

Chemical examination of gymnema leaves / by Frederick B. Power and Frank Tutin.

Contributors

Power, Frederick B. 1853-1927.
Tutin, Frank.
Royal College of Surgeons of England

Publication/Creation

London : Wellcome Chemical Research Laboratories, [1904]

Persistent URL

<https://wellcomecollection.org/works/sxvdra3y>

Provider

Royal College of Surgeons

License and attribution

This material has been provided by This material has been provided by The Royal College of Surgeons of England. The original may be consulted at The Royal College of Surgeons of England. where the originals may be consulted. You have permission to make copies of this work under a Creative Commons, Attribution license.

This licence permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited. See the Legal Code for further information.

Image source should be attributed as specified in the full catalogue record. If no source is given the image should be attributed to Wellcome Collection.



Wellcome Collection
183 Euston Road
London NW1 2BE UK
T +44 (0)20 7611 8722
E library@wellcomecollection.org
<https://wellcomecollection.org>

No. 48.

(23)

CHEMICAL EXAMINATION
OF
GYMNEA LEAVES

BY
FREDERICK B. POWER, PH.D.
AND
FRANK TUTIN

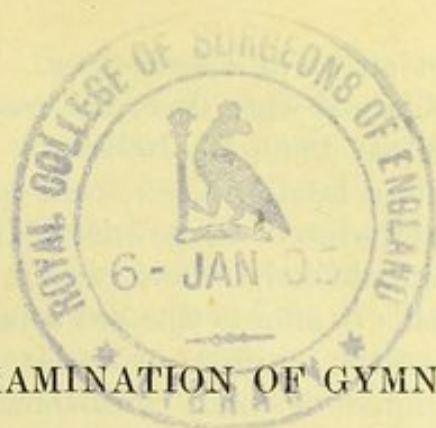


THE WELLCOME CHEMICAL RESEARCH LABORATORIES
FREDERICK B. POWER, PH.D., *Director*
6, King Street, Snow Hill
LONDON, E.C.

TITLES OF PUBLISHED PAPERS FROM
THE WELLCOME CHEMICAL RESEARCH LABORATORIES

1. SOME NEW GOLD SALTS OF HYOSCINE, HYOSCYAMINE, AND ATROPINE
2. THE CHARACTERS AND METHODS OF ASSAY OF THE OFFICIAL HYPOPHOSPHITES
3. NOTE ON THE MYDRIATIC ALKALOIDS
4. PREPARATION OF ACID PHENYLIC SALTS OF DIBASIC ACIDS
5. A NEW METHOD FOR THE ANALYSIS OF COMMERCIAL PHENOLS
6. THE ASSAY OF PREPARATIONS CONTAINING PILOCARPINE
7. PILOCARPINE AND THE ALKALOIDS OF JABORANDI LEAVES
8. A NEW GLUCOSIDE FROM WILLOW BARK
9. THE CONSTITUTION OF PILOCARPINE Part I.
10. THE COMPOSITION AND DETERMINATION OF CERIUM OXALATE
11. RESEARCHES ON MORPHINE—Part I.
12. OBSERVATIONS RELATING TO THE CHEMISTRY OF THE BRITISH PHARMACOPŒIA
13. MERCUROUS IODIDE
14. THE COMPOSITION OF BERBERINE PHOSPHATE
15. A CONTRIBUTION TO THE PHARMACOGNOSY OF OFFICIAL STROPHANTHUS SEED
16. THE CHEMISTRY OF THE JABORANDI ALKALOIDS
17. A NEW ADMIXTURE OF COMMERCIAL STROPHANTHUS SEED
18. RESEARCHES ON MORPHINE—Part II.
19. THE CONSTITUTION OF PILOCARPINE—Part II.
20. THE CHEMISTRY OF THE BARK OF ROBINIA PSEUD-ACACIA, *Linn.*
21. THE ANATOMY OF THE BARK OF ROBINIA PSEUD-ACACIA, *Linn.*
22. A SOLUBLE MANGANESE CITRATE AND COMPOUNDS OF MANGANESE WITH IRON
23. THE CHEMICAL CHARACTERS OF SO-CALLED IODO-TANNIN COMPOUNDS
24. THE CONSTITUTION OF PILOCARPINE—Part III.

(Continued on page 3 of cover)



CHEMICAL EXAMINATION OF GYMNEMA LEAVES.¹

BY FREDERICK B. POWER, PH.D., AND FRANK TUTIN.

Gymnema sylvestre, Br., from which the leaves under consideration are obtained, is a shrubby, climbing plant, belonging to the family of Asclepiadaceæ, and indigenous to Banda and the Deccan Peninsula (compare *Pharmacographia Indica*, 2, 450). Although various medicinal properties have been attributed to this plant by the Hindus, it was brought more prominently to notice several years ago in consequence of the observation that the leaves, when chewed, have the property of rendering imperceptible the sweet taste of sugar and other saccharine substances, and also, but in a less marked degree, the taste of many bitter substances.

The leaves appear to have been first chemically examined by Hooper (*Pharm. Journ.*, 1887, 17, 867, and *Chemical News*, 1889, 59, 159), and no subsequent publication is known to us in which the chemical nature of their constituents has been made the subject of further study.

In a summary of the investigation by Hooper (*loc. cit.*) the following principal products are stated to have been obtained:—

- (1) An ether extract, containing chlorophyll and resins.
- (2) An alcoholic extract, containing *gymnemic acid*, tartaric acid, glucose, a neutral bitter principle, resin, etc.
- (3) An aqueous extract, containing gum, glucose, carbohydrate, and extractive.

The above-mentioned peculiar properties of the leaves were attributed to the substance designated as “*gymnemic acid*,” which was stated to exist in them as a potassium salt, and to be best prepared by treating an aqueous solution of the alcoholic extract with a mineral acid, washing the precipitate, and drying

¹ Contributed to the British Pharmaceutical Conference at Sheffield, August, 1904, and reprinted from “*The Year-Book of Pharmacy*.”

it in a current of hot air or in a desiccator. The characters of the substance thus obtained were described essentially as follows (compare *Chemical News*, 1889, **59**, 159):—

“Gymnemic acid is a brittle, black, resinous substance, of a greenish colour when reduced to powder. It is insoluble in water, but soluble in alcohol, ether, benzol, and chloroform. With the caustic alkalis it affords fine red solutions, from which it is re-precipitated on the addition of acids. It fuses at about 60°C. into a black liquid of thick consistence; above 100° it gives off creosotic fumes, and, at a higher temperature, burns with a bright, smoky flame, leaving no ash. It is precipitated by the salts of lead, iron, silver, barium, and calcium, but not by tannin, picric acid, and gelatin solution. It forms insoluble salts with alkaloids, and this accounts for its masking the taste of quinine and other bitter substances. From analyses of the acid, the formula $C_{32}H_{55}O_{12}$ may be deduced.”

“The silver and lead salts of gymnemic acid form black powders, and the analysis of these affords evidence that the acid is monobasic, while the amount of alkali required for its neutralisation indicates a molecular weight corresponding to the above formula. The acid or its salts have not been obtained in anything approaching a crystalline condition; they dry as do tannic acid and the tannates. The acid is a glucoside. After boiling for about an hour with dilute acid, a dark resinous mass, devoid of the peculiar property of gymnema leaves, remains, and the liquid contains a body which readily reduces Fehling's solution.”

It has, furthermore, been noted by Hooper that “chloroform agitated with an alkaline solution of the leaf left a crystalline residue of a brownish colour; it had a bitter taste, and acted as a sialagogue. With the ordinary alkaloidal reagents it afforded coloured precipitates, but was a neutral principle.”

The action of the leaves of *Gymnema sylvestre* on the sense of taste was investigated several years ago by L. E. Shore, of Cambridge, whose results are recorded in a paper, entitled, “A Contribution to Our Knowledge of Taste Sensations” (*The Journal of Physiology*, 1892, **13**, 191–217). From a large number of experiments in this direction the following conclusions were drawn:—

“It is accepted that tastes may be divided into four classes, namely, sweet, bitter, acid, and salt, and as examples of these, glycerin, quinine sulphate, sulphuric acid, and sodium chloride

were employed. By the action of gymnema the sweet taste is very readily prevented in all regions of the tongue. The bitter taste is easily prevented, but not so readily as the former, especially at the back of the tongue. Acid taste, in dilute solutions, is not affected at all. Salt taste is very slightly, if at all, influenced. It therefore possesses a marked differentiating action. Solid saccharin, placed on the tongue, is tasteless after the action of gymnema; with a solution of quinine sulphate (1 : 1000) a slight acid taste is noticed at the tip of the tongue. The taste excited by the action of socotrine aloes is a pure bitter. Very dilute solutions of picric acid are intensely bitter at the back of the tongue, but after the action of gymnema even a saturated solution excites no sensation. The taste of a very large number of substances used in ordinary life is but little affected by gymnema, only such bitter and sweet components as are present being removed. It does not seem to have so ready or so powerful an action on bitter as on sweet taste; the reverse seems to be the case with cocaine. The action of gymnema on tactile perception, such as induction shocks, is very slight. It has no action on pain. The prick of a needle excites the same sensations."

"The action of gymnema is therefore believed to be best explained by supposing that the nerve fibres or nerve endings capable of being stimulated by pure sweet and bitter substances are different from those which are excited only by acid and salt. The selective action of cocaine, not only on the nerve endings concerned with taste, but on others associated with more general sensory impressions, points also to the multiplicity of the kind of endings of sensory nerves in the tongue. The more powerful action of cocaine on bitter taste than on sweet, and of gymnema on sweet taste than on bitter, may also be an indication that the nerve fibres or nerve endings concerned with these tastes are also distinct."

EXPERIMENTAL.

The present state of information respecting the constituents of gymnema leaves, as outlined in the introductory portion of this paper, suggested the desirability of subjecting them to a more complete examination. It seemed, moreover, of particular interest to determine somewhat more precisely the

chemical character of the substance designated as "gymnemic acid." For this purpose we were supplied with a large quantity of the leaves, which had been freshly gathered in India, and dried by exposure to the air.

It has been noted by Greshoff (*Ber. d. deutsch. chem. Ges.*, 1890, 23, 3548) that the leaves of *Gymnema latifolium*, Wall., contain a large quantity of amorphous amygdalin, but no hydrolysing enzyme, and therefore that no hydrocyanic acid is developed in contact with water or even by heating with dilute sulphuric acid. In contact with emulsin, however, it was observed that hydrolysis was quickly effected, and that the distilled liquid then contained hydrocyanic acid and benzaldehyde. This behaviour of the substance is very remarkable, inasmuch as amygdalin is known to be readily hydrolysed by heating with dilute mineral acids.

With consideration of the above observation, a portion of the leaves of *Gymnema sylvestre* was tested for a cyanogenetic compound, but neither by contact with water nor by the action of dilute acids or emulsin was any hydrocyanic acid developed.

In order to ascertain the general character of the constituents of the leaves, the following preliminary experiments were made.

Fifty grammes of the ground leaves were extracted successively in a Soxhlet apparatus with various solvents, when the following percentages of extract, dried at 100°C., were obtained:—

(1) Petroleum (b.p. 40–60°C)	gave 2.34 per cent.
(2) Ether	gave 1.16 per cent.
(3) Chloroform	gave 0.88 per cent.
(4) Alcohol	gave 17.02 per cent.
	<hr/>
Total	21.40 per cent.

The petroleum, ether, and chloroform extracts were resinous in character, and insoluble in water. The alcoholic extract was soluble in water, affording a solution which gave an abundant precipitate with mineral acids, a dark coloration with ferric chloride (not due to tannin), and reduced Fehling's solution. All the extracts were very dark in colour and were free from alkaloid. A special test for alkaloids was also made by digesting a portion of the leaves with Prollius' fluid, but with a negative result. The leaves, after complete extraction with alcohol, were treated with boiling water, which, however, removed only gum, colouring matter, and other indefinite substances. The air-dried leaves yielded, on ignition, 8.6 per cent.

of inorganic residue, which was found to contain: CaO, 19.3 per cent.; Fe_2O_3 and Al_2O_3 , 17.9 per cent.; MgO, 2.7 per cent.; the remainder consisting chiefly of alkali carbonates with traces of manganese and silica.

The above-mentioned alcoholic extract yielded, on ignition, 0.59 per cent. of inorganic residue, which contained all the metals identified in the leaves.

For the purpose of a complete examination of the constituents of the leaves a large quantity of them was extracted by continuous percolation with hot alcohol. After removing the greater part of the alcohol by distillation, a quantity of water was added to the concentrated extract, and the mixture heated on a water-bath in order to eliminate the remainder of the alcohol. A soft, dark green resinous mass was thus obtained, amounting to about 5 per cent. of the weight of the leaves. For subsequent reference this substance may be designated as (A).

To the filtrate from the above precipitate diluted sulphuric acid was added in slight excess. This precipitated a quantity of a dark-coloured resinous substance, which amounted to about 17 per cent. of the weight of the leaves. It was separated from the liquid and washed with hot water until free from mineral acid. In the subsequent description of its properties it will be designated as (B).

The acid filtrate from (B) was shaken with chloroform, which removed only a very small quantity of resinous substance. It was then made alkaline with barium hydroxide, the precipitate of barium sulphate removed by filtration, and the liquid again shaken with chloroform, when only a little resinous substance was obtained. The alkaline liquid was then neutralised, concentrated, and an excess of basic lead acetate added, which produced a bulky, bright yellow precipitate. This was collected on a filter, washed with a little water, and then suspended in water and decomposed by hydrogen sulphide. The liquid filtered from the lead sulphide was concentrated under diminished pressure, when a brownish-yellow, amorphous substance was deposited, which was not again readily soluble in water. As it resisted all attempts to obtain it in a crystalline state, it was not further examined.

The filtrate from the lead acetate precipitate was treated with hydrogen sulphide for the removal of the lead, filtered, and concentrated under diminished pressure. The examination of this liquid will be described under (C).

(A) *Examination of the Resin separated by the addition of Water to the Alcoholic Extract of the Leaves.*

This product was a soft, greenish-coloured mass. A portion of it was mixed with sawdust, dried, and extracted successively with the following solvents :—

(1) Petroleum (b.p. 40·60°C.)	extracted 78·75 per cent.
(2) Ether	extracted 6·25 per cent.
(3) Chloroform	extracted 1·87 per cent.
(4) Ethyl acetate	extracted 4·37 per cent.
(5) Alcohol	extracted 8·12 per cent.

Total . . . 99·36 per cent.

The petroleum extract was a dark, brownish-green, soft solid. A quantity (238 Gm.) of it was boiled for some time with an alcoholic solution of potassium hydroxide (60 Gm.), the alcohol then removed and water added, when nothing separated. The aqueous solution, which had not the character of a soap, was extracted repeatedly with ether. The ethereal liquids had a yellow colour, and, after the removal of the ether, afforded a crystalline residue. This was distilled under 20 Mm. pressure, when a clear liquid was obtained, which rapidly became crystalline. The boiling point of the substance was evidently very high. It was finally re-crystallized several times from ethyl acetate, when about 3 Gm. of the pure substance were obtained in the form of nearly colourless, pearly leaflets, melting at 68°C.

On analysis :—

0·1072 gave 0·3338 CO₂ and 0·1406 H₂O. C=84·9 ; H=14·6.

C₃₁H₆₄ requires C=85·3 ; H=14·7 per cent.

The substance was thus found to be a hydrocarbon, and to correspond in its characters to *hentriacontane*, the melting point of which is given as 68·1°. It is of interest to recall the fact that the same hydrocarbon was found quite recently in the seeds of *Brucea sumatrana* (Power and Lees, *Year-Book of Pharmacy*, 1903, p. 512), but otherwise it has only been known to occur in nature in beeswax.

The liquid from which the hentriacontane had been extracted was acidified with sulphuric acid, when a large amount of black tarry matter was precipitated. After distilling off the volatile acids in steam, the liquid was shaken with ether, which removed a quantity of soft, black amorphous substance, from which

nothing crystalline could be obtained, and which apparently consisted simply of an acid resin. The volatile acids were converted into a barium salt, the solution of which gave the reactions characteristic of *formic acid*, and when acidulated with sulphuric acid developed a strong odour of *butyric acid*. A portion of the crystallised salt was dried and analysed :—

0.3192 gave 0.3456 BaSO₄. Ba=52.0 per cent.

As barium formate requires 60.4 per cent. Ba, and barium butyrate 44.1 per cent. Ba, the substance analysed would appear to have been a mixture of these two salts in about equal proportions.

The portions of the original resin (A) which had been dissolved by extraction with ether, chloroform, ethyl acetate, and alcohol respectively, were all very dark-coloured substances, from which nothing crystalline could be separated, and they were, therefore discarded. The ethyl acetate extract appeared to consist chiefly of the substance described under (B).

B) *Examination of the Precipitate produced by the addition of Sulphuric Acid to the Aqueous Filtrate from (A).*

This product, which is a crude and complex mixture of substances, would appear to represent the "gymnemic acid" described by Hooper (*loc. cit.*), although as obtained by us it differs considerably in some of its characters from those recorded by the former investigator. It amounted to about 17 per cent. of the weight of the leaves. For the purpose of its purification it was dissolved in alcohol, the solution mixed with sawdust which had previously been deprived of soluble substances, and, after thorough drying, extracted successively in a Soxhlet apparatus with the following solvents :—

1. Petroleum (b.p. 40–60°C.). This removed a very small amount of a yellowish fatty matter.

2. Ether. This extracted a small amount of a black resin.

3. Chloroform. This also extracted a relatively small amount of a soft black resin, containing much chlorophyll.

4. Ethyl acetate. This extracted an amount of substance corresponding to about 35 per cent. of the original precipitate or 6 per cent. of the weight of air-dried leaves.

5. Alcohol. This finally dissolved all of the residual substance.

Of the above extracts, only that obtained with ethyl acetate was capable of affecting or temporarily destroying the sense of taste for sweet substances. We are, therefore, unable to confirm the observation of Hooper (*loc. cit.*) that the anti-saccharine principle is soluble in ether, benzene and chloroform.

(a) *Characters of the Substance obtained from Precipitate (B) by extraction with Ethyl Acetate. (Gymnemic Acid.)*

The name of gymnemic acid has hitherto been applied to the complex mixture of substances precipitated by sulphuric acid, but as it has been shown by the above-described method of purification that the only portion of the mixture which possesses the peculiar anti-saccharine property of the leaves is that extracted by ethyl acetate, it would appear desirable that the name should at least be restricted to this active portion. Notwithstanding the fact that even the latter substance does not appear to be a chemical entity, the name of gymnemic acid may conveniently be retained for it, and it will, therefore, be so designated in the following description of its chemical characters.

Gymnemic acid is a resinous substance, of a greenish-brown colour, and has feebly acidic properties. It is only sparingly soluble in ethyl acetate, and is, therefore, extracted very slowly by this solvent from the crude product containing it. It is readily soluble in alcohol and in acetone, but is insoluble in petroleum, ether, chloroform, benzene and water. When gradually heated it softens, and melts indefinitely between 150° and 175°C.

An attempt was made to further purify the substance, and, as it could not be obtained in anything approximating a crystalline state, to ascertain by other means whether it was homogeneous in composition. It was, therefore, first extracted with chloroform, which removed a further small quantity of chlorophyll. A portion of the substance was then dissolved in alcohol, mixed with sawdust, thoroughly dried, and extracted in a Soxhlet apparatus with ethyl acetate in such a manner as to obtain five successive fractions. Of these the first, third, and fifth fractions, after drying at 100°C., were analysed.

First Fraction.—0.1041 gave 0.2207 CO₂ and 0.0736 H₂O.

Third Fraction.—0.1384 gave 0.2940 CO₂ and 0.0917 H₂O.

Fifth Fraction.—0.1085 gave 0.2287 CO₂ and 0.0684 H₂O.

These figures correspond to the following percentages, which are in fairly close agreement :—

—	I.	III.	V.
C. . .	57.8	57.9	57.4 per cent.
H. . .	7.8	7.3	7.8 per cent.

Twenty Gm. of the substance were dissolved in alcohol, and boiled repeatedly with animal charcoal until the colour of the liquid no longer became appreciably lighter. After filtering, and removing the solvent, a brown, varnish-like mass was obtained, which could be reduced to a very light brown powder, and amounted to 11 Gm. This, after drying at 100°C., was also analysed, with the following result :—

0.1090 gave 0.2428 CO₂ and 0.0796 H₂O.
C=60.7; H=8.1 per cent.

This purified substance melted at 145–155°C., with decomposition, and, although rendered much lighter in colour by the above treatment, it possessed the same general characters as before. The result obtained by its analysis, however, afforded evidence that it had become appreciably changed in composition, and that it could not be regarded as an individual substance.

Gymnemic acid is readily dissolved by the caustic alkalis, forming dark coloured solutions, the sodium and potassium compounds being only sparingly soluble in the cold. The ammonium compound is a black, amorphous solid, which dissolves readily in water, forming a solution which froths strongly on agitation, and still possesses the anti-saccharine property. From the solutions of these compounds the gymnemic acid is precipitated by mineral acids and by acetic acid, but it is readily re-dissolved by an excess of the latter. The potassium compound was boiled for a few minutes with a 20 per cent. aqueous solution of potassium hydroxide, and the gymnemic acid then re-precipitated, when it was found to have lost its anti-saccharine property. The acid dissolves slightly in cold sodium carbonate, no carbon dioxide being evolved until the liquid is boiled, when the acid dissolves freely. When the solution in sodium carbonate was boiled with animal charcoal, and the filtered liquid subsequently acidified, no precipitate was obtained, the gym-

nemic acid having been completely absorbed. The compounds of gymnemic acid with mercury, lead, silver, copper, iron, barium and calcium were prepared by the precipitation of a salt of the respective metal with a solution of the ammonium compound of the acid. When dry, they all formed black, amorphous products, soluble in alcohol, but which were quite unsuitable for analysis.

On warming gymnemic acid with concentrated nitric acid, a yellow, resinous compound was obtained, which was readily soluble in hot alcohol, and on cooling separated in an amorphous state.

If gymnemic acid is boiled for a few minutes with dilute sulphuric or hydrochloric acid, its anti-saccharine property is destroyed. The resinous substance which is formed shows practically the same behaviour towards solvents as the original acid, with the exception of being insoluble in alkalis. An alcoholic solution of hydrogen chloride had the same action as the aqueous acid. No sugar is produced by the above treatment, and we are, therefore, unable to confirm the observation of Hooper (*loc. cit.*), apparently made, however, with the crude substance, that gymnemic acid is a glucoside.

Action of Acetic Anhydride and of Benzoyl Chloride.—A portion of gymnemic acid was boiled with acetic anhydride and anhydrous sodium acetate, when it readily dissolved. On pouring this solution into water, a black, brittle, resinous solid separated. Another portion of the acid was dissolved in a solution of potassium hydroxide and shaken with benzoyl chloride, when a light brownish resin was obtained. These products were soluble in alcohol, ethyl acetate, and chloroform, but insoluble in alkalis, thus indicating that some change had been effected in the acid. Neither of them, however, could be obtained in a crystalline state.

Fusion with Potassium Hydroxide.—Forty Gm. of gymnemic acid were introduced into 200 Gm. of the caustic alkali, to which a little water had been added, and the mixture gradually heated. At about 170°C. gas commenced to be evolved rapidly, and this continued until a temperature of about 220°C. was attained. A considerable portion of the acid did not pass into solution, but formed a spongy mass on the surface, which remained undissolved even when kept for some time at 250–270°C. After cooling, water was added, when the greater part of the fused mass was dissolved, but there remained an insoluble slimy product, which was separated by filtration, and, after treatment

with dilute sulphuric acid, gave a resinous substance that was insoluble in all the ordinary solvents. The filtered liquid was acidified with sulphuric acid, distilled in steam, the volatile acid converted into a barium salt, and the concentrated solution of the latter precipitated in five fractions by silver nitrate. All of these fractions were analysed :—

Fraction 1.—0.2017 gave 0.1275 Ag. Ag=63.2 per cent.

Fraction 2.—0.2148 gave 0.1358 Ag. Ag=63.2 per cent.

Fraction 3.—0.1658 gave 0.1048 Ag. Ag=63.2 per cent.

Fraction 4.—0.2205 gave 0.1402 Ag. Ag=63.6 per cent.

Fraction 5.—0.2091 gave 0.1344 Ag. Ag=64.3 per cent.

$C_2H_3O_2$ Ag requires Ag=64.6 per cent.

The volatile acids formed by the fusion therefore consisted essentially of *acetic acid*, with apparently a very small amount of a higher acid.

The liquid remaining in the distillation flask was extracted with ether, the ethereal solution washed, dried, and the ether removed. A product was thus obtained which, when dissolved in hot water, treated with animal charcoal, and the solution concentrated, afforded colourless crystals. These melted at $192^\circ C.$, and this melting point was not changed by further crystallisation. The substance gave the colour reactions characteristic of *protocatechuic acid*, and, after drying at $110^\circ C.$, was analysed with the following result :—

0.1003 gave 0.2100 CO_2 and 0.0367 H_2O . C=57.1 ; H=4.1.

0.0961 gave 0.2008 CO_2 and 0.0366 H_2O . C=57.0 ; H=4.2.

The compound $C_7H_6O_4 + C_7H_6O_3$ requires C=57.5 ; H=4.1 per cent.

From the characters and analysis of this substance it is highly probable that it represents the compound formed by the union of equal molecules of *protocatechuic* and *para-oxybenzoic* acids. This well-defined molecular compound of these two acids has previously been obtained by the fusion of *benzoin* with potassium hydroxide. Its components cannot be separated by crystallisation nor by fractional precipitation with lead acetate, since it is converted by the latter into a crystalline lead salt of constant composition. (Compare *Beilstein's Handbuch der organischen Chemie*, Bd. II., p. 1740.)

Oxidation with Potassium Permanganate.—Forty Gm. of *gymnemic acid* and 10 Gm. of potassium hydroxide were dis-

solved in about 4 litres of water, and a cold, dilute solution of permanganate gradually added. The colour of the permanganate was rapidly discharged at first, but after 105 Gm. had been added it remained unchanged for some time. The liquid was filtered, the light yellow filtrate concentrated, and then acidified with sulphuric acid, when a very large amount of carbon dioxide was evolved, and a small quantity of resinous matter separated. On subsequent distillation in steam, an acid distillate was obtained from which a barium salt was prepared, and this afforded the reactions of *formic acid*.

The liquid remaining in the distillation flask was saturated with ammonium sulphate and repeatedly extracted with ether, but the latter removed only a small amount of brownish-coloured substance, from which nothing crystalline could be obtained.

(β) *Characters of the Acid Resin from Precipitate (B), which was insoluble in Ethyl Acetate.*

It has been noted that in the purification of the precipitate designated as (B) a considerable portion was insoluble in ethyl acetate, and was, therefore, finally extracted by alcohol, in which it was readily soluble. This substance was of a resinous nature, and when dry could be reduced to a light brown powder. It was freely soluble in alkalis, but completely devoid of the anti-saccharine property which particularly characterises the substance soluble in ethyl acetate.

It was thought of interest to ascertain whether there were any marked points of distinction in chemical behaviour between this acid resin and the gymnemic acid with which it was associated in the crude product. It was therefore dried at 100°C. and analysed.

0.0984 gave 0.2061 CO₂ and 0.0612 H₂O.
C=57.1 ; H=6.9 per cent.

It is thus seen not to differ very greatly in its elementary composition from the substance designated as gymnemic acid.

Fusion with Potassium Hydroxide.—Forty Gm. of the acid resin were fused with 200 Gm. of potassium hydroxide in the manner described under gymnemic acid, and the products of the reaction were similarly treated. The volatile acids were converted into a barium salt, and were found to consist chiefly of *formic acid*, with apparently a small amount of acetic acid.

In this respect the acid resin differs essentially from gymnemic acid, since the latter yielded no formic acid by potash fusion.

After the separation of the volatile acids, the liquid remaining in the distillation flask was extracted with ether. A crystalline acid was thus obtained, which melted at 192°C., and gave the colour reactions characteristic of protocatechuic acid. It was dried at 110°C., and then analysed, with the following result:—

0.1040 gave 0.2181 CO₂ and 0.0390 H₂O. C=57.2; H=4.1.

The compound C₇H₆O₄ + C₇H₆O₃ requires C=57.5; H=4.1 per cent.

This substance evidently represents the molecular compound of protocatechuic and para-oxybenzoic acids, and is, therefore, identical with that obtained by the potash fusion of gymnemic acid.

(C) *Examination of the Aqueous Liquid remaining after the separation of the Precipitates (A) and (B).*

This liquid, as previously noted, was deprived of colouring matter, and concentrated under diminished pressure. It then formed a thick syrup, which slowly deposited a quantity of colourless crystals. This crystalline substance consisted of a *lævo-rotatory modification of quercitol*, a pentatomic alcohol of the formula C₆H₇(OH)₅.H₂O, which has already been fully described by the authors (*Journal of the Chemical Society*, 1904, 85, 624). The syrupy liquid contained an abundance of sugar, from which an osazone was prepared. This crystallised in handsome, bright yellow needles, and melted at 218–219°C., which is the melting point of the osazone of *inactive glucose*. It was analysed with the following result:—

0.1056 gave 0.2339 CO₂ and 0.0590 H₂O. C=60.3; H=6.2.

C₁₈H₂₂N₄O₄ requires C=60.3; H=6.1 per cent.

On heating the above-mentioned syrupy liquid with potassium hydroxide, an abundance of ammonia was evolved, which indicated that it also contained some proteid matter.

The Fruits of Gymnema sylvestre.

From the large quantity of gymnema leaves available to us, it was possible to separate a sufficient quantity of the fruits of the plant for a comparative examination. They were ground,

and extracted by percolation with alcohol, which yielded an amount of dry extract corresponding to 7.7 per cent. of their weight. This extract was treated in a similar manner to that obtained from the leaves, and, although not quite so completely examined, it appeared to have the same general characters as the latter. No quercitol, however, could be obtained from it, and most of the products were dark in colour, amorphous, and of a resinous nature.

SUMMARY AND CONCLUSIONS.

From the somewhat extended details of this investigation the essential results and deductions therefrom may be briefly summarised.

1. The leaves of *Gymnema sylvestre* contain no cyanogenetic compound, such as has been observed by Greshoff to exist in the leaves of *G. latifolium* (*Ber. d. deutsch. chem. Ges.*, 1890, **23**, 3548).

2. From an alcoholic extract of the leaves, water precipitates a quantity of soft, dark-coloured, resinous matter, of an acid nature, the chief portion of which is soluble in petroleum. This petroleum extract, after treatment with an alcoholic solution of potassium hydroxide, yielded to ether a substance crystallising in pearly leaflets, melting at 68°C., which was identified as *hentriacontane*, $C_{31}H_{64}$. It is contained to the extent of about 0.05 per cent. in the leaves. The alkaline liquid, when acidified and distilled, yielded *formic acid* and a *butyric acid*.

3. The filtrate from the above precipitate, when acidified with sulphuric acid, yielded a quantity of a dark coloured resinous product. This would appear to represent the substance described several years ago by Hooper (*Chemical News*, 1899, **59**, 159), who observed it to possess the peculiar, anti-saccharine property of the leaves, and designated it as "gymnemic acid." He regarded it as a glucoside, existing in the leaves as a potassium salt, and assigned to it the formula $C_{32}H_{55}O_{12}$, although neither the substance itself nor any derivative of it was obtained in a crystalline state. The results of our investigation have shown that the precipitate obtained by the above described method is an exceedingly impure and complex mixture of substances. By its successive treatment with various solvents, a portion was extracted by ethyl acetate, which alone possessed the property of destroying the sense of taste for sweet substances,

and which amounted to about 35 per cent. of the original precipitate, or about 6 per cent. of the weight of air-dried leaves employed. For the portion thus dissolved by ethyl acetate the name of *gymnemic acid* may conveniently be retained as a distinguishing title, although, even in the purest form in which we have been able to obtain it, there is no assurance that it represents a homogeneous substance. It is amorphous, and apparently incapable of yielding any crystalline salt or other simple crystalline derivative. For these reasons it is undesirable that any chemical formula should be assigned to it. It is sparingly soluble in ethyl acetate, readily soluble in alcohol, and insoluble in ether, chloroform, benzene, and water. It is not a glucoside, but has very weak acidic properties, so that it is readily separated from a solution of the soluble combination in which it exists in the leaves by the addition of a mineral acid. Its anti-saccharine properties are destroyed by heating with the fixed alkalis or with dilute mineral acids. When fused with potassium hydroxide it afforded *acetic acid* and a molecular compound of *protocatechuic and para-oxybenzoic acids*, melting at 192°C. On oxidation with potassium permanganate the only product that could be identified was *formic acid*.

It has been stated by Hooper (*loc. cit.*) that "as gymnemic acid forms insoluble salts with alkaloids, this accounts for its masking the taste of quinine and other bitter substances." It is evident, however, from the physiological tests that have been made with the acid, that its property of affecting the sense of taste is due to its action upon the nerve fibres or nerve endings of the tongue, although this is less marked in the case of bitter than of sweet substances.

4. The resinous substance associated with the gymnemic acid in the crude product as first precipitated, but which is insoluble in ethyl acetate, is readily dissolved by alcohol. It is also of an acidic nature, but is completely devoid of any anti-saccharine property. When fused with potassium hydroxide it yielded *formic acid* and apparently a small amount of acetic acid, together with the same crystalline compound of *protocatechuic and para-oxybenzoic acids*, as was obtained from gymnemic acid by this treatment.

5. The aqueous liquid from which the preceding substances had been separated, after being deprived of colouring matter, afforded a handsomely-crystalline substance. This consisted of a *lævo-rotatory modification of quercitol*, which has already

been described by the authors (*Journ. Chem. Soc.*, 1904, 85, 624). It was associated with a sugar, the crystalline osazone of which melted at 218 to 219°C., and, therefore, corresponds with that of *inactive glucose*.

6. The constituents of the fruits of *Gymnema sylvestre* appear to be similar to those of the leaves, but no quercitol could be obtained from them.

7. Some experiments conducted in the Wellcome Physiological Research Laboratories have shown that neither the gymnemic acid nor the resin insoluble in ethyl acetate possesses toxic properties, nor could any pronounced physiological effects be observed when these substances were administered to the lower animals in doses of 0.5 to 1 Gm.

THE WELLCOME CHEMICAL RESEARCH LABORATORIES,
LONDON, E.C.

TITLES OF PUBLISHED PAPERS FROM
THE WELLCOME CHEMICAL RESEARCH LABORATORIES

continued

25. A NEW SYNTHESIS OF α -ETHYLTRICARBALLYLIC ACID
26. THE CONSTITUENTS OF THE ESSENTIAL OIL OF ASARUM CANADENSE
27. DERIVATIVES OF GALLIC ACID
28. THE OCCURRENCE OF SALICIN IN DIFFERENT WILLOW AND POPLAR BARKS
29. THE CONSTITUENTS OF COMMERCIAL CHRYSAROBIN
30. THE CONSTITUENTS OF AN ESSENTIAL OIL OF RUE
31. METHYL β -METHYLHEXYL KETONE
32. INTERACTION OF KETONES AND ALDEHYDES WITH ACID CHLORIDES
33. THE ANATOMY OF THE STEM OF DERRIS ULIGINOSA, *Benth.*
34. THE CHEMISTRY OF THE STEM OF DERRIS ULIGINOSA, *Benth.*
35. THE CONSTITUTION OF PILOCARPINE—Part IV.
36. PREPARATION AND PROPERTIES OF DIMETHYLGLYOXALINE AND DIMETHYLPYRAZOLE
37. THE ELECTROLYTIC REDUCTION OF PHENO- AND NAPHTHOMORPHOLONES
38. CHEMICAL EXAMINATION OF KÔSAM SEEDS (BRUCEA SUMATRANA, *Roxb.*)
39. COMPARATIVE ANATOMY OF THE BARKS OF THE SALICACEÆ—Part I.
40. THE CONSTITUTION OF CHRYSOPHANIC ACID AND OF EMODIN
41. THE CONSTITUTION OF EPINEPHRINE
42. A LEVO-ROTATORY MODIFICATION OF QUERCITOL
43. THE CONSTITUENTS OF THE ESSENTIAL OIL OF CALIFORNIAN LAUREL
44. SOME DERIVATIVES OF UMBELLULONE
45. THE CONSTITUENTS OF CHAULMOOGRA SEEDS
46. THE CONSTITUTION OF CHAULMOOGRIC ACID—Part I.
47. CHEMICAL EXAMINATION OF CASCARA BARK
48. CHEMICAL EXAMINATION OF GYMNEMA LEAVES

