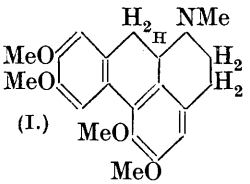
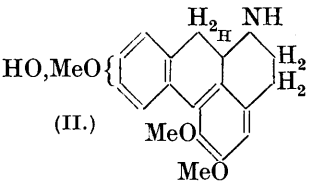
CCCLXXXIV.—*The Constitution of Laurotetanine.*

By GEORGE BARGER and ROBERT SILBERSCHMIDT.

LAUROTETANINE is an alkaloid discovered by Greshoff ("Verslag v.h. onderzoek v.d. Plantenstoffen Ned. Indie," 1890, 98; *Ber.*, 1890, **23**, 3537) in various *Lauraceæ;* injected, it causes tetanic convulsions. It was studied by Filippo (*Arch. Pharm.*, 1898, **236**, 601) and particularly by Gorter (*Bull. Jard. bot. Buitenzorg*, 1921, **3**, 180). The latter showed that the anhydrous base is $C_{16}H_{10}(OH)(NH)(OMe)_3$ and being impressed by the chemical and physiological resemblance of its *N*-methyl methyl ether to the alkaloid glaucine, he assumed the same carbon skeleton in both alkaloids. Now glaucine (I) has been synthesised from laudanosine by Gadamer (*Arch. Pharm.*, 1911, **249**, 680) and is a derivative of phenanthrene and *iso*quinoline. Since Gorter regarded the *N*-methyl



(I.)



(II.)

methyl ether of laurotetanine as different from glaucine, he named it *iso*glaucine and considered the isomerism due to different positions of two methoxy-groups. A further study of these compounds and the products of their exhaustive methylation by Hofmann's method has, however, convinced us that glaucine and Gorter's "*iso*glaucine" are identical, so that laurotetanine may be represented by (II).

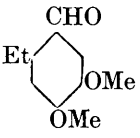
The one free hydroxyl group is as indicated in the benzene ring attached to two carbon atoms, for on oxidation of laurotetanine Gorter found that the benzene ring attached to three carbon atoms survives as a dimethoxybenzenetricarboxylic acid. The choice between the two possible formulæ indicated by (II) might perhaps be made by exhaustive methylation of laurotetanine ethyl ether and comparison of the product with two synthetic ethoxytrimethoxyethylphenanthrenes.

We carried out a number of preliminary experiments with this end in view. Although we could readily obtain from laurotetanine and glaucine a vinylphenanthrene and a phenanthrenecarboxylic acid, we were quite unable to decarboxylate the acid. We therefore decided to reduce the vinyl to the ethyl compound and attempt the synthesis of the latter.

For this purpose the required 3 : 4-*dimethoxyethylbenzene* was

obtained both by reducing 3 : 4-dimethoxyvinylbenzene (from 3 : 4-dimethoxyphenylethylcarbinol by Klages' method, *Ber.*, 1903 **36**, 3588) and by reducing acetoveratrone according to Clemmensen (*Ber.*, 1913, **46**, 1838). The latter alternative avoids the complications due to the polymerisation of the vinyl compound and is much the better method. Acetoveratrone was prepared by oxidation of the phenylmethylcarbinol as well as directly from veratrole ; the latter route is the cheaper.

The dimethoxyethylbenzene was converted by Gattermann's reaction into an *aldehyde*, presumably (III), but this constitution has not been proved rigorously.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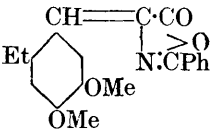
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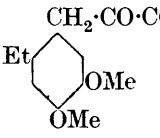
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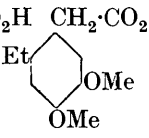
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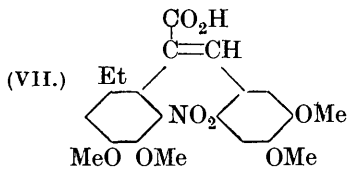
By condensation with hippuric acid to the *azlactone* (IV), hydrolysis to the *pyruvic acid* (V), and oxidation, a *dimethoxyethylphenylacetic acid* (VI) was obtained (compare Kropp and Decker, *Ber.*, 1909, **42**, 1184 ; Cain, Simonsen, and Smith, J., 1913, **103**, 1036 ; Haworth, Perkin, and Rankin, J., 1924, **125**, 1686). This was condensed with 6-nitro-3 : 4-dimethoxybenzaldehyde to the *nitro-acid* (VII), which after reduction and diazotisation was expected to yield a *phenanthrenecarboxylic acid* (VIII). The product of the decarboxylation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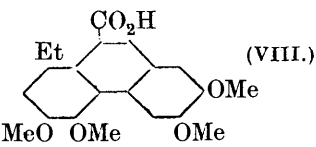
(VIII.)

of (VIII) was very similar to that obtained by the degradation of laurotetanine, but did not appear to be identical with it. Further work will be necessary to clear up this discrepancy.

EXPERIMENTAL.

A. *Exhaustive Methylation of Laurotetanine.*

Eike Gorter, we prepared the alkaloid from the bark of *Litsea citrata* from Java, using, however, a different process from the usual Stas–Otto method employed by Gorter and by Filippo, for by extraction of an alkaline solution with immiscible solvents we obtained hardly any crystalline alkaloid. An alcoholic extract prepared for us by Messrs. Duncan, Flockhart and Co. of Edinburgh was a thick, tarry mass representing about 10% by weight of the

bark. We dissolved it in two-thirds of its weight of hot glacial acetic acid, poured the solution into 5—6 volumes of water with vigorous mechanical stirring, and filtered off a precipitate (A). The brown filtrate, on being made slightly acid to litmus by addition of ammonia, gave a precipitate (B), partly composed of a weak base which probably contained several free hydroxyl groups and was readily oxidised. This alkaloid, which has some properties in common with boldine, a dihydroxydimethoxyphenanthrene alkaloid occurring in *Peumus Boldus* (*Monimiaceæ*, related to *Lauraceæ*), could not be crystallised. To the filtrate from (B) we added excess of ammonia and obtained a voluminous, pale yellow precipitate of crude laurotetanine, which was washed free from ammonia and dried on a porous tile in a vacuum. Precipitate (A) was redissolved in glacial acetic acid and treated as before, whereby it yielded a further quantity of crude laurotetanine. The dried alkaloid was powdered, mixed with sand, and extracted with ether for several days in a Soxhlet apparatus. Crystalline material soon began to separate from the ether, in which it is very sparingly soluble. It was recrystallised from acetone by the addition of a few drops of water. The hydrated alkaloid melted in its water of crystallisation at 124—134° (Gorter gives 125°; Filippo, 134°). It did not darken on exposure to light as stated by Gorter (at Buitenzorg). Yield, 0·3—0·4% (Gorter obtained 0·38%).

Complete Methylation. Dimethyl-laurotetanine Methiodide, $C_{22}H_{28}O_4NI$.—Preliminary experiments, in which laurotetanine and methyl iodide were heated in methyl-alcoholic sodium hydroxide, yielded two crystalline iodides, ultimately identified as the methiodide and methine hydriodide, but as the yield was unsatisfactory, diazomethane was used for the first step. Laurotetanine (1 g.) in 10 c.c. of methyl alcohol was mixed with an ethereal solution of diazomethane from 7 c.c. of nitrosomethylurethane. Next day, after evaporation, the methyl ether was separated by means of sodium hydroxide and treated as syrup with excess of methyl iodide in methyl alcohol. The methiodide, obtained in a yield of 60%, formed bundles of colourless needles, m. p. 210°, the same as that recorded for glaucine methiodide; the melting point remained constant on recrystallisation and did not rise to 226°, that given by Gorter for "*iso*glaucine methiodide." The methiodide was very sparingly soluble in water and could be completely extracted from aqueous solution by chloroform.

Dimethyl-laurotetaninemethine, $C_{22}H_{27}O_4N$. — The methiodide (0·5 g.) was heated for 5 hours with 40 c.c. of 10% alcoholic potassium hydroxide. The solution, which became slightly red, was concentrated and water was added; an oily base then separated.

When this was neutralised with acid, the *hydriodide* of the methine separated in colourless, rhomb-shaped leaflets, m. p. 265°, almost insoluble in water and in alcohol (Found : I, 26·2. $C_{22}H_{27}O_4N,HI$ requires I, 25·6%). The free methine, isolated after addition of ammonia and extraction with ether, could not be crystallised, and remained oily after distillation at 200° in a high vacuum. Yield, 0·4 g. The methine tenaciously retained a small quantity of an impurity, which gave it a pink colour and was less soluble in hot water than in cold. This impurity was absent from the methine prepared from glaucine, which could be obtained in colourless needles, m. p. 76°, from the dry ethereal solution, and probably accounts for the somewhat lower melting points of Gorter's "*iso*-glaucine" salts, as compared with those of glaucine.

Dimethyl-laurotetaninemethine methiodide, $C_{23}H_{30}O_4NI$, obtained on mixing the components in ethereal solution, formed white needles, m. p. 276°, very sparingly soluble in hot water.

2 : 3 : 5 : 6-*Tetramethoxy*-8-*vinylphenanthrene*.—The methine methiodide (0·37 g.; 0·75 millimol.) was heated with 40 c.c. of 10% methyl-alcoholic potassium hydroxide on the water-bath, the trimethylamine evolved being swept into 0·1*N*-hydrochloric acid. After 6 hours, 6·5 c.c. of the acid (= 90% of the theoretical) had been neutralised. (The trimethylamine was isolated as the chloroplatinate, m. p. 253° after crystallising from alcohol. Found : Pt, 37·2. Calc., 36·7%.) When the solution was concentrated, crystals separated; the quantity was increased by addition of water and neutralisation. On extraction with ether and crystallisation from alcohol, slightly pink, rhomb-shaped plates, m. p. 142°, were obtained. They decolorised potassium permanganate in cold acetone. Yield, 60% (Found : C, 74·05, 74·0; H, 6·3, 6·4. $C_{20}H_{20}O_4$ requires C, 74·0; H, 6·2%).

2 : 3 : 5 : 6-*Tetramethoxyphenanthrene*-8-*carboxylic Acid*.—To a cold solution of 5·1 g. of the vinyl compound in 10 c.c. of acetone, 0·175 g. of potassium permanganate (= 5O) was added during 1 hour. The manganese dioxide was filtered off and reduced with bisulphite and the solution was added to the residue left on evaporation of the acetone filtrate. Ether extracted from the alkaline solution a little unchanged vinyl compound and then, after acidification, a substance which crystallised from acetone–water or benzene–ligroin in yellow needles, m. p. 215°, very soluble in ether (Found : equiv., by back titration, 339. $C_{19}H_{18}O_6$ requires equiv., 342).

B. *Exhaustive Methylation of Glaucine.*

The above degradation had not been carried out by Gadamer (*loc. cit.*), who deduced the constitution by converting laudanosine

into glaucine. We isolated the latter alkaloid from 10 kg. of leaves and pods of *Glaucium luteum* from the Royal Botanic Gardens, Edinburgh, and from the coast near Cockburnspath. Fischer's method (*Arch. Pharm.*, 1901, **239**, 426) was greatly simplified by utilising the minute solubility of glaucine hydriodide in the presence of potassium iodide, the glaucine salt being under these conditions almost as sparingly soluble as the precipitate with Mayer's reagent (limit at 1 : 10,000). The material was covered with water, heated to boiling, strained, passed through a mincer, stirred for 2—3 hours in the original solution after addition of 2% acetic acid, strained, and extracted a second time. The filtered extracts were concentrated in a vacuum to a half-syrupy consistency and filtered through glass wool. Potassium iodide was then dissolved in the solution; after 2 days, a layer of brownish crystals had separated. One recrystallisation from much alcohol gave long, silky needles, still slightly pink, m. p. 243° (yield, 0·2% of the fresh leaves and pods). Thus this alkaloid is readily accessible. The filtrate from the crystals gave with ammonia a small precipitate yielding the colour reactions of protopine (Fischer, *loc. cit.*).

To obtain the pure base, the iodide was shaken with aqueous ammonia and ether. The former retained the colouring matter and the latter left on evaporation colourless prisms, m. p. 118—119°, after several crystallisations.

We prepared glaucine methiodide, the methine, methine methiodide, the vinylphenanthrene, and the carboxylic acid. We also prepared the characteristic iodide of dimethyl-laurotetanine (from laurotetanine and diazomethane). We thus had six different compounds, each obtained from two natural sources. In all cases they were identical; these results, tabulated below, dispose of Gorter's alleged difference between glaucine and dimethyl-laurotetanine. We have also tabulated the melting points given by Warnat (*Ber.*, 1925, **58**, 2768) for mixtures of derivatives of dimethylboldine and glaucine. Warnat was the first to publish the Hofmann degradation of glaucine, after we had already carried it out independently.

	Dimethyl-lauro-tetanine alone.	Glaucine alone.	Glaucine + dimethyl-lauro-tetanine.	Glaucine + dimethyl-boldine.
Hydriodide	243°	243°	243°	239—241°
Methiodide	219	219	219	221
Methine hydriodide	264	264	263	—
Methine methiodide	276	278	276	276—278
Vinylphenanthrene	142	142	142	142—143
Phenanthrene acid	215	214	214	—

Having established this identity, we carried out some experiments

as first steps in the determination of the position of the free hydroxyl in laurotetanine. In order to have a tetramethoxy-compound which could be synthesised, we attempted to decarboxylate the tetramethoxyphenanthrenecarboxylic acid. We entirely failed to do this, whether by distillation (when the acid sublimed at a high temperature), or by heating with lime, or by heating in glacial acetic acid. We therefore decided to base the comparison on the ethyl compound.

2 : 3 : 5 : 6-Tetramethoxy-8-ethylphenanthrene was prepared by reducing 0·485 g. (1·5 millimols.) of the vinyl compound (from glaucine), dissolved in the minimum amount of glacial acetic acid, which coloured the solution red, with hydrogen after addition of palladium chloride and gum arabic. The reaction stopped abruptly after 20 minutes, when 35 c.c. of hydrogen had been taken up. The solution was diluted with water, and the resulting mixture extracted with ether. The extract crystallised from methyl alcohol. Yield, 80%. The crystals were distilled in a high vacuum with the bath at 200°. The colourless distillate crystallised from alcohol in narrow plates, m. p. 120°.

Synthesis of 2 : 3 : 5 : 6-*Tetramethoxy*-8-*ethylphenanthrene.*

As we required large quantities of methyl vanillin and of veratrole, we paid some attention to the methylation of vanillin and of guaiacol, finally obtaining a practically quantitative yield by a rapid method.

Vanillin (152 g.; 1 mol.) is melted in a wide-mouthed bottle provided with a condenser, a stirrer, and two tap-funnels. With rapid stirring, 1·5 mols. (82 g.) of potassium hydroxide in 120 c.c. of water are run in at the rate of two drops a second, and 20 seconds after this has started, addition of 1·25 mols. (160 c.c.) of methyl sulphate is begun at the same rate. (The methyl sulphate has stood over potassium carbonate and is neutral to Congo.) The external heating is soon stopped. The reaction mixture keeps a pale reddish-brown and alkaline; green indicates an acid reaction. When three-quarters of the reagents have been added, the reaction mixture becomes turbid and separates into two layers. As soon as all has been run in (about 20 minutes from the start) the reaction mixture is poured into a porcelain basin. The upper layer solidifies to a hard, white mass of practically pure methyl vanillin, m. p. 43°. It is washed with water and dried. Yield, 165 g., or more than 99%. Veratrole was similarly obtained in a yield of 95% (after extraction with ether and distillation). Pure, crystalline guaiacol must be used. Acetoveratrone was obtained from veratrole and acetyl chloride by means of aluminium chloride in carbon disulphide

solution in a yield of 80% and was then reduced by Clemmensen's method (*Ber.*, 1913, **46**, 1838).

3 : 4-*Dimethoxyethylbenzene.*—720 G. of zinc, amalgamated for 1 hour with double the weight of 5% aqueous mercuric chloride solution, was washed and covered with 10% hydrochloric acid. The liquid was heated to boiling and 180 g. of acetoveratrone were added in the course of an hour. The boiling was continued for 5 hours, 50—100 c.c. of acid being added per hour. The oily layer removed by ether distilled mostly at 110—112°/9 mm.; the boiling point then suddenly rose to 230—240° and a polymeride distilled. Thus the lower fraction was at once pure and stable to permanganate. Yield, 68%. The substance could not be crystallised.

3 : 4-*Dimethoxy*-6-*ethylbenzaldehyde* (III).—To 54 g. of dimethoxyethylbenzene in 75 g. of anhydrous benzene, cooled by ice, 60 g. of freshly powdered aluminium chloride were slowly added, then 55 g. of hydrogen cyanide. Hydrogen chloride was passed for 3 hours at 0° and for ½ hour at 30°. Next day the imide was poured on ice and finally decomposed by a current of steam. The *aldehyde* distilled at 150—159°/9 mm. and crystallised on long standing in plates, m. p. 28—30°, very soluble in organic solvents (Found: C, 67·7; H, 7·2. $C_{11}H_{14}O_3$ requires C, 68·0; H, 7·3%). The semicarbazone formed colourless crystals, m. p. 197—199°.

3 : 4-*Dimethoxy*-6-*ethylbenzoic acid*, prepared by oxidising the aldehyde with alkaline permanganate, had m. p. 138° (Found: equiv., 210·5. $C_{11}H_{14}O_4$ requires equiv., 210).

Azlactone of 3 : 4-*Dimethoxy*-6-*ethylbenzylidenehippuric Acid* (IV).—A mixture of 39 g. of the aldehyde, 40 g. of hippuric acid, 82 c.c. of acetic anhydride, and 16 g. of sodium acetate was heated for 40 minutes on the water-bath. After cooling, the semi-solid mass was washed with much ether and with water; it then separated from hot alcohol, in which it was rather sparingly soluble, in yellow crystals, m. p. 155°. Yield, 66% (Found: C, 71·0; H, 6·0. $C_{20}H_{19}O_4N$ requires C, 71·2; H, 6·0%). The acid itself, m. p. 212°, was made by heating the lactone with 1% potassium hydroxide solution until it was colourless.

3 : 4-*Dimethoxy*-6-*ethylphenylpyruvic Acid* (V).—20 G. of the azlactone were boiled with 30 g. of potassium hydroxide in 100 c.c. of water, the ammonia evolved being swept into *N*-acid. In 1 hour 53 c.c. had been neutralised (= 94% of the theoretical). The solution was acidified with sulphur dioxide, and the benzoic acid filtered off. The solution, which contained the pyruvic acid as bisulphite compound, was acidified with hydrochloric acid and heated under reduced pressure to remove sulphur dioxide. A crystalline precipitate separated. Recrystallised from glacial

acetic acid and from alcohol, it formed pale yellow crystals, m. p. 181°; yield, 71%.

3 : 4-*Dimethoxy*-6-*ethylphenylacetic Acid* (VI).—The keto-acid (10 g.) in a large excess of dilute sodium hydroxide solution was oxidised at 0° by 37 c.c. of 3% hydrogen peroxide. After neutralisation with sulphur dioxide (to keep the unchanged acid in solution) the oxidation product separated as an oil which solidified next day, and was extracted with ether. It formed slightly pink bundles of needles, m. p. 67°; yield, 78% (Found : equiv., 222·5. $C_{12}H_{16}O_4$ requires equiv., 224).

α-3 : 4-*Dimethoxy*-6-*ethylphenyl*-β-6-*nitro*-3 : 4-*dimethoxyphenylacrylic Acid* (VII).—A solution of 5 g. of the previous acid in methyl alcohol was neutralised by methyl-alcoholic potassium hydroxide and evaporated in a round-bottom flask. The potassium salt was dried in a vacuum at 120—130° for 3 hours, and 10 g. of 6-nitro-3 : 4-dimethoxybenzaldehyde (Pschorr, *Ber.*, 1899, **32**, 3412), 8 c.c. of acetic anhydride, and 0·2 g. of anhydrous zinc chloride were then added. The flask was fitted with a long air-condenser and dry nitrogen was very slowly passed over the mixture to prevent oxidation. The mixture was heated for 20 hours at 120°, and then for 1 hour on the water-bath after addition of 20 c.c. of water. It was then made alkaline with ammonia and filtered from a brown tar, which was re-extracted with ammonia. After charcoal treatment, acidification gave a voluminous precipitate of the crude *acid* (yield, 52%), which separated from hot methyl alcohol in yellow crystals, m. p. 208° after sintering at 203° (Found : equiv., 418. $C_{21}H_{23}O_8N$ requires equiv., 417).

Amino-acid from the above. The nitro-acid (2·08 g.; 5 millimols.) was dissolved in 20 c.c. of dilute ammonia, heated to 80°, and slowly added to a reduction mixture at the same temperature, prepared by adding 8 c.c. of 25% ammonia to 9·2 g. of ferrous sulphate (33 millimols.) in a little water. The mixture was shaken and left on the water-bath for 2 hours and filtered. Acetic acid precipitated from the dark brown solution a deep yellow, amphoteric precipitate, m. p. 192° in the crude state.

2 : 3 : 5 : 6-*Tetramethoxy*-8-*ethylphenanthrene*-9-*carboxylic Acid* (VIII) *and* 2 : 3 : 5 : 6-*Tetramethoxy*-8-*ethylphenanthrene* (?).—The above amino-acid (0·9 g.; 2·5 millimols.), dissolved in the calculated quantity of sodium carbonate solution, was mixed with 2·5 c.c. of *N*-sodium nitrite and dropped very slowly into 9 c.c. of well-cooled and stirred *N*-sulphuric acid. After filtration, the solution of the diazonium salt was heated for 2 hours on the water-bath; it then no longer coupled with β-naphthol. A reddish-brown acid separated on acidification, but could not be crystallised; yield, 0·5 g. (crude).

A small-scale experiment with amyl nitrite and copper powder in the cold yielded hardly any of the acid after 12 hours. The crude acid was decarboxylated by heating 0·2 g. in a bath at 230—250°/9 mm. An ethereal solution of the oily distillate, after being washed with sodium carbonate, was evaporated; the residue crystallised from hot methyl alcohol in pale brown leaflets, m. p. 106°, and 112° when mixed with the corresponding compound from laurotetanine. The product (some 40 mg.) was redistilled and crystallised as before; it was now completely colourless and melted at 118°, but the mixture with the laurotetanine derivative melted at 106°. The lowering of the melting point is as yet unexplained; it is not clear which of the two substances should first be suspected of having a wrong constitution assigned to it. Both substances showed sparing solubility in methyl alcohol.

Some preliminary experiments were also undertaken with laurotetanine ethyl ether, prepared by means of ethyl *p*-toluenesulphonate. The oily base gave a hydriodide, m. p. 198—200°, a methiodide, m. p. 223—226°, a methine methiodide, m. p. 275°, and a trimethoxyethoxyvinylphenanthrene, m. p. 139°.

6-*Nitro*-3-*methoxy*-4-*ethoxybenzaldehyde* (ethyl nitrovanillin) was prepared by adding ethyl vanillin to a mixture of equal parts of fuming and ordinary concentrated nitric acids. After distillation under reduced pressure, it melted at 159—160° and was resublimed in a vacuum at 130—140° (Found: C, 53·3; H, 4·8. $C_{10}H_{11}O_5N$ requires C, 53·3; H, 4·8%). The condensation of this aldehyde with potassium dimethoxyethylphenylacetate has not yet proceeded in normal fashion.

We wish to express our great indebtedness to Dr. R. Wind, Director of the Experimental Forestry Station, and to Dr. W. Docters van Leeuwen, Director of the Botanic Gardens, Buitenzorg, Java, for a supply of the bark of *Litsea citrata*; also to Professor W. Wright Smith, Director of the Royal Botanic Gardens, Edinburgh, for help in securing a quantity of *Glaucium luteum*, and to the Moray Fund of Edinburgh University for a grant towards the expenses of the research.

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