassium hydroxide, and the other facts recorded in this paper, render it possible to state with a high degree of probability the constitutional formulæ of both pilopic and homopilopic acids. The formation of acetic and propionic acids by the oxidation of *iso*pilocarpine and of normal butyric acid by the fusion of pilopic acid with potassium hydroxide, prove that the *iso*butyl group does not exist in *iso*pilocarpine, and that the formation of *iso*butyric acid by the fusion of the alkaloid with potassium hydroxide is probably due to the action of the fused alkali on the normal acid first formed. That *iso*pilocarpine must contain the *n*-butyl grouping follows from the formation of α-ethyltricarballylic acid,

$$C_2H_5\cdot CH(CO_2H)\cdot CH(CO_2H)\cdot CH_2\cdot CO_2H.$$

The formation of the tricarboxylic acid from the hydroxydicarboxylic acid (as potassium salt) can only be explained on the assumption that the $CH_2\cdot OH$ group is oxidised to CO_2H. The ethyltricarballylic acid may therefore be derived from one of the three following hydroxydibasic acids:

$$C_2H_5\cdot\underset{CH_2\cdot OH}{CH}\text{———}\underset{CO_2H}{CH}\cdot CH_2\cdot CO_2H \quad : \quad C_2H_5\cdot\underset{CO_2H}{CH}\text{——}\underset{CH_2\cdot OH}{CH}\cdot CH_2\cdot CO_2H \quad :$$

$$C_2H_5\cdot\underset{CO_2H}{CH}\text{——}\underset{CO_2H}{CH}\cdot CH_2\cdot CH_2\cdot OH \quad .$$

On account of the stability of homopilopic acid, it is most reasonable to assume that it is a γ-lactonic acid. From the above formulæ, four γ-lactonic acids may be derived:

(1).

$$C_2H_5\cdot\underset{CH_2\cdot O\cdot CO}{CH}\text{——}\underset{}{CH}\cdot CH_2\cdot CO_2H$$

(2).

$$C_2H_5\cdot\underset{CO\cdot O\cdot CH_2}{CH}\text{—}\underset{}{CH}\cdot CH_2\cdot CO_2H$$

(3).

$$C_2H_5\cdot\underset{CO_2H}{CH}\text{——}\underset{CH_2\cdot O\cdot CO}{CH}\text{——}CH_2$$

(4).

$$C_2H_5\cdot\underset{CO_2H}{CH}\text{——}\underset{CO\cdot O\cdot CH_2}{CH}\text{—}CH_2$$

If we regard pilopic acid as derived from homopilopic acid by the loss of carbon dioxide and oxidation of the contiguous carbon atom, only formulæ (1) and (2) are possible, as (3) and (4) would yield acids containing less than seven carbon atoms.

The formation of pilopic acid during the oxidation of *iso*pilocarpine may be assumed to be due either to oxidation of the homopilopic acid first formed, or, as has been previously pointed out, to simultaneous

oxidation at two different points in the molecule. Since, however, both acids contain the *n*-butyl group, it is most probable that they bear the relation to each other previously suggested, since oxidation must have occurred at that portion of the molecule containing the nitrogen atoms. If this argument be admitted, the possible formulæ for pilopic acid will be,

(1). (2).

$$C_2H_5\cdot CH\text{——}CH\cdot CO_2H \quad \text{or} \quad C_2H\ CH\text{—}CH\cdot CO_2H$$
$$\quad\ CH_2\cdot O\cdot CO \qquad\qquad\quad CO\cdot O\cdot CH_2$$

but of these (1) is clearly not admissible as it is a malonic acid derivative and should therefore lose carbon dioxide on heating at 200°, which, as has been shown, is not the case. There remains therefore for pilopic acid the second formula, and the corresponding formula for homopilopic acid is

$$C_2H_5\cdot CH\text{—}CH\cdot CH_2\cdot CO_2H$$
$$\quad\ CO\cdot O\cdot CH_2$$

Lactonic acids corresponding to these formulæ have not hitherto been prepared, but pilopic acid resembles ethylparaconic acid in many of its physical properties, and the relation between the two is shown by their formulæ:

$$C_2H_5\cdot CH\text{—}CH\cdot CO_2H \qquad C_2H_5\cdot CH\text{—}CH\cdot CO_2H$$
$$\quad\ CO\cdot O\cdot CH_2 \qquad\qquad\quad O\cdot CO\cdot CH_2$$

Pilopic acid. Ethylparaconic acid.

It remains, therefore, to synthesise acids having the constitution assigned to pilopic and homopilopic acids, and to contrast the properties of the acids prepared synthetically with those recorded in this paper. Experiments with this end in view are now in progress.

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CXLII.—*A New Synthesis of α-Ethyltricarballylic Acid.*

By Hooper Albert Dickinson Jowett.

In the preceding paper, it was shown that by the fusion of homopilopic acid with potassium hydroxide an acid of the formula $C_8H_{12}O_6$ was formed and that the properties of this acid corresponded with those of α-ethyltricarballylic acid. As, however, two different melting points have been assigned to this acid and as no derivatives have been described other than the silver salt, it was necessary to prepare the

substance and to examine it more fully, so as to be able to identify completely the acid formed from homopilopic acid with the synthetical compound.

α-Ethyltricarballylic acid was first prepared by Auwers (*Ber.*, 1891, 24, 307, 2897) by the condensation of ethyl ethylsodiomalonate with ethyl fumarate and subsequent hydrolysis. The acid, after recrystallisation, melted at 147—148° and was readily soluble in water, but only moderately so in ether; on evaporation, the ethereal solution left an oil which became crystalline. The substance was prepared more recently by Michael (*Ber.*, 1900, 33, 3745) by the same method, but he found that after further purification it melted at 155—157°.

In order to prepare the acid for the purpose of comparison, it was deemed advisable to accomplish the synthesis by an entirely different method, as this at the same time would afford additional proof of the correctness of the constitution of the synthetical acid prepared by Auwers.

Ethyl α-cyano-β-ethylsuccinate was prepared by the general method of preparation of the alkyl-substituted succinic acids described by Bone and Sprankling (Trans., 1899, 75, 839), by condensing the sodium compound of ethyl cyanoacetate with ethyl α-bromobutyrate.

The sodium compound of ethyl α-cyano-β-ethylsuccinate was then condensed with ethyl bromoacetate with the formation of ethyl β-cyano-α-ethyltricarballylate. This stage of the synthesis may be presented by the following equation:

$$CH_2Br\cdot CO_2Et + CO_2Et\cdot CNa(CN)\cdot CH(C_2H_5)\cdot CO_2Et = CO_2Et\cdot CH_2\cdot C(CN)(CO_2Et)\cdot CH(C_2H_5)\cdot CO_2Et + NaBr.$$

The ethyl cyanoethyltricarballylate, on hydrolysis, yielded an acid from which carbon dioxide was eliminated by heating at 180° with the formation of α-ethylcarballylic acid.

These condensations took place readily, the yield of cyano-esters in the first and second stages of the synthesis being 50 and 70 per cent. respectively of the calculated amounts. As these cyano-esters have not been previously prepared, their boiling points and densities were determined.

α-Ethyltricarballylic acid as thus prepared was found to agree in its general properties with those previously recorded, and after recrystallisation melted at 157° (corr.), thus confirming the figure given by Michael. Of its derivatives described later, the calcium salt is very characteristic; it is fairly soluble in cold water, but on warming the aqueous solution, the liquid is converted into a solid jelly, which liquefies on cooling. This gelatinisation of the calcium salt, coupled with the melting point of the acid, affords the best means of identifying the substance.

Experimental.

Ethyl α-Cyano-β-ethylsuccinate, $CO_2Et\cdot CH(CN)\cdot CH(C_2H_5)\cdot CO_2Et$.

This ester was prepared as follows:—28·5 grams of ethyl cyanoacetate were mixed with a solution of 5·75 grams of sodium in 70 grams of absolute alcohol, and to the resulting thick paste 48·75 grams of ethyl α-bromobutyrate were added. The mixture, which became warm, was heated on a water-bath until neutral, which generally required 2—3 hours; it was then cooled, poured into water, and the oil which separated was extracted with ether. The ethereal solution was washed with water, dried over calcium chloride, and then distilled. The residual oil was fractionated under 20 mm. pressure, when a quantity of unchanged ethyl cyanoacetate and ethyl α-bromobutyrate distilled below 100°, but the greater portion of the liquid distilled at 160—170°, leaving only a very small residue in the flask. On refractionation, the portion distilling at 160—170° was collected. Of this a portion boiling constantly at 167—168° was set aside for analysis; it had a density d 15°/15° = 1·0647. The yield of product boiling at 160—170° under 20 mm. pressure was 50 per cent. of the theoretical. On analysis:

0·1338 gave 0·285 CO_2 and 0·0916 H_2O. C = 58·1; H = 7·6.
0·1512 „ 9·4 c.c. nitrogen at 24° and 764 mm. N = 6·8.
$C_{11}H_{17}O_4N$ requires C = 58·1; H = 7·5; N = 6·2 per cent.

The correctness of the formula ascribed to the cyano-ester was proved by the fact that on hydrolysis the compound furnished ethylsuccinic acid melting at 98°, which on analysis yielded the following result:

0·132 gave 0·2372 CO_2 and 0·086 H_2O. C = 49·0; H = 7·2.
$C_6H_{10}O_4$ requires C = 49·3; H = 6·9 per cent.

The characteristic calcium ethylsuccinate was also prepared.

Ethyl β-Cyano-α-ethyltricarballylate,
$CO_2Et\cdot CH(C_2H_5)\cdot C(CN)(CO_2Et)\cdot CH_2\cdot CO_2Et$.

This cyano-ester was prepared as follows:—To 6 grams of sodium dissolved in 60 grams of absolute alcohol, 45·4 grams of ethyl-α-cyano-β-ethylsuccinate were added. The mixture became warm and very viscid, but no solid separated; after cooling, 33·4 grams of ethyl bromoacetate were added, when a reaction at once took place with evolution of heat and separation of sodium bromide. The mixture was heated for half-an-hour on a water-bath, cooled, and then poured into water. The oil which separated was extracted with ether, the ethereal solution washed with water, dried with calcium chloride, and distilled. The residual oil was carefully fractionated under 20 mm. pressure. Scarcely

any liquid distilled below 195°, the greater portion came over between 195° and 215°, leaving a small residue in the flask. On refractionation, the portion distilling at 205—208° under 17—21 mm. pressure was collected, the greater portion, boiling constantly at 208° under 21 mm. pressure, being put aside for analysis and for the determination of its density. The yield of refractionated product was 70 per cent. of the theoretical. On analysis:

0·1716 gave 0·3602 CO_2 and 0·1144 H_2O. C = 57·3; H = 7·4.
0·1756 „ 7·5 c.c. nitrogen at 24° and 764 mm. N = 4·7.
$C_{15}H_{23}O_6N$ requires C = 57·5; H = 7·3; N = 4·5 per cent.

The density at 16° compared with water at 16° was 1·0972.

α-Ethyltricarballylic Acid, $CO_2H{\cdot}CH(C_2H_5){\cdot}CH(CO_2H){\cdot}CH_2{\cdot}CO_2H$.

This acid was obtained from the ethyl cyanoethyltricarballylate in the following manner. The ester was first boiled with five times its weight of 40 per cent. aqueous sulphuric acid in a reflux apparatus until all the oily drops had disappeared, which generally required a period of 24—48 hours. The acid liquid was then saturated with ammonium sulphate and extracted with ether six times. The ethereal extract was distilled to a convenient bulk and extracted with dilute sodium carbonate solution. This was acidified with sulphuric acid, saturated with ammonium sulphate, and extracted with ether. The ethereal solution was washed with water, dried with calcium chloride, distilled to a low bulk, and then poured into ten times its volume of benzene. On standing, crystals separated which were filtered off; these, when dried, melted at 117° with effervescence. The acid was then heated in a small flask in a paraffin bath at 180° until all evolution of gas had ceased. After cooling, the mass was dissolved in a little hot water, boiled with animal charcoal, and filtered. On standing and stirring with a glass rod, the solution became pasty. The crystals were drained on a porous tile and recrystallised from a little water until of constant melting point; thus purified, they melted at 156—157° (corr.). The pure acid required about 20 times its weight of ether to dissolve it, but on evaporating this solution at the ordinary temperature, no crystals separated until all the ether had evaporated, when an oil was left which gradually became solid. On analysis:

0·1974 gave 0·341 CO_2 and 0·106 H_2O. C = 47·1; H = 6·0.
0·198 „ 0·342 CO_2 „ 0·1064 H_2O. C = 47·1; H = 6·0.
$C_8H_{12}O_6$ requires C = 47·1; H = 5·9 per cent.

The *anhydro-acid* was prepared by adding the acid to three times its weight of acetyl chloride and boiling for 2 hours in a reflux apparatus. The acid slowly dissolved and the excess of acetyl chloride was removed

by evaporation on the water-bath and subsequent standing in a vacuous desiccator over solid potassium hydroxide. In this way, a hard, sticky varnish was obtained, which could not be crystallised. On analysis :

0·2218 gave 0·4146 CO_2 and 0·1108 H_2O. C = 51·0 ; H = 5·5.
$C_8H_{10}O_5$ requires C = 51·6 ; H = 5·4 per cent.

The anhydro-acid dissolved in benzene ; on the addition of aniline to the solution, heat was developed, the liquid immediately became cloudy, and an amorphous precipitate separated which, however, could not be obtained crystalline.

The *barium* salt was prepared by digesting an aqueous solution of the acid with excess of barium carbonate until neutral, and filtering. On warming the filtrate, crystals appeared which, however, redissolved on cooling. The filtrate was evaporated to a small bulk, filtered while hot, and the crystals dried, first on a porous tile and finally between blotting paper. Analysis showed that the air-dried crystals contained 7 mols. of water of crystallisation, of which six are removed by heating at 180° :

0·3156 at 180° lost 0·036 H_2O and gave 0·234 $BaSO_4$. H_2O = 11·4 ; Ba = 43·6.
0·1716 gave on combustion 0·0526 H_2O. H = 3·4.
$(C_8H_9O_6)_2Ba_3,7H_2O$ requires H_2O = 13·4 ; Ba = 43·8 ; H = 3·4 per cent.

The *calcium* salt was prepared by digesting an aqueous solution of the acid with excess of calcium carbonate and filtering. The filtrate had the remarkable property of becoming a firm jelly when heated to 100°, and liquefying on cooling. The liquid was evaporated to a small bulk in a vacuum and the calcium salt precipitated with alcohol in a microcrystalline form. On analysis :

0·1886 at 150° lost 0·044 H_2O and on ignition gave 0·0454 CaO.
H_2O = 23·3 ; Ca = 17·2.
$(C_8H_9O_6)_2Ca_3,9H_2O$ requires H_2O = 23·7 ; Ca = 17·5 per cent.

The *copper* salt, prepared by adding copper acetate solution to an aqueous solution of the ammonium salt of the acid and boiling, was a greenish-blue, flocculent precipitate. On analysis :

0·1972 at 150° lost 0·0272 H_2O and on ignition gave 0·0696 CuO
H_2O = 13·8 ; Cu = 28·2.
$(C_8H_9O_6)_2Cu_3,5H_2O$ requires H_2O = 13·2 ; Cu = 28·0 per cent.

The *triethyl ester*, prepared in the usual manner by the action of sulphuric acid and ethyl alcohol on the acid, was a limpid, colourless liquid boiling at 170—175° under 16 mm. pressure. On analysis, it furnished the following results :

0·2136 gave 0·457 CO_2 and 0·1574 H_2O. C = 58·3 ; H = 8·2.
$C_{14}H_{24}O_6$ requires C = 58·3 ; H = 8·3 per cent.

An attempt to prepare the amide by mixing the ester with excess of strong ammonia was unsuccessful.

Addendum.—Since this paper was in type the author has received a communication from Dr. Bone pointing out the following passage in a paper, "Researches on the Alkyl Substituted Succinic Acids," by Bone and Sprankling (Trans., 1899, **75**, 864): "Finally, we are studying the interaction of the sodium derivative of ethylic cyanosuccinates and the ethylic salts of α-bromo-fatty acids."

As the purpose for which reference was made to the paper in question only necessitated the reading of that portion of it relating to the details of preparation of the analogous ethyl cyanomethylsuccinate, the concluding paragraph containing the passage referred to most unfortunately escaped my attention.

The necessity for the preparation of α-ethyltricarballylic acid and certain of its derivatives was due to the great importance of identifying beyond question the acid formed by the fusion of homopilopic acid with potassium hydroxide.

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XLIX.—*The Constitution of Pilocarpine. Part IV.*

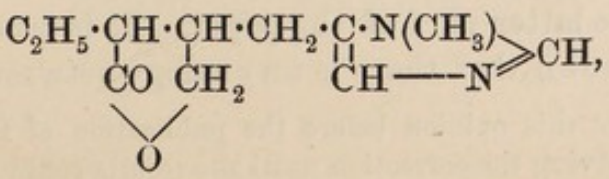
By Hooper Albert Dickinson Jowett.

In previous papers on this subject (Trans., 1900, **77**, 494, 851; 1901, **79**, 580, 1331), in which various substances obtained by the bromination and oxidation of *iso*pilocarpine were described, the discussion of the results was reserved for a future communication, as it was desired to obtain as much experimental evidence as possible before proposing any formulæ for the alkaloids, pilocarpine and *iso*pilocarpine.

In the last paper, in particular, when the structure of the greater part of the molecule was rendered obvious by the determination of the constitution of homopilopic acid, the discussion of these results in their relation to the constitution of the alkaloid was postponed, as it was hoped to afford conclusive proof of the correctness of the proposed formula for homopilopic acid by its synthesis. Although several attempts have been made to accomplish this, the results so far have been unsuccessful.

Before stating the results of the present investigation it appears necessary to comment briefly on a paper by Pinner and Schwarz (*Ber*, 1902, **35**, 2441) in which they suggest a constitutional formula for pilocarpine. This formula is based on (1) the constitution of homopilopic acid and (2) certain analogies shown to exist between pilocarpine derivatives and glyoxaline, especially with regard to the behaviour of the quaternary ammonium compounds towards alkalis. The possibility of the existence of a glyoxaline complex in pilocarpine was suggested by the subtraction of the homopilopic residue from the empirical formula of pilocarpine and by the formation of methylurea during oxidation.

It is on these grounds that Pinner and Schwarz suggest the following formula for pilocarpine:



leaving the question of the isomerism between pilocarpine and *iso*pilocarpine an open one.

Although the experimental results recorded in this paper show that the above formula is possibly correct, yet the evidence from which Pinner and Schwarz deduced their formula is open to criticism.

Whilst these authors have confirmed my statements regarding the formation of homopilopic acid by the oxidation of *iso*pilocarpine, they have not proved that this acid is produced when pilocarpine is oxidised. The diamide of the acid obtained by them from pilocarpine by oxidation melted at 182°, whilst the diamide of homopilopic acid melts at 208°. Therefore either the acid produced by the oxidation of pilocarpine is isomeric and not identical with homopilopic acid, or the diamide obtained by Pinner and Schwarz was impure. The experiments recorded in this paper show that the latter is the correct explanation, and that when pilocarpine is oxidised, homopilopic acid, identical in all respects with that previously described, is produced. Pinner and Schwarz have also stated that certain conclusions made by me with regard to the existence of the :NH group in *iso*pilocarpine are incorrect,* and offer another explanation of the facts. I have proved that their explanation is correct by preparing directly from *iso*pilocarpine methiodide the picrate previously supposed to be methyl *iso*pilocarpine picrate. The compound thus previously described should therefore be named *iso*pilocarpine methyl picrate.

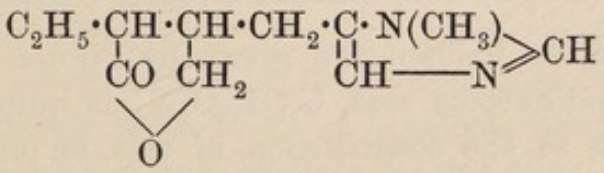
In former papers I showed that *iso*pilocarpine and pilocarpine contain no ordinary double bond, and further experiments recorded in this paper demonstrate the extraordinary stability of *iso*pilocarpine towards reducing agents, and also prove its mono-acidic character, which had previously been shown to be due to the nitrogen atom other than the methylamine residue.

Pinner and Schwarz seem to have overlooked these facts, as they describe (*Ber.*, 1902, **35**, 204, 2443) similar experiments to prove that bromine forms substitution and not additive compounds, and make no mention of my results. Moreover, I gave the most conclusive proof of the composition of the dibromo-compounds by reducing them to the original alkaloids (Trans., 1901, **79**, 601). It is unfortunate that Pinner and Schwarz were not able to adduce more conclusive proof of the formation of methylurea by the oxidation of pilocarpine. If, however, a small amount of this base were formed it would not necessitate the existence of a glyoxaline ring in the pilocarpine molecule, as it might be equally well produced from the betaine formula mentioned in the latter part of the paper. It is true that Pinner and Schwarz have shown that there is an analogy between certain reactions

* I had arrived at this opinion before the publication of Pinner and Schwarz's paper, but was reserving the correction until the results could be fully discussed.

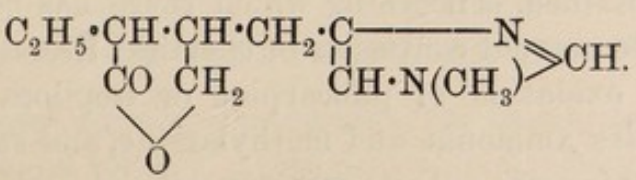
of pilocarpine and its derivatives and those of glyoxaline, yet, as is shown in the latter part of this paper, there are several other formulæ equally probable, including the isomerides dependent on the point of attachment of the non-nitrogenous group to the nitrogen ring. In consideration of all these points, and in view of the experimental results recorded in this and the following paper (p. 464), I am able to confirm with a certain reservation the formula for pilocarpine suggested by Pinner and Schwarz.

The constitution of homopilopic acid having been determined, the problem of the nature of the remainder of the molecule has been attacked from two points. Firstly, by preparing substances containing possibly the same group as *iso*pilocarpine, and studying their behaviour with certain reagents in order to determine if there was any analogy between them and this alkaloid. The substances thus studied were 1 : 4(or 1 : 5)-dimethylglyoxaline and 1 : 3-dimethylpyrazole, and the results are recorded in the following paper. Secondly, by endeavouring to obtain from *iso*pilocarpine substances containing the nitrogen ring intact, which has been accomplished by distillation with soda lime. From the crude product thus obtained 1-methylglyoxaline, 1 : 4(or 1 : 5)-dimethylglyoxaline, 1 : 4(or 1 : 5)-methylamyl glyoxaline, and probably 1 : 4(or 1 : 5)-methylamyleneglyoxaline have been isolated and identified. The identification of the first three substances has been rendered conclusive by the preparation and analysis of crystalline derivatives and by their products of oxidation. It may be mentioned that the aurichloride of the substance described in an early paper (Trans., 1900, **17**, 853) as methylpyridine was undoubtedly dimethylglyoxaline. The analytical figures agree equally well for the latter substance, and a small amount of impurity would account for the low melting point. The bearing of these and previous results on the constitution of *iso*pilocarpine is fully discussed, and the following formula proposed for *iso*pilocarpine. It will be seen that I is identical with that suggested by Pinner and Schwarz,



I.

or



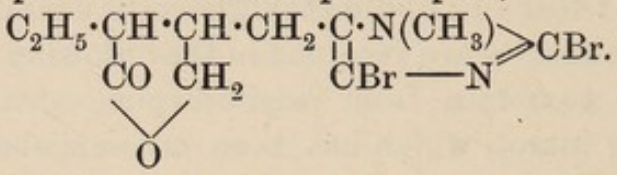
II.

The difficulty in deciding between these two formulæ is due to the fact that the constitution of the 4(or 5)-methylglyoxaline of Gabriel and Pinkus (*Ber.*, 1893, **26**, 2205) cannot be stated with certainty. The determination of the constitution of the latter will conclusively decide between formulæ I and II.

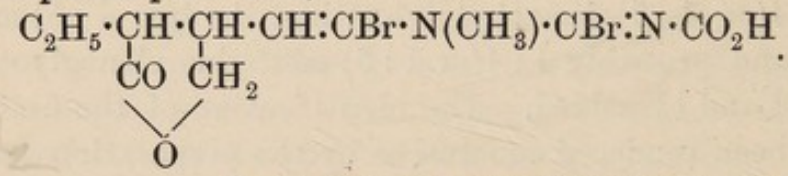
The relationship of pilocarpine to *iso*pilocarpine is considered, and the conclusion arrived at is that they are stereoisomerides, the asymmetric carbon atom involved being that contiguous to the carboxyl residue. This conclusion is strongly supported by the fact that the absorption spectra of the two alkaloids are absolutely identical.

The formulæ for the following substances are suggested after consideration of their reactions; they are based on formula I for *iso*pilocarpine, and if II should prove correct will require slight modification.

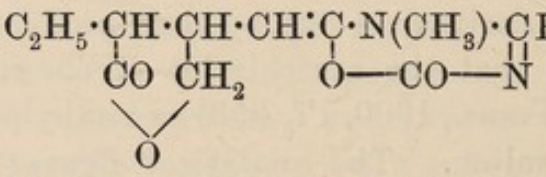
Dibromopilocarpine or dibromo*iso*pilocarpine,



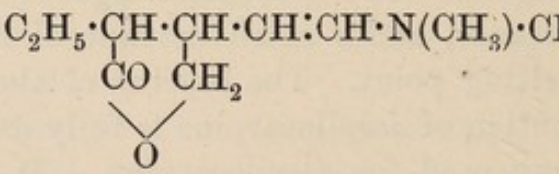
Dibromo*iso*pilocarpinic acid,



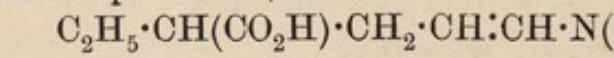
*iso*Pilocarpinolactone,



*iso*Pilocarpinic acid,



Bromocarpinic acid,



It is shown that the explanation given by Pinner and Schwarz of the formation of pilocarpoic acid, $C_{11}H_{16}O_5N_2$, is quite untenable, and no suggestions as to the constitution of this, or pilomalic acid, are offered.

It seems desirable at this stage of the inquiry to state briefly some of the results obtained, concerning which there has been some uncertainty, in order to prevent confusion in chemical literature. From the products of the oxidation of pilocarpine or *iso*pilocarpine with permanganate, besides ammonia and methylamine, and small amounts of acetic and propionic acids, pilopic and homopilopic acids—lactonic acids of the formula $C_7H_{10}O_4$ and $C_8H_{12}O_4$—have been isolated in amounts

varying with the conditions of experiment. These substances occur in the oxidation liquors as the potassium salts of the corresponding hydroxy-acids. These acids may be considered to be the lactonic acids of α-hydroxymethyl-β-ethylsuccinic and β-hydroxymethyl-α-ethylglutaric acids, though complete proof of this can only be obtained by synthesis. The acid obtained by Pinner and Kohlhammer by the oxidation of pilocarpine, which was first named by them *piluvic* acid, $C_8H_{12}O_5$, and subsequently *homopilomalic* acid, $C_8H_{14}O_5$, was undoubtedly homopilopic acid, though the salts examined by them were those of the corresponding hydroxydibasic acid. As this hydroxy-acid is unstable, immediately losing water with formation of the lactonic acid, and as Pinner and Schwarz in their last paper definitely accept my conclusions regarding this acid, the names *piluvic* and *homopilomalic* acid may be abandoned. The acid, $C_7H_{12}O_5$, obtained by Pinner and Kohlhammer by the oxidation of pilocarpoic acid, which was first named *iso*hydrochelidonic acid and subsequently pilomalic acid, appears to be a distinct acid, and not the hydroxy-acid of pilopic acid; it should therefore be considered as an acid of unknown constitution.

With regard to Pinner and Schwarz's suggestion to alter the name of the acid, first described by me, from dibromo*iso*pilocarpinic to dibromo*iso*carpoic acid on account of its possible confusion with dibromo*iso*pilocarpic acid, I see no reason for such a change, and would prefer to retain the original name for this acid. The confusion has arisen by an incorrect translation of these names into German, the acid obtained from dibromo*iso*pilocarpine and corresponding to the acid of which *iso*pilocarpine is the lactone is dibromo*iso*pilocarpic acid or "dibromo*iso*pilocarpinsäure," whereas dibromo*iso*pilocarpinic acid would be "dibromo*iso*pilocarpininsäure," and not "dibromo*iso*pilocarpinsäure" as rendered by Pinner and Schwarz (compare also *Chem. Centr.*, 1901, i, 1059). The difference in terminology would thus be the same in German as in English, and no new name is required.

Finally, in view of the researches which have resulted in establishing the constitution of pilocarpine (compare Feist, *Arch. Pharm.*, 1902, 240, 3), it would seem important that the widely quoted statements of Hardy and Calmels regarding this alkaloid, including its alleged synthesis by them, should now cease to receive scientific recognition.

Experimental.

Distillation of iso*Pilocarpine with Soda-lime.*

After several preliminary experiments, the best method of procedure was found to be as follows. One part of *iso*pilocarpine nitrate was mixed with eight times its weight of a mixture of equal parts of soda-lime and slaked lime and the mixture then placed in an iron tube,

which fitted into an ordinary combustion furnace, replacing the bed on which the glass combustion tube usually rests. The tube was closed at each end by caps, which could be unscrewed, and contained iron delivery tubes at each end. It was filled so that a clear space was left above the powder, the air displaced with hydrogen, the tube then gradually heated, and the gases, &c., evolved passed into aqueous hydrochloric acid. When the whole length of tube had been heated and no more gas was evolved, hydrogen was passed through. The acid solution was then boiled with a little animal charcoal, filtered, and extracted with ether; the acid liquid was then made decidedly alkaline with excess of caustic alkali, extracted several times with ether, the ethereal solution washed with water, dried over potassium carbonate and distilled.

The residue, which was a dark-coloured, strongly smelling liquid, was next distilled under diminished pressure, and the portion distilling between 100° and 200° under 10 mm. pressure collected. The yield of distillate was about 17 per cent. of the base taken. It was then distilled several times under 10 mm. pressure and the following fractions collected:

(1) B. p. 100—120°, 1 part. (2) B. p. 120—150°, 1 part. (3) B. p. 150—160°, 5 parts.

These fractions were then further examined.

Fraction b. p. 100—120° *under* 10 *mm. Pressure.*

This was a colourless liquid with the characteristic odour of a glyoxaline, rapidly becoming brown, and immiscible with water. An attempt to distil it under the ordinary pressure was not very successful, and though the chief portion boiled at 210—215°, decomposition occurred, and a large proportion of the whole was left behind in the flask as a thick, red oil.

The distillate was analysed:

0·1406 gave 0·3418 CO_2 and 0·108 H_2O. C = 66·3; H = 8·5.

$C_5H_8N_2$ requires C = 62·5; H = 8·3 per cent.

The analytical figures and its behaviour towards various reagents (*vide infra*) showed that this fraction, despite its fairly constant boiling point, was far from pure.

Fraction b. p. 120—150° *under* 10 *mm. Pressure.*

This fraction, which was very small for such a wide range of temperature, was so evidently a mixture that it was not further examined. It

became oxidised and discoloured much more rapidly than either of the other fractions.

Fraction b. p. 150—160° *under* 10 *mm. Pressure.*

This was by far the largest fraction, and was a thick, yellow liquid which slowly became discoloured, but was much more stable than either of the other fractions. A portion, which boiled at 158—160° under 10 mm. pressure, was analysed with the following result:

0·213 gave 0·5610 CO_2 and 0·176 H_2O. C = 71·8; H = 9·2.
0·1956 „ 0·5148 CO_2 „ 0·160 H_2O. C = 71·8; H = 9·1.
0·075 „ 12·8 c.c. nitrogen at 17° and 745 mm. N = 19·0.
$C_9H_{14}N_2$ requires C = 72·0; H = 9·3; N = 18·7 per cent.
$C_9H_{16}N_2$ „ C = 71·1; H = 10·5; N = 18·4 „

Although the analytical figures agree remarkably well for those required for the formula $C_9H_{14}N_2$, an amount of impurity might be present, as in the first fraction, to cause the slight variation from the numbers required for the formula $C_9H_{16}N_2$. Other experiments, shortly to be described, show that the fraction contained substances corresponding to both of the above formulæ.

Identification of 1-*Methylglyoxaline.*

In view of the fact that the lowest members of the glyoxaline series are not readily extracted from their aqueous solution by ether, the alkaline liquid after extraction with ether (p. 465) was neutralised, evaporated to a low bulk, and treated by the method described for the isolation of methylglyoxaline. The ethereal solution left, after removal of the ether by distillation, a liquid which distilled completely at 200—205°. This colourless liquid had the peculiar odour characteristic of the glyoxalines, was miscible with water, alcohol, or ether, and was quite stable:

0·1882 gave 0·4146 CO_2 and 0·134 H_2O. C = 60·1; H = 7·9.
$C_4H_6N_2$ requires C = 58·5; H = 7·3 per cent.
$C_5H_8N_2$ „ C = 62·5; H = 8·3 „

The analytical figures indicating a mixture of methyl- and dimethylglyoxalines, the picrate and platinichloride were prepared and examined.

Platinichlorides.—These were prepared by fractional precipitation with platinic chloride.

The first fraction melted at 239° and on analysis:

0·1028 gave 0·0334 Pt. Pt = 32·5.
$(C_5H_8N_2)_2,H_2PtCl_6$ requires Pt = 32·4 per cent.

The fifth fraction melted at 195° and on analysis:

0·0678 gave 0·0228 Pt. Pt = 33·6.

$(C_4H_6N_2)_2,H_2PtCl_6$ requires Pt = 33·9 per cent.

The intermediate fractions melted between 195° and 239°.

The *picrates* were prepared by precipitating a solution of the hydrochloride with picric acid. The crystalline precipitate melted at 157°, but after frequent recrystallisation two fractions were obtained melting at 158° and 167° respectively, and further crystallisation did not affect these melting points; 1-methylglyoxaline boils at 197—199°, its platinichloride melts at 190—191°, and its picrate at 158° (*Ber.*, 1889, **22**, 1359).

The constants of dimethylglyoxaline and its salts agree with these recorded in the next section of the paper.

The fraction b. p. 200—205° contained, therefore, 1-*methylglyoxaline* and *dimethylglyoxaline*.

Identification of 1 : 4(or 1 : 5)-*Dimethylglyoxaline.*

The hydrochloride of the fraction boiling at 100—120° under 10 mm. pressure, does not crystallise, and the platinichloride and aurichloride, when prepared in the usual way, undergo reduction, but the former double salt was obtained as follows. The oil was shaken up with three times its volume of water and the aqueous layer separated and converted into the hydrochloride. On adding platinic chloride a yellow crystalline precipitate was obtained which was separated and examined. When prepared in this way the platinichloride had no tendency to undergo reduction.

Dimethylglyoxaline platinichloride, when crystallised from hot water, separated in orange-coloured, cubical crystals, which melted at 238—239° with decomposition. The melting point was not affected by further recrystallisation. When mixed with an equal weight of 1 : 4(or 1 : 5)-dimethylglyoxaline platinichloride (see following paper) the mixture melted at 238—239°:

0·0678 gave 0·0222 Pt. Pt = 32·7.

0·104 „ 0·0336 Pt. Pt = 32·3.

0·1174 „ 0·0380 Pt, 0·087 CO_2, and 0·039 H_2O. Pt = 32·4; C = 20·2; H = 3·7.

0·112 gave 8·8 c.c. nitrogen at 17° and 745 mm. N = 8·9.

$(C_5H_8N_2)_2,H_2PtCl_6$ requires Pt = 32·4; C = 20·0; H = 3·0; N = 9·3 per cent.

Dimethylglyoxaline aurichloride was prepared from the hydrochloride obtained by decomposing the platinichloride with hydrogen sulphide. On adding auric chloride to the aqueous solution of the hydrochloride,

yellow acicular crystals separated, which, when collected and dried, first on a porous tile, and then in a desiccator over sulphuric acid, melted at 214—215°:

0·1096 gave 0·0492 Au, 0·0566 CO_2, and 0·0252 H_2O. Au = 44·9; C = 14·1; H = 2·5.

$C_5H_8N_2,HAuCl_4$ requires Au = 45·1; C = 13·8; H = 2·1 per cent.

The pure hydrochloride in 2 per cent. aqueous solution was optically inactive.

Dimethylglyoxaline picrate, prepared from the pure hydrochloride by precipitation with picric acid, formed yellow acicular crystals which melted at 167°. The melting point was not affected by further recrystallisation from water or alcohol. When mixed with an equal weight of 1:4(or 1:5)-dimethylglyoxaline picrate (m. p. 167°), the mixture melted at 140—145°. This experiment was repeated several times with different specimens with confirmatory results.

Oxidation of the fraction boiling at 100—120° *with potassium permanganate.*—In order to obtain further proof of the presence of dimethylglyoxaline in this fraction, the whole liquid containing the portions both soluble and insoluble in water was oxidised with permanganate and the products of oxidation isolated and examined.

Three grams of oil required 15 grams of permanganate in 1 per cent. aqueous solution to produce a permanent colour, and the resulting liquid after removing the manganese peroxide by filtration was worked up in the usual manner.

The bases obtained were identified as ammonia and methylamine. The platinum salt of the latter base was analysed.

0·1758 gave 0·0732 Pt. Found Pt = 41·6. Calculated Pt = 41·3 per cent.

The acids obtained were acetic and butyric acids (the latter derived from the portion of the oil insoluble in water).

The silver salts were fractionally precipitated and analysed.

1st fraction. 0·061 gave 0·034 Ag. Ag = 55·7.

$C_4H_7O_2Ag$ requires Ag = 55·4 per cent.

The last fraction. 0·0582 gave 0·0374 Ag. Ag = 64·3.

$C_2H_3O_2Ag$ requires Ag = 64·7 per cent.

The analyses of the platinichloride and aurichloride prove the existence in the first fraction of a substance of the formula $C_5H_8N_2$, whilst its general properties and its oxidation to ammonia, methylamine, and acetic acid, prove that it is a dimethylglyoxaline with one methyl group attached to nitrogen.

It therefore remains to be decided to which of the three carbon atoms the other methyl group is attached. Of the three isomerides possible, two are known and have been characterised (compare p. 464).

The constitution of 1 : 2-dimethylglyoxaline is definitely established, but that of the other synthetical dimethylglyoxaline is still uncertain; the dimethylglyoxaline derived from *iso*pilocarpine is, however, *not* identical with it.

The following table shows the close similarity existing between the derivatives of these isomerides:

		1 : 2.	1 : 4 (or 1 : 5).	From *iso*pilocarpine.
Base.....	b. p.	205—206°	203°	210—215°
Aurichloride	m. p.	215°	215°	214—215°
Platinichloride	m. p.	230°	238—239°	238—239°
Picrate.....	m. p.	179°	167°	167°

The only difference between the 1 : 4(or 1 : 5)-isomeride and that from *iso*pilocarpine is that the mixture of equal weights of their picrates, each melting at 167°, melts at 140—145°.

The glyoxalines are, therefore, isomeric and not identical. The constituent of the fraction, b. p. 100—120° under 10 mm. pressure, and soluble in water, is therefore 1 : 4(or 1 : 5)-*dimethylglyoxaline*.

Identification of 1 : 4(*or* 1 : 5)-*Methylamylglyoxaline*.

The attempted preparation of a crystalline hydrochloride from the fraction b. p. 150—160° under 10 mm. pressure being unsuccessful, the platinichloride was prepared as follows. The base was dissolved in alcohol, made faintly acid with aqueous hydrochloric acid, and platinic chloride in aqueous solution added. A yellow, crystalline precipitate separated, which was allowed to remain in contact with the solution until the latter began to darken, the crystals were then rapidly collected, washed, and dried.

Methylamylglyoxaline platinichloride forms light brownish-coloured, tabular crystals which melt at 198°. The melting point was not altered by further crystallisation. On analysis:

0·172 gave 0·0466 Pt. Pt = 27·1.

0·1092 ,, 0·0298 Pt, 0·1254 CO_2, and 0·0468 H_2O. Pt = 27·3 C = 31·3; H = 4·8.

0·124 gave 0·034 Pt, 0·1418 CO_2, and 0·054 H_2O. Pt = 27·4; C = 31·2; H = 4·8.

$(C_9H_{16}N_2)_2,H_2PtCl_6$ requires Pt = 27·3; C = 30·3; H = 4·7 per cent.

The hydrochloride, obtained from the pure platinichloride by treatment with hydrogen sulphide, yielded a crystalline *picrate* melting at 134°, but an amorphous *aurichloride*. In 5 per cent. aqueous solution the hydrochloride was optically inactive.

The *methylamylglyoxaline*, obtained from the pure hydrochloride by

adding excess of potassium carbonate and extracting with ether, was a colourless, viscid oil with a feebly basic odour, insoluble in water:

0·1026 gave 0·2672 CO_2 and 0·0954 H_2O. C = 71·0; H = 10·3.
0·1092 „ 0·2844 CO_2 „ 0·0982 H_2O. C = 71·0; H = 10·0.
$C_9H_{16}N_2$ requires C = 71·1; H = 10·5 per cent.

Oxidation of the Fraction, b. p. 145—160° *under* 10 *mm., with Permanganate.*

This fraction, containing, besides methylamylglyoxaline, the substance boiling between it and dimethylglyoxaline and yielding butyric acid on oxidation, was oxidised with excess of permanganate, and the bases and acids isolated by the usual method.

The bases formed were identified as ammonia and methylamine; the platinichloride of the latter on analysis gave: Pt = 41·2 (calculated Pt = 41·3 per cent).

The acids formed were identified as *n*-hexoic and *n*-butyric acids.

The crude acids were purified by distillation, when three fractions were obtained: (1) a small fraction below 125°, chiefly ether, &c.; (2) 125—160°; (3) 160—190°. Only a slight residue was left in the distilling flask.

Fraction (2) smelt very strongly of butyric acid, whilst fraction (3) had a pleasanter smell and was more viscid.

Fraction (2), *b. p.* 125—160°.

The acid was converted into the barium salt and precipitated in two fractions with silver nitrate. The silver salts were analysed, with the following result:

1. 0·1176 gave 0·0656 Ag. Ag = 55·7.
2. 0·24 „ 0·1334 Ag. Ag = 55·6.
$C_4H_7O_2Ag$ requires Ag = 55·4 per cent.

The anilide, prepared in the usual way and recrystallised from ligroin, melted at 82°. *iso*Butyranilide melts at 102·5°, and butyranilide at 90°. The low melting point is due to the presence of the higher anilide which is sparingly soluble in ligroin, and is not easily separated. The corresponding acid was undoubtedly *n*-butyric acid.

Fraction (3), *b. p.* 160—190°.

The acid was converted into the silver salts by fractional precipitation in the usual manner, and on analysis:

1. 0·114 gave 0·0546 Ag. Ag = 47·9.
2. 0·0364 „ 0·0178 Ag. Ag = 48·8.

$C_6H_{11}O_2Ag$ requires Ag = 48·4 per cent.

The anilide, prepared in the usual way and purified by recrystallisation from ligroin until of constant melting point, melted at 94—95°; *n*-hexoanilide melts at 95°. The acid was therefore *n*-hexoic acid.

The analyses of the pure base and platinichloride, and the products of oxidation, prove that one constituent of this fraction is methylamylglyoxaline, and from the fact that it is formed with 1 : 4(or 1 : 5)-dimethylglyoxaline, it may be inferred that the substituent groups occupy the same position in each case. With regard to the more volatile constituent of the fraction yielding butyric acid on oxidation, the analysis of the fraction boiling at 145—160° under 10 mm. pressure would indicate that it has the formula $C_9H_{14}N_2$. In this case it might be regarded as *methylamyleneglyoxaline*,

$$C_2H_5\cdot CH_2\cdot CH{:}CH\cdot \begin{matrix} C \\ \| \\ CH \end{matrix} \cdot N(CH_3) \begin{matrix} \\ \\ \!\!-\!\!- N \end{matrix} \!\!> CH,$$

which, with permanganate, would oxidise at the double bond as well as the glyoxaline ring, giving *n*-butyric acid. The unsaturated nature of the compound would also account for its readiness to oxidise. This fraction (b. p. 145—160° under 10 mm. pressure) contains, therefore, 1 : 4(or 1 : 5)-*methylamylglyoxaline*, and probably 1 : 4(or 1 : 5)-*methylamyleneglyoxaline*.

*iso*Pilocarpine, when distilled with soda-lime, yields ammonia, methylamine, 1-methylglyoxaline, 1 : 4(or 1 : 5)-dimethylglyoxaline, 1 : 4(or 1 : 5)-methylamylglyoxaline, and probably 1 : 4(or 1 : 5)-methylamyleneglyoxaline.

Oxidation of iso*Pilocarpinolactone.*

In order to determine the constitution of dibromo*iso*pilocarpinic acid and of *iso*pilocarpinolactone, the latter was oxidised with permanganate, and the resulting product treated in the same manner as in the case of *iso*pilocarpine, in order to isolate the bases and acids formed.

Five grams of *iso*pilocarpinolactone (m. p. 83°) dissolved in water were oxidised with a slight excess (9 grams) of permanganate in 1 per cent. aqueous solution. At 12°, the colour of the permanganate was immediately discharged until the above quantity had been added, when the slight excess remained unchanged. In this respect the lactone differs from *iso*pilocarpine, where the colour is not discharged so readily. The amount of oxidising agent used agreed well with the following equation. Calculated 8·3 grams; required 9 grams.

$$C_{11}H_{14}O_4N_2 + 4O + 2H_2O \rightarrow C_7H_{10}O_4 + NH_2\cdot CH_3 + NH_3 + 3CO_2.$$

The bases obtained were identified as ammonia and methylamine The platinichloride of the latter was analysed.

0·109 gave 0·045 Pt. Found Pt = 41·3. Calculated Pt = 41·3 per cent.

The acid was isolated as the ethyl ester, which distilled completely at 160—170° under 20 mm. pressure; titration of this derivative showed that the acid was monobasic and lactonic.

After hydrolysis, 0·3886 ester required for neutralisation 2·2 c.c. *N*-alkali solution cold and 4·0 c.c. hot. Calculated for an acid $C_7H_{10}O_4$ the requisite quantities are 2·1 c.c. and 4·2 c.c. respectively.

On hydrolysis, the acid was obtained as a colourless oil which quickly became solid. The crystals, after crystallisation from hot benzene, melted at 104°, and on analysis:

0·1052 gave 0·2044 CO_2 and 0·0616 H_2O. C = 53·0; H = 6·5.
$C_7H_{10}O_4$ requires C = 53·2; H = 6·3 per cent.

The aqueous solution of the acid was dextrorotatory, and with barium carbonate yielded a barium salt which was analysed with the following result.

0·128 gave 0·066 $BaSO_4$. Ba = 30·3.
$(C_7H_9O_4)_2Ba$ requires Ba = 30·4 per cent.

The acid was therefore pilopic acid.

The oxidation products of *iso*pilocarpinolactone with permanganate solution are therefore ammonia, methylamine, and pilopic acid.

Oxidation of Pilocarpine with Potassium Permanganate.

Twenty grams of pure pilocarpine were oxidised with 63 grams of permanganate at the ordinary temperature, and the products of oxidation isolated as described under the oxidation of *iso*pilocarpine.

The ethyl ester obtained was distilled under 10 mm. pressure, and the following fractions separated:

1. 165—175° 2. 175—180° 3. Above 180°.

The first fraction was analysed with the following result:

0·128 gave 0·277 CO_2 and 0·0918 H_2O. C = 59·0; H = 8·0.
$C_{10}H_{16}O_4$ requires C = 60·0; H = 8·0 per cent.

These figures are almost identical with those previously obtained by Pinner and Kohlhammer (*Ber.*, 1900, **33**, 1424).

When the ester was treated with excess of strong aqueous ammonia, the crystalline diamide of homopilopic acid (m.p. 208°) was obtained.

The second fraction was hydrolysed and the acid obtained titrated; in the cold, 0·42 required 24·4 c.c. normal alkali solution for neutralisation, and, when boiled with excess of alkali and titrated back with acid,

47·6 c.c.: calculated for an acid, $C_8H_{12}O_4$, the requisite quantities are 24·4 c.c. and 48·8 c.c. respectively.

The specific rotation in aqueous solution was determined with the following result:

$\alpha_D = +2·1°$; $l = 1$ dcm.; $c = 4·78$; $[\alpha]_D = +43·9°$; homopilopic acid has $[\alpha]_D$ +45·4°.

The barium salt was prepared in the usual way by neutralisation with barium carbonate:

0·393 gave 0·1914 $BaSO_4$. Ba = 28·6.

$(C_8H_{11}O_4)_2Ba$ requires Ba = 28·6 per cent.

When treated with ammonia, the third fraction yielded the diamide m. p. 208°, which furnished the following data:

0·0976 gave 0·1834 CO_2 and 0·0764 H_2O. C = 51·2; H = 8·7.

$C_8H_{16}O_3N_2$ requires C = 51·1; H = 8·5 per cent.

A determination of its specific rotation gave the following result:

$\alpha_D = +0·2°$; $l = 1$ dcm.: $c = 0·934$; $[\alpha]_D = +21·4°$.

Pinner and Schwarz found for the amide (m. p. 206°) $[\alpha]_D = +20·8°$. The above results prove conclusively that the oxidation product of pilocarpine with permanganate at the ordinary temperature consists almost entirely of homopilopic acid.

Miscellaneous Experiments with iso*Pilocarpine.*

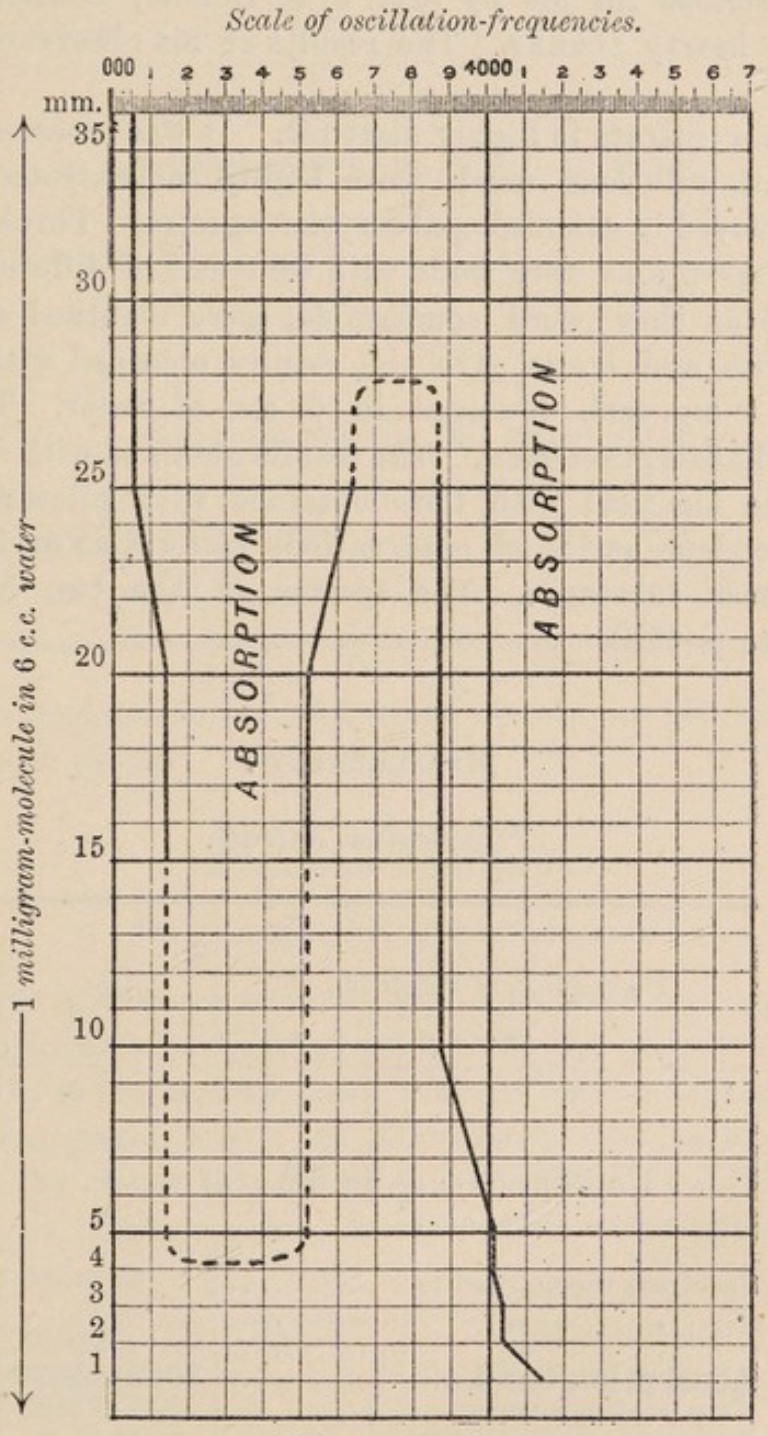
The extraordinary stability of *iso*pilocarpine towards reducing agents has already been noticed, even sodium in boiling amyl-alcoholic solution failing to attack it. An attempt was therefore made to reduce it electrolytically by Tafel's method (*Ber.*, 1900, **33**, 2209), but this was also unsuccessful.

Attempts were also made to form diacidic salts of *iso*pilocarpine, first, by heating with excess of methyl iodide in a sealed tube for 2 hours at 100°, and, secondly, by heating with concentrated hydrochloric acid at 180°, but in both cases only the normal mono-acidic products were obtained.

In view of the suggestion by Pinner and Schwarz that the compounds previously described as methyl*iso*pilocarpine derivatives were not such, but *iso*pilocarpine methyl compounds, picric acid was added directly to an aqueous solution of *iso*pilocarpine methiodide when a crystalline picrate was obtained (m. p. 136°), identical with that previously described as methyl*iso*pilocarpine picrate. As no change other than simple replacement of the iodine by the picric acid radical can be assumed, it follows that the substance described as methyl*iso*pilocarpine

picrate is really *iso*pilocarpine methyl picrate and that *iso*pilocarpine contains the :N and not the :NH residue.

A previous attempt to titrate *iso*pilocarpine not having given perfectly satisfactory results, the experiment was repeated:



Pilocarpine nitrate.

0·1316 when boiled with excess of caustic alkali and titrated back with acid required, 6·8 c.c. decinormal soda for neutralisation, with phenolphthalein as indicator, the calculated amount being 6·3 c.c. *iso*Pilocarpine behaved therefore as a normal lactone.

The Absorption Spectra of Pilocarpine Nitrate and iso*Pilocarpine Nitrate.*

The absorption spectra of pilocarpine and *iso*pilocarpine have been kindly determined for me by Prof. J. J. Dobbie, to whom I wish to express my hearty thanks. The results of his observations were as follows.

Pilocarpine nitrate is highly diactinic. It was necessary therefore to work with solutions much more highly concentrated than those usually employed in examining absorption spectra. Three independent series of observations were made with solutions of different strengths, and, so far as they were comparable, gave identical results. The measurements and curve give the results obtained with a solution containing 1 mg. mol. dissolved in 6 c.c. of water. There is one well-marked absorption band. The results obtained with *iso*pilocarpine nitrate were identical with those obtained with pilocarpine nitrate; three independent series of observations being also made in the case of the former substance. The spectra of the two nitrates were identical (see p. 452).

MEASUREMENTS.

Pilocarpine Nitrate.

Thickness of layer of liquid in millimetres.	Description of spectrum.	$^1/\lambda$.	λ.
	1 milligram-mol. in 6 c.c. of water.		
35	Spectrum transmitted to	3053	3275
30	,, ,, ...	3053	3275
25	,, ,, ...	3053	3275
	Absorption band.....	**3053** to **3633**	**3275** to **2752**
	Very weak spectrum.....	3633 to 3880	2752 to 2577
20 and 15	Spectrum transmitted to	3140	3185
	Absorption band.....	**3140** to **3520**	**3185** to **2841**
	Spectrum	3520 to 3880	2841 to 2577
10	Spectrum transmitted to ..... Very weak in position of absorption band.	3880	2577
5 and 4	Spectrum transmitted to ..... Weak in position of absorption band.	4008	2495
3 and 2	Spectrum transmitted to	4040	2475
1	,, ,, ...	4133	2419

Pilocarpine Nitrate (continued).

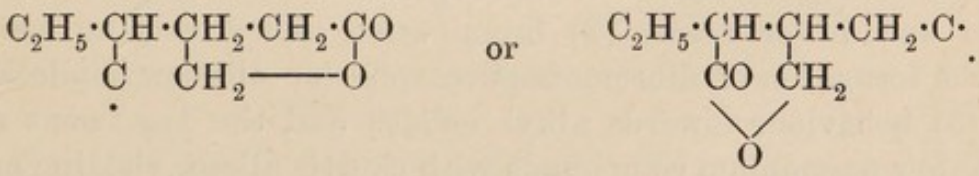
Thickness of layer of liquid in millimetres.	Description of spectrum.	$^1/\lambda$.	λ.
	1 milligram-mol. in 30 c.c. of water.		
4 and 3	Spectrum transmitted to	4133	2419
2	,, ,, ...	4183	2391
1	,, ,, ...	4250	2353
	1 milligram-mol. in 150 c.c. of water.		
5, 4 and 3	Spectrum transmitted to	4250	2353
2	,, ,, ...	4340	2304
1	,, ,, ...	4370	2288

The measurements for *iso*pilocarpine nitrate are identical with those for pilocarpine nitrate.

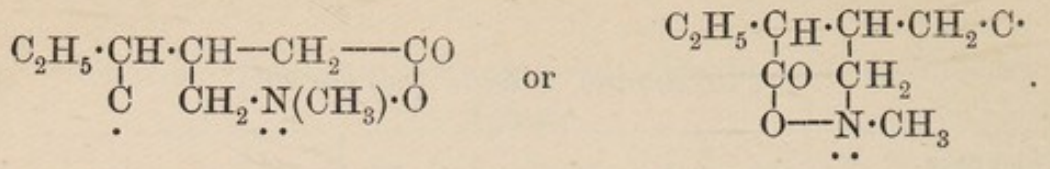
Discussion of Results.

The Constitution of iso*Pilocarpine.*

Since *iso*pilocarpine on oxidation with potassium permanganate yields the potassium salt of a hydroxy-acid, $C_8H_{14}O_5$, of which homopilopic acid, $C_8H_{12}O_4$, is the lactonic acid, and as the constitutional formula of the hydroxy-acid has been determined with a high degree of probability to be $C_2H_5\cdot CH(CO_2H)\cdot CH(CO_2H)\cdot CH_2\cdot CO_2H$, it is permissible to assume that *iso*pilocarpine contains one of the following complexes:

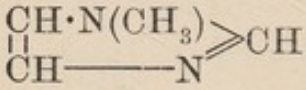


Since a formula containing either of these complexes would yield nomopilopic acid on oxidation, it follows that either formula would be equally valid. There is also a third possibility to consider in connection with a formula for *iso*pilocarpine, that is the existence of a betaine grouping in the molecule. If this were the case the following type of formula would account for the products of oxidation:



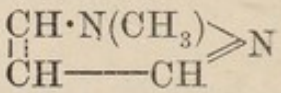
or

On oxidation the nitrogen atom is split off as methylamine and is replaced by hydroxyl, yielding homopilopic acid. In addition to the existence of one of the above complexes in the molecule the occurrence of the nitrogen atoms as N⁝ and ⁝N·CH_3 has been proved, and subtracting these groups from the empirical formula of *iso*pilocarpine, $C_{11}H_{16}O_2N_2$, the condition of two carbon and two hydrogen atoms remains to be explained. Assuming first that the two nitrogen atoms are present in the same complex, I have been able to construct only two possible formulæ for the group to be attached to the homopilopic residue, one being a glyoxaline and the other a pyrazole residue.



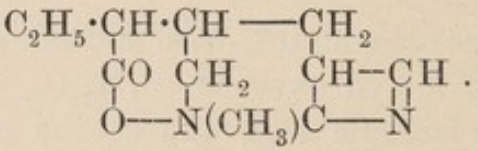
1-Methylglyoxaline.

or

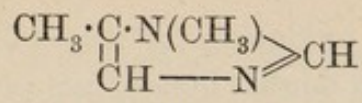


1-Methylpyrazole.

The homopilopic residue might be attached to any of the three carbon atoms in the ring by replacement of hydrogen, and the alkaloid would thus be a substituted glyoxaline or pyrazole. If, however, one of the nitrogen atoms, for example, that in the methylamine residue, is quinquevalent, the homopilopic complex must be attached to a ring containing one nitrogen atom, and, on these grounds, the following formula for the alkaloid might be suggested:

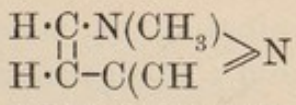


Besides these three possibilities there are also many others depending on the point of attachment of the two groups. The reactions of *iso*pilocarpine which have been most thoroughly studied are (1) oxidation with permanganate, yielding ammonia, methylamine, homopilopic and lower acids, (2) bromination under varying condition with the formation of dibromo*iso*pilocarpine or dibromo*iso*pilocarpinic acid, (3) behaviour towards alkyl iodides and the treatment of the quaternary ammonium compounds with caustic alkali, yielding methylamine and the respective alkylamine. These reactions would not enable us to decide between the three types of formulæ previously mentioned. In order to trace the analogy which might exist between *iso*pilocarpine and glyoxaline or pyrazole derivatives, the following dimethyl-compounds of the latter have been prepared and their reactions studied (see pp. 465, 467):



1 : 5(or 1 : 4)-Dimethylglyoxaline.

and



1 : 3-Dimethylpyrazole.

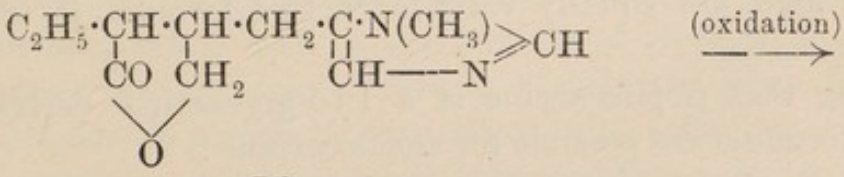
The results of the examination showed a remarkable analogy between the behaviour of *iso*pilocarpine and that of dimethylglyoxaline, and a striking dissimilarity between that of *iso*pilocarpine and dimethylpyrazole.

The results are shown in the following table:

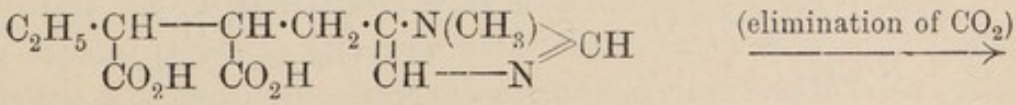
Reaction.	*Pyrazole.*	*Glyoxaline.*	iso*Pilocarpine.*
Oxidation with permanganate	Pyrazole carboxylic acid	Ammonia, methylamine and acetic acid	Ammonia, methylamine and homopilopic acid
Bromination	Dibromo-compound	Dibromo-compound	Dibromo-compound
Bromination at 100°	Dibromo-compound	Acid containing N and Br	Dibromo*iso*pilocarpinic acid
Methiodide with caustic potash at 180°	Very slight reaction	Methylamine and acetic acid	Methylamine and homopilopic acid

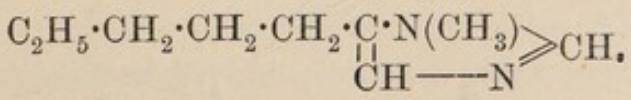
These results amplify and confirm the analogy existing between pilocarpine and glyoxaline derivatives, first pointed out by Pinner and Schwarz (*loc. cit.*), and exclude any formula for pilocarpine or *iso*pilocarpine containing a pyrazole ring. The glyoxaline formula for *iso*pilocarpine suggested by the above-mentioned analogy receives conclusive proof from the formation of various glyoxaline derivatives by distillation of the alkaloid with soda-lime. The crude product thus formed has been proved to contain 1-methylglyoxaline, 1 : 4(or 1 : 5)-dimethylglyoxaline, 1 : 4(or 1 : 5)-methylamylglyoxaline and probably 1 : 4(or 1 : 5)-methylamyleneglyoxaline together with ammonia and methylamine. Their formation is most readily explained by supposing that the (CH_2OH) group is oxidised to carboxyl with subsequent elimination of carbon dioxide yielding methylamylglyoxaline.

Assuming the following formula for *iso*pilocarpine, the change might be thus represented.



*iso*Pilocarpine.

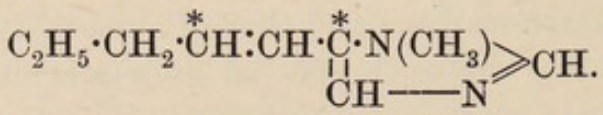




Methylamylglyoxaline.

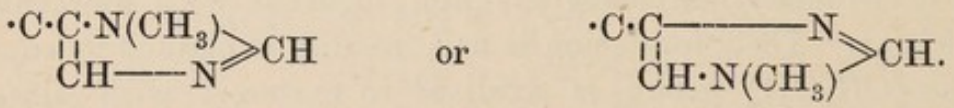
The unsaturated methylamyleneglyoxaline is probably produced by

the elimination of water prior to the oxidation of the (CH_2OH) group, and a change in the position of the double bond thus produced with subsequent oxidation and elimination of carbon dioxide, giving rise to the following glyoxaline.



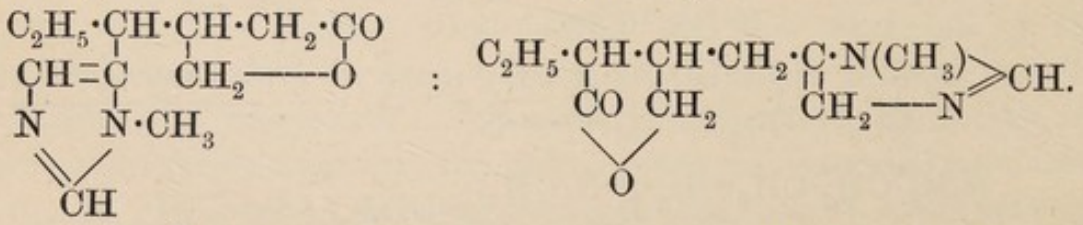
This compound oxidises at both the points marked with an asterisk, yielding butyric and carbonic acids, ammonia, and methylamine. The other glyoxalines, as well as ammonia and methylamine, are produced by further disruption of the molecule and the final fission of the glyoxaline ring itself. It has previously been shown that glyoxaline derivatives are very stable under similar conditions, for example, 2-methylglyoxaline was prepared by passing 1-methylglyoxaline through a red-hot tube (Wallach, *Ber*, 1883, **16**, 542). These facts, taken in conjunction with the analogies shown to exist between *iso*pilocarpine and glyoxaline derivatives, seem to afford conclusive evidence that *iso*pilocarpine is a glyoxaline derivative, and accordingly the betaine formula suggested for *iso*pilocarpine must be abandoned. The glyoxaline formula having been adopted, two problems remain for solution: (1) the determination of the point of attachment of the glyoxaline and homopilopic complexes, (2) choice of the appropriate formula for the homopilopic residue.

The identification of the dimethylglyoxaline formed with the 1 : 4- (or 1 : 5)-isomeride proves that one of the following complexes must exist in *iso*pilocarpine.



Inasmuch as it is impossible at the present time to decide between these formulæ, during the remainder of the discussion it will be assumed that *iso*pilocarpine is a 1 : 5-glyoxaline derivative. The arguments used are equally valid for a 1 : 4-formula, if this should prove to be correct.

Assuming that *iso*pilocarpine is a 1 : 5-glyoxaline derivative the following formulæ are possible for *iso*pilocarpine:

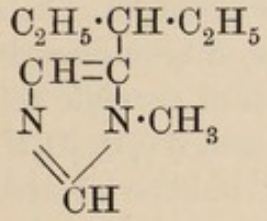


I. II.

The determination of the constitution of the methylamylglyoxaline formed renders it possible to decide between these two formulæ.

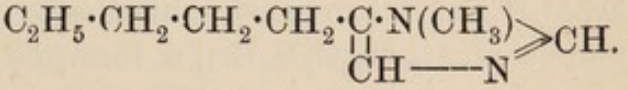
The formation of the methylamylglyoxaline can best be explained, as already indicated, by the oxidation of the (CH_2OH) group to carboxyl and subsequent elimination of carbon dioxide.

The methylamylglyoxaline thus derived from formula I or II will have the following constitution:



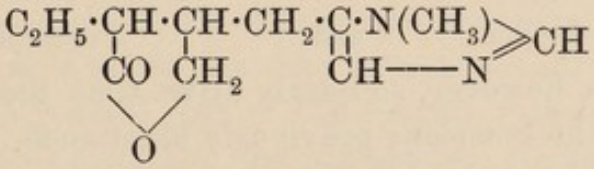
I.

or



II.

These compounds would yield on oxidation either (I) diethylacetic acid or (II) *n*-hexoic acid, besides ammonia and methylamine. The identification of the acidic product as *n*-hexoic acid justifies the adoption of formula II. From the experimental results recorded in this and previous papers and the deductions made therefrom, it is possible to propose the following constitutional formula for *iso*pilocarpine with a considerable degree of certainty, although complete proof can be furnished by synthesis alone. The formula given subject to the reservation previously mentioned.



*iso*Pilocarpine.

The Constitution of Pilocarpine.

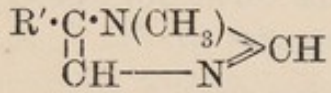
In former papers it was explained that, after having arrived at the constitution of *iso*pilocarpine, the more stable of the two alkaloids, it might be possible from a consideration of its formula and a comparison of the two alkaloids to pass to the formula of pilocarpine. This procedure seemed also necessary, since in certain reactions, for example, fusion with caustic potash or distillation with soda-lime, the products obtained would be those produced from *iso*pilocarpine and not from pilocarpine. Since *iso*pilocarpine is formed from pilocarpine by simple means, such as heating alone or with water, the relationship must be a close one. Furthermore, since pilocarpine, by oxidation with permanganate, at ordinary temperatures, yields homopilopic acid, identical in all respects with that obtained from *iso*pilocarpine, both alkaloids must contain this complex. The formation of homopilopic acid from pilocarpine might be thought to exclude the possibility of stereoisomerism, for the only asymmetric carbon atoms present are those in

the homopilopic complex and the glyoxalines obtained were optically inactive.

There are also two reactions in which the alkaloids apparently differ, which, as Pinner and Schwarz have pointed out, would favour the view of structural isomerism. These reactions are (1) the action of bromine under pressure, when, with pilocarpine, bromocarpinic acid, $C_{10}H_{15}O_4N_2Br$, is produced, whilst with *iso*pilocarpine, dibromo*iso*pilocarpinic acid, $C_{11}H_{14}O_4N_2Br_2$, is formed; (2) the action of chromic acid on pilocarpine leading to the formation of pilocarpoic acid, $C_{11}H_{16}O_5N_2$, whilst in the case of *iso*pilocarpine, no definite product could be isolated, and the action seemed to result in the general disruption of the molecule.

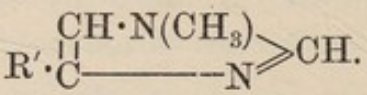
If the alkaloids are structural isomerides, the difference must lie in the point of attachment of the homopilopic complex to the glyoxaline ring, since both alkaloids must contain the homopilopic residue, and from the very close relationship of the alkaloids and their method of conversion it is almost certain that both contain a glyoxaline ring.

The relationship would be shown thus, where R′ = homopilopic complex:



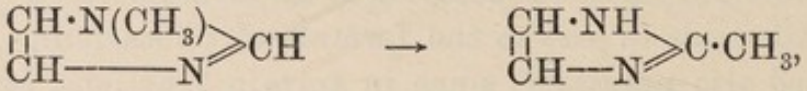
*iso*Pilocarpine

and



Pilocarpine.

These formulæ are, however, so nearly alike that they fail to explain the differences in the reactions previously mentioned. The mechanism of such an isomeric change is not easy to conceive, for, as it takes place by heat alone, it is most likely to occur by a migration either of the homopilopic residue to the contiguous carbon atom, or of the methyl group from one nitrogen atom to the other with rearrangement of the double linkings. A migration of the methyl group from the nitrogen to the contiguous carbon atom takes place in the formation of 2-methylglyoxaline by passing 1-methylglyoxaline through a red-hot tube,



but this cannot take place in the conversion of pilocarpine into *iso*pilocarpine, since both alkaloids contain the $:N\cdot CH_3$ group. The theory of structural isomerism therefore appears improbable.

If the alkaloids are stereoisomerides, this must be due to the asymmetric carbon atoms which occur only in the homopilopic residue. The formation of homopilopic acid from pilocarpine by oxidation can be readily explained when it is considered that the homopilopic acid is produced in the presence of free alkali and purified by the distillation of its ester, both of which factors would conduce to the formation of the stable modification.

With regard to the other two apparent differences, a satisfactory explanation can be offered of at least one of the reactions.

When bromine acts on pilocarpine or *iso*pilocarpine under pressure, the crystalline compound isolated in each case is different, but it by no means follows that the reaction does not proceed similarly in both cases. It may be that more than one substance is formed, and that they differ in their ability to crystallise, just as pilocarpine methiodide is amorphous whilst *iso*pilocarpine methiodide is crystalline. Further, I have previously shown (Trans., 1901, 79, 598) that there is experimental evidence that the reactions are similar, and that dibromopilocarpinic acid might be present in the mother liquors from which bromocarpinic acid has crystallised, just as bromo*iso*carpinic acid might be present in the filtrates from which dibromo*iso*pilocarpinic acid has been obtained. The yield of crystalline acid obtained is in harmony with this suggestion. The reaction of the alkaloids with chromic acid has not been sufficiently studied to permit of any deductions being made in support of either theory.

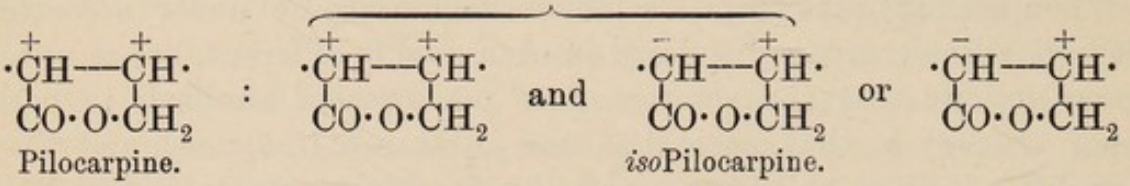
A consideration of the specific rotation of pilocarpine and its derivatives affords support both for the theory of the stereoisomerism of the alkaloids and also for the constitutional formula proposed. In the cases of pilocarpine, *iso*pilocarpine, homopilopic acid, and pilopic acids it has been noted that the specific rotation of the substances in alkaline solution is much less than in aqueous solution. This is in harmony with the general rule of the specific rotation of lactones in alkaline and aqueous solution, and the gradual change in the specific rotation of an aqueous solution of pilocarpine, from $+100°$ to $+77°$ is undoubtedly due to the opening of the lactone ring.

Three possible explanations as to the nature of the stereoisomerism may be briefly mentioned.

1. The fact that the conversion of pilocarpine into *iso*pilocarpine is accompanied by a change in the specific rotation from $+100{\cdot}5°$ to $+42{\cdot}8°$ suggests that partial racemisation may be the cause of the isomeric change. Of the two asymmetric carbon atoms in *iso*pilocarpine, one is attached to a carboxyl residue, whilst the other is attached to a methylene group. In accordance with general views on this subject, it may be assumed that the first of these asymmetric carbon atoms undergoes inversion, and that the optical activity of *iso*pilocarpine, homopilopic and pilopic acids is due to the carbon atom attached to the $-CH_2-$ group. Many examples of this type of change could be quoted, but it will be sufficient to refer to the example given by Pasteur of the change of quinine into quinicine.

2. That pilocarpine contains two non-equivalent asymmetric carbon atoms, and that in the formation of *iso*pilocarpine complete inversion of the asymmetric carbon atom attached to the carboxyl residue takes

place. The relation between pilocarpine and *iso*pilocarpine might then be represented as follows:



Pilocarpine. *iso*Pilocarpine.

In the latter case, the specific rotation of *iso*pilocarpine would be the difference between the rotatory powers of the two asymmetric carbon atoms, and *iso*pilocarpine would be incapable of resolution.

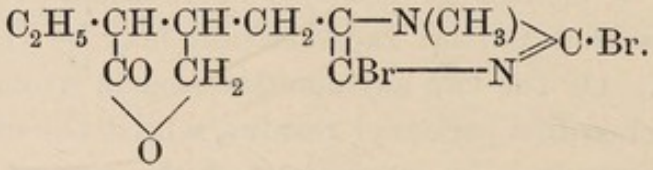
3. That, in addition to either of the above causes of isomerism, the bases may be also geometrical isomerides, corresponding to *cis*- and *trans*-modifications. The difference in the melting points of the salts and in the stability of pilocarpic and *iso*pilocarpic acids (Pinner and Schwarz, *Ber.*, 1902, **35**, 201) tends to support this view.

These conclusions are also supported by the fact that the alkaloids, pilocarpine and *iso*pilocarpine, give absolutely identical absorption spectra, and it has been shown (Hartley, *Phil. Trans.*, 1885, ii, 471) that stereoisomeric alkaloids give identical spectra, whilst any considerable difference in structure is accompanied by a corresponding difference in the absorption spectra.

In view of all these facts, it seems most probable that pilocarpine and *iso*pilocarpine are stereoisomerides, and must therefore be represented by the same structural formula.

Formulæ of various Derivatives of Pilocarpine and iso*Pilocarpine.*

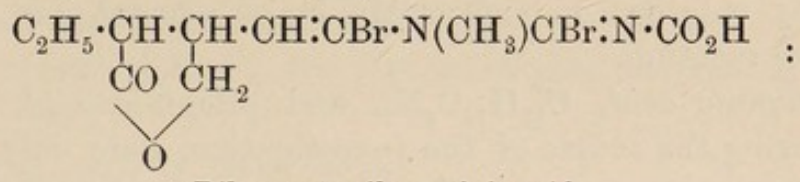
Dibromo-pilocarpine and iso*Pilocarpine.*—From the analogy with the glyoxalines examined there can be no doubt that substitution takes place in the glyoxaline ring, and the constitutional formulæ of each of these substances may be written:



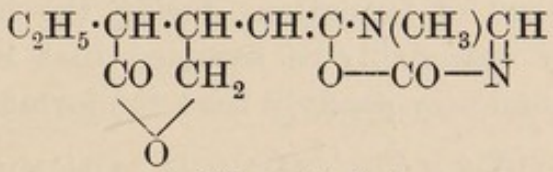
*Dibromo*iso*pilocarpinic Acid and* iso*Pilocarpinolactone.*—The constitution of these two substances can be proposed with a great degree of certainty from the formation of ammonia, methylamine, and pilopic acid by the oxidation of *iso*pilocarpinolactone and from the formation of pilopic acid by the action of sodium and alcohol on dibromo*iso*-pilocarpinic acid (Trans., 1901, **79**, 592).

The formation of pilopic acid proves the existence of the pilopic complex in these substances, and the oxidation must therefore have taken place in the glyoxaline ring.

The following formulæ are proposed:

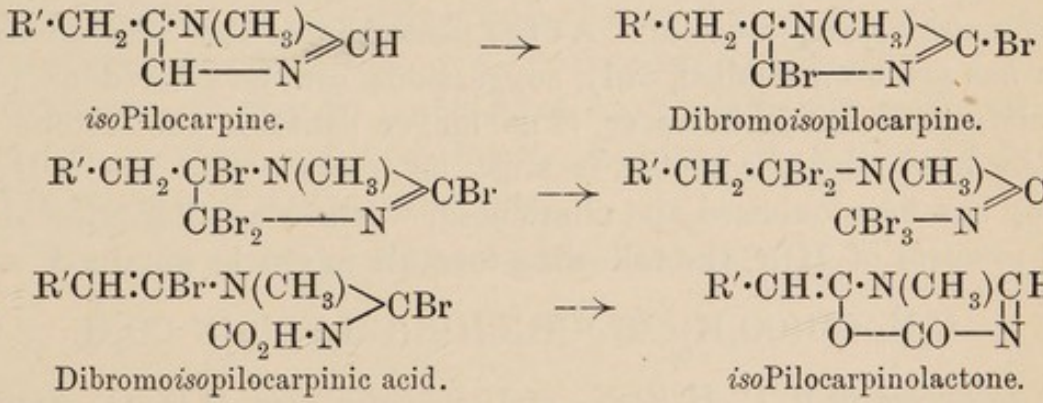


Dibromo*iso*pilocarpinic acid.



*iso*Pilocarpinolactone.

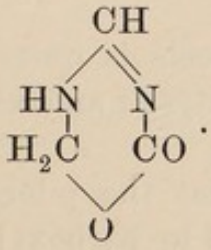
The mode of formation of these substances may be explained by assuming that bromination first takes place with formation of dibromo*iso*pilocarpine, which unites with excess of bromine with disruption of the double bond and formation of a tribromo-acid, this product then losing HBr, yielding dibromo*iso*pilocarpinic acid. The stages in the reaction may be expressed in the following manner, where R′ = the pilopic residue.



*iso*Pilocarpine. Dibromo*iso*pilocarpine.

Dibromo*iso*pilocarpinic acid. *iso*Pilocarpinolactone.

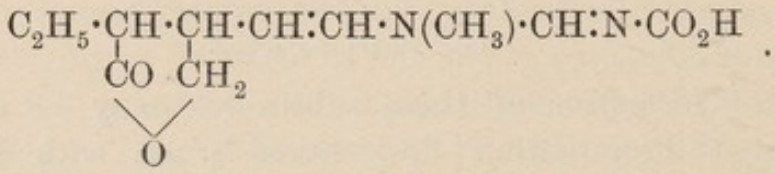
These formulæ explain perfectly the formation of pilopic instead of homopilopic acid on oxidation and the greater readiness to oxidise with permanganate, as well as the formation of ammonia and methylamine and the other characters of these compounds. If these formulæ are correct, and they are strongly supported by experimental evidence, then *iso*pilocarpinolactone must be regarded as the first member of a new type of ring compounds of which the parent substance would be



Preliminary experiments on the action of bromine under pressure on dimethylglyoxaline (p. 467) indicate that a similar reaction occurs with other derivatives of glyoxaline, and this change will be further

studied with these substances, as *iso*pilocarpinolactone cannot be conveniently obtained in sufficiently large amount for a complete study of the reaction.

iso*Pilocarpinic acid*, $C_{11}H_{16}O_4N_2$, and *pilopinic acid*, $C_8H_{11}O_4N$, obtained during the course of the investigation, were only isolated in very small amounts, and the first not even characterised. Having regard to its method of formation, during the bromination of *iso*-pilocarpine, it may reasonably be assumed that it is related to *iso*-pilocarpinolactone and may possibly have the formula



I can offer no suggestion as to the constitution of pilopinic acid, and the very small yield obtained precludes its further examination.

Bromocarpinic acid, $C_{10}H_{15}O_4N_2Br$, was obtained by Pinner and Kohlhammer by the action of bromine under pressure on pilocarpine, the reaction being similar to that yielding with *iso*pilocarpine, dibromo*iso*pilocarpinic acid. As the behaviour of this acid on oxidation has not been studied, only suggestions can be offered as to its constitution. Since, however, it no longer contains the lactone ring of the homopilopic complex, it must be assumed that the (CH_2OH) group has been oxidised and eliminated, when by an alternate addition and removal of HBr the following formula might be obtained.

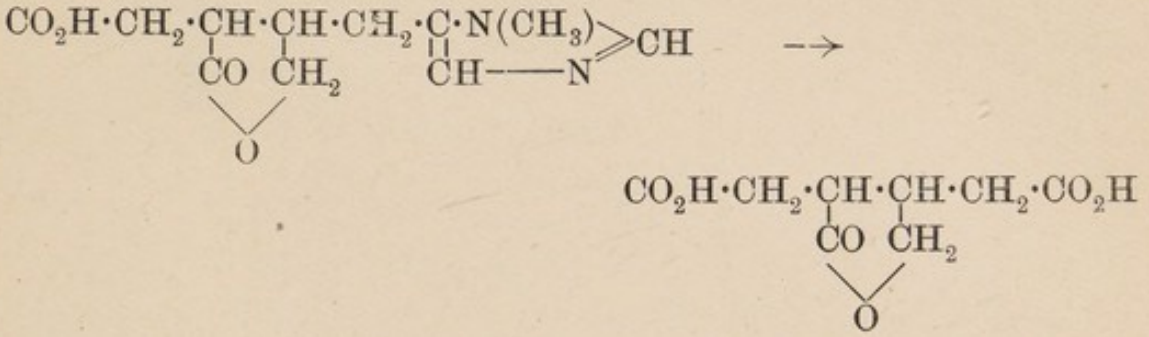
$$C_2H_5\cdot CH(CO_2H)\cdot CH_2\cdot CH{:}CH\cdot N(CH_3)\cdot CBr{:}N\cdot CO_2H.$$

Pilocarpoic acid, $C_{11}H_{16}O_5N_2$, and *pilomalic acid*, $C_7H_{12}O_5$, were obtained by Pinner and Kohlhammer, the former by the oxidation of pilocarpine with chromic acid, and the latter by oxidation of the pilocarpoic acid with permanganate. The only information available about the first of these acids is that it is dibasic towards baryta, but neither the acid nor its barium salt were obtained crystalline. The second acid is crystalline, dibasic, lævorotatory, and apparently non-lactonic, although the simple experiment of titration does not appear to have been made.

The formation of these acids is quite inexplicable if the formula proposed for pilocarpine by myself and also by Pinner and Schwarz is correct.

These authors suggest that the pilocarpoic acid is formed by the oxidation of a methyl group to carboxyl, but a consideration of the formula for pilocarpine shows that this explanation is quite untenable. There are only two methyl groups in pilocarpine: (1) the methyl of the homopilopic residue; (2) the methyl of the methyl-

amine residue. If the former were oxidised, pilocarpoic acid on oxidation with permanganate should yield a dibasic and lactonic acid having the formula $C_8H_{10}O_6$, the glyoxaline ring undergoing oxidation in the usual way:



This, however, is not the case, as pilomalic acid, $C_7H_{12}O_5$, is dibasic. If the methyl of the methylamine residue were oxidised, then pilocarpoic acid on oxidation with permanganate should yield homopilopic acid, which again does not occur. Furthermore, both of these reactions are, *a priori*, extremely unlikely.

The explanation of Pinner and Schwarz is therefore quite untenable, and in the absence of any information as to the reactions of these acids no suggestions as to their constitution can be offered.

In conclusion, it may be pointed out that pilocarpine and *iso*pilocarpine are the first and only known members of a new class of alkaloids, containing a glyoxaline ring.

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