392. *The Chemistry of* Aspergillus *Colouring Matters. Part II.*

By J. H. Cruickshank, H. Raistrick, and Robert Robinson.

A further study of flavoglaucin and auroglaucin and their reduction products is interpreted in the light of the hypothesis that the pigments are derived from quinol. Flavoglaucin is regarded as a *n*-octoyl*iso*pentenylquinol or a *n*-octoylvinyl*iso*propylquinol, and auroglaucin has the same skeleton with three more double bonds.

In Part I (Raistrick, Robinson, and Todd, J., 1937, 80) a preliminary investigation of the functional groups of flavoglaucin, $C_{19}H_{28}O_3$, and auroglaucin, $C_{19}H_{22}O_3$, was recorded and the results were held to justify the following conclusions: (1) Auroglaucin is constituted like flavoglaucin but contains three more double bonds. This statement was based on the results of catalytic hydrogenation of the pigments and especially on the formation under certain conditions of one and the same substance, $C_{19}H_{32}O_2$, from both of them.

(2) The pigments are ketones and probably contain two hydroxyl groups. The evidence was not decisive on the latter point.

(3) Flavoglaucin contains the group $CH_3{\cdot}[CH_2]_6{\cdot}C{:}$ in its molecule.

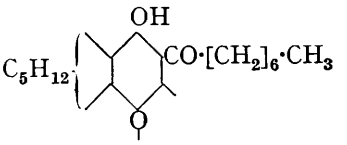
(4) The pigments may be benzene derivatives and in that case flavoglaucin might be additionally constructed from one or more aliphatic chains including the above fragment, a carbonyl group, a double bond (four in auroglaucin) and two hydroxyl groups (or one hydroxyl and a reducible cyclic ether group and no double bond).

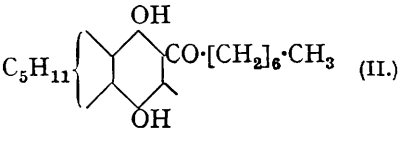
The work described in this paper confirms and extends these deductions and in order to clear the ground for discussion it may be stated at once that the pigments are undoubtedly benzene derivatives. A qualitative experiment (p. 2062) showed that a reduced and methylated flavoglaucin derivative could be nitrated, and the product reduced to

a diazotisable amine. Furthermore the ultra-violet absorption of tetrahydroflavoglaucin dimethyl ether gave strong indication of the quinol structure.

The whole behaviour of flavoglaucin and auroglaucin and of their transformation products is in harmony with this hypothesis and we consider that little risk is taken in assuming its validity in the sequel.

With palladised strontium carbonate as the catalyst, hydrogenation of both flavoglaucin and auroglaucin affords *dihydroflavoglaucin*, $C_{19}H_{30}O_3$, which is a ketone. The monomethyl ether of this substance affords *n*-octoic acid on oxidation with hydrogen peroxide in alkaline solution. On the quinol theory this must be an illustration of Dakin's reaction and therefore flavoglaucin must be (I). This decomposition is in itself evidence in favour of a ketophenolic structure.

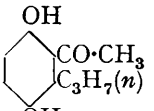
(I.) 

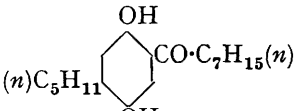
 (II.)

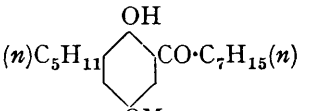
The next stage in the argument is to expand (I) to (II). In Part I (*loc. cit.*, p. 83) it was noted that auroglaucin appears to contain two active hydrogen atoms (Zerewitinoff), although the estimations gave somewhat low results (9·5, 9·8 instead of 11·4). We may now ascribe this to firm co-ordination of the hydrogen in the *o*-keto-phenol system. Flavoglaucin gave clear evidence of the existence of two hydroxyls in the molecule (10·9, 11·3. Calc., 11·2%).

Auroglaucin readily yields a *monomethyl ether*, which exhibits a ferric reaction; its *oxime* forms a complex copper derivative and hence it is probable that it contains a free phenolic hydroxyl in the *o*-position to the ketoxime group. For these reasons we believe that the pigments are dihydroxylic, flavoglaucin being represented by (II). It follows that the C_5H_{11} moiety contains the double bond indicated by catalytic hydrogenation experiments. The results of side-chain methyl estimations by the Kuhn–Roth method, depending on vigorous oxidation to acetic acid, would appear to indicate that flavoglaucin and auroglaucin contain only one structure capable of giving rise to acetic acid, although a second portion of the molecule furnishes a poor yield of acetic acid in the case of auroglaucin [Found (Part I): flavoglaucin, side-methyl, 0·9, 0·9; auroglaucin, side-methyl, 1·3]. It should be noted that *p*-phenylphenacyl *n*-octoate gives rise to about 1 mol. of acetic acid (p. 2061), so the molecule of acetic acid from the pigments is probably derived by terminal oxidation of the *n*-octoyl (or unsaturated corresponding group) residue. Furthermore tetrahydroflavoglaucin and some of its derivatives afford 2 mols. of acetic acid, suggesting that a part structure $\therefore C \therefore CH_2$ (no acetic acid on oxidation) becomes $\therefore CH \cdot CH_3$.

It is difficult to arrange the group C_5H_9 so that it should not give acetic acid on oxidation and, in view of the ready destruction of a substituted quinol by chromic acid, this difficulty is greatly increased unless the group is taken as a single substituent in the benzene nucleus. Undoubtedly methyl or ethyl groups attached to a quinol nucleus should be oxidised to acetic acid under the conditions of the experiment. Synthetic substances available are (III), (IV), and (V), and comparisons with these are of value because it has been found that certain properties are common to several members of the series in which the alkyl groups of the acyl residue and the direct benzene substituents are varied.


(III.)


(IV.)


(V.)

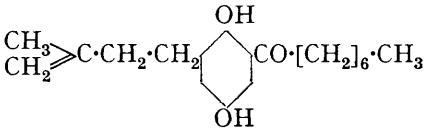
6-*n*-Propylquinacetophenone (III) and 6-allylquinacetophenone (Baker and Lothian, J., 1936, 279) exhibit little resemblance to dihydroflavoglaucin. The points of divergence are the lack of colour of (III), the ferric reactions, the non-reactivity of (III) to reagents for the carbonyl group (this point shows clearly that the pigments are not substituted in

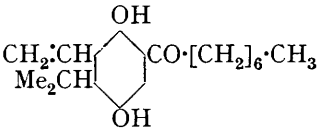
position 6), the ease of coupling of (III) with diazonium salts, the ready solubility of (III) in aqueous alkalis, and the ease of formation of the dimethyl ether of (III).

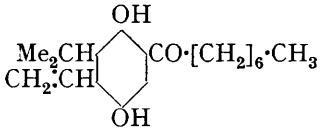
The ketone (IV) closely resembles dihydroflavoglaucin in many of its properties, but it is less intensely coloured, more stable to oxidising agents, and more readily soluble in aqueous sodium hydroxide. The ferric reaction of (IV) and its behaviour with alcoholic sodium hydroxide are almost the same as those of dihydroflavoglaucin. This is strong confirmation of the general correctness of our views.

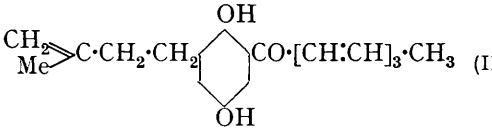
The dihydroxy-compound corresponding to (V) is unfortunately not yet available for study, but (V) is not identical with *dihydroflavoglaucin monomethyl ether* prepared by the catalytic reduction of auroglaucin monomethyl ether.

The evidence is not conclusive, but it is best collated if one of the alternative constitutions (VI), (VII), and (VIII) is attributed to flavoglaucin. The failure to derive a molecule of acetic acid from the C_5H_9 group of (VI) in the Kuhn–Roth estimation may then be due to oxidation of the methyl group adjacent to the double bond.

(VI.) 


(VII.)

(VIII.) 


(IX.)

Auroglaucin would be (IX) on the basis of (VI), or the analogous structures related to (VII) and (VIII), or, less probably, one of the double bonds in the acyl group could be transferred to the C_5 chain.

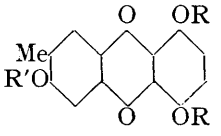
These provisionally proposed orientations may be thought to be supported by the difficulty experienced in characterising the second hydroxyl of the pigments by the preparation of derivatives.

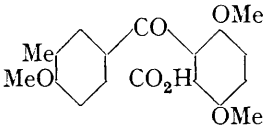
The constitutions to be attributed to the reduction products on the above basis are obvious on inspection. Dihydroflavoglaucin, $C_{19}H_{30}O_3$, is an *iso*amylquinoctophenone or ethyl*iso*propylquinoctophenone, and as such gives rise to a 2 : 4-dinitrophenylhydrazone; tetrahydroflavoglaucin, $C_{19}H_{32}O_3$, is the corresponding *sec.*-alcohol and accordingly it forms a triacetyl derivative and does not react with reagents for the carbonyl group. Tetrahydrodeoxyflavoglaucin, $C_{19}H_{32}O_2$, is an *iso*amyl-*n*-octylquinol or ethyl*iso*propyl-*n*-octylquinol and yields a *dimethyl* ether and a bisphenylurethane. Fully reduced flavoglaucin, $C_{19}H_{38}$ (Part I; *loc. cit.*, p. 85), is an *iso*amyl-*n*-octyl*cyclo*hexane or an ethyl*iso*propyl-*n*-octyl*cyclo*hexane.

It is hoped that further synthetic work will throw more light on the problem.

In Part I (*loc. cit.*) reasons were given for the belief that the congener of flavoglaucin and auroglaucin, termed rubroglaucin, is a methylmethoxyquinizarin, but experiments recently conducted in the laboratory of one of us (H. R.) have indicated that the material is not homogeneous. An account of this work will shortly be published and in the meantime further discussion of rubroglaucin would be valueless.

Before this development we synthesised 5 : 8-*dihydroxy*-3-*methoxy*-2-*methylanthraquinone* (X; R = H, R′ = Me), which on earlier information might have been identical with rubroglaucin.

(X.) 


(XI.)

Condensation of 3 : 6-dimethoxyphthalic anhydride with *o*-tolyl methyl ether in the presence of aluminium chloride affords a substituted benzoylbenzoic acid which by analogy

with previous work should be (XI) (compare Walsh and Weizmann, J., 1910, **97**, 685; Mitter and Sen, *J. Indian Chem. Soc.*, 1928, **5**, 631; Keimatso and Hirano, *J. Pharm. Soc. Japan*, 1930, **50**, 644).

Ring-closure by means of hot sulphuric acid is accompanied by partial demethylation, giving (X; R = H, R′ = Me), the orientation being again clear from the work of the above-mentioned authors. This *substance* is insoluble in cold aqueous sodium carbonate and hence does not contain a free β-hydroxyl group. To remove ambiguity due to the partial demethylation the fully demethylated *compound* (X; R = R′ = H) was prepared. It retains water tenaciously and even on sublimation; the *triacetyl* derivative has the anticipated composition.

Experimental.

Isolation of Pigments.—The crude mixture of colouring matters (Gould and Raistrick, *Biochem. J.*, 1934, **28**, 1640; Raistrick, Robinson, and Todd, J., 1937, 80) was repeatedly crystallised from ethyl alcohol to obtain pure auroglaucin, m. p. 152—153°. The mother-liquors were evaporated under diminished pressure. Extraction (Soxhlet) of the semi-solid residue with light petroleum (b. p. 40—60°) left most of the gummy impurities undissolved. On cooling, yellow needles separated, m. p. 87°, raised to 92° by one recrystallisation from 70% alcohol. Further recrystallisations from light petroleum and aqueous alcohol did not raise the m. p. above 94°. This product consisted mainly of flavoglaucin, for it readily gave pure derivatives; a pure specimen was isolated by hydrolysis of the condensation product with *o*-phenylenediamine. This derivative (Raistrick, Robinson, and Todd, *loc. cit.*) (70 mg.) was refluxed for 2 hours with hydrochloric acid (20 c.c. of 20%) in the presence of light petroleum (20 c.c.). After cooling, the organic layer was washed with water and evaporated in a vacuum, and the residue recrystallised from light petroleum. It formed long yellow needles, m. p. 104°. The impurity in crude flavoglaucin was auroglaucin, because reduction gave an almost quantitative yield of pure dihydroflavoglaucin, identical with the product from auroglaucin. The semicarbazone of flavoglaucin was precipitated from an ethereal solution by the addition of light petroleum in small colourless needles, m. p. 135°, but it could not be recrystallised.

Auroglaucin Monomethyl Ether.—A mixture of auroglaucin (1 g.), potassium carbonate (1 g.), methyl iodide (2 g.), and acetone (20 c.c.) was refluxed for 1 hour. Potassium carbonate (1 g.) was then added, and heating continued for 3 hours, with the addition of a few drops of methyl iodide from time to time. The acetone was removed under diminished pressure, water added, and the solid collected (0·84 g.). Crystallisation from alcohol gave yellowish-brown needles, m. p. 100° (Found: C, 76·8; H, 7·8; MeO, 9·5. $C_{20}H_{24}O_3$ requires C, 76·9; H, 7·7; 1MeO, 10·0%). The *ether* was almost insoluble in cold aqueous sodium hydroxide, and dissolved sparingly on heating, with darkening; with aqueous alcoholic sodium hydroxide it gave a deep yellowish-brown, and with alcoholic ferric chloride a permanent dark greenish-brown coloration. The presence of a carbonyl group was confirmed by the ready formation of a crystalline 2 : 4-dinitrophenylhydrazone by means of Brady's reagent (J., 1931, 756).

All attempts to prepare a dimethyl ether from auroglaucin were unsuccessful, and the hydroxyl group in the monomethyl ether, indicated by the ferric reaction, could not be diagnosed by the formation of any derivative.

Oxime of Auroglaucin Monomethyl Ether.—The methyl ether (0·3 g.) in alcohol (20 c.c.) together with a solution of hydroxylamine hydrochloride (0·07 g.) and sodium acetate (0·14 g.) in water (3 c.c.) was refluxed on the steam-bath for 6 hours. Water (5 c.c.) was added, and part of the alcohol removed in a vacuum; on cooling and keeping, a small quantity of greenish-yellow crystals separated. The *oxime* crystallised from light petroleum (b. p. 40—60°) in yellowish-green needles, m. p. 117° (decomp.) (Found: C, 73·2; H, 7·9. $C_{20}H_{25}O_3N$ requires C, 73·4; H, 7·6%).

The oxime formed a complex copper salt, soluble in benzene. Taylor and Ewbank (J., 1926, 2818) have shown that the formation of metallic chelate compounds of oximes indicates an enolic structure, such as an *o*-hydroxyl group in a benzene ring, adjacent to the oximino-group.

Dihydroflavoglaucin.—A solution of crude flavoglaucin (1 g.), m. p. 92°, in ethyl acetate (25 c.c.) was shaken for 2 hours with hydrogen at 2—3 atms. in presence of a palladised strontium carbonate catalyst (0·3 g. of 2%). Evaporation of the filtered solution left a solid residue and one recrystallisation from light petroleum afforded yellow needles (0·92 g.), m. p. 97°, raised by one recrystallisation to 98°; m. p. under water, 89° (Found: C, 74·6; H, 9·8. $C_{19}H_{30}O_3$

requires C, 74·5; H, 9·8%). A solution of *dihydroflavoglaucin* (50 mg.) and *o*-phenylenediamine (25 mg.) in alcohol (10 c.c.) was refluxed for 1 hour. On dilution with water and cooling, the product crystallised in yellowish-brown needles. Two recrystallisations from aqueous alcohol gave deep yellow needles, m. p. 150° (Found: C, 75·4; H, 9·2; N, 7·3. $C_{25}H_{36}O_2N_2$ requires C, 75·7; H, 9·1; N, 7·1%). The *substance* is therefore a Schiff base formed from its generators by elimination of one molecule of water.

A solution of dihydroflavoglaucin (50 mg.) in alcohol (2 c.c.) was added to Brady's reagent (1 c.c.) diluted with alcohol (1 c.c.), and the mixture kept at room temperature for 1 hour. The magma of deep red needles was drained and washed with a little alcohol. The 2 : 4-*dinitrophenylhydrazone*, recrystallised from alcohol, formed deep red, hair-like needles, m. p. 203° (Found: C, 61·6; H, 7·1; N, 11·3. $C_{25}H_{34}O_6N_4$ requires C, 61·7; H, 7·0; N, 11·5%).

Dihydroflavoglaucin was also obtained by shaking a solution of auroglaucin (1 g.) in ethyl acetate (25 c.c.) in hydrogen at 2—3 atms. for 4 hours, with palladised strontium carbonate (0·5 g. of 2%) as catalyst. The product, after solidifying, crystallised from light petroleum in yellow needles, m. p. 98° alone or mixed with the product from flavoglaucin. The condensation product with *o*-phenylenediamine had m. p. 150° (mixed m. p. 150°) and the 2 : 4-dinitrophenylhydrazone had m. p. 203° (mixed m. p. 203°).

Oxidation of Dihydroflavoglaucin.—The substance (0·7 g.) was dissolved in pyridine (50 c.c.) and oxidised with 4% aqueous potassium permanganate, volumes (5·8 c.c.) corresponding to 1 atom of oxygen being added successively. The oxidation proceeded rapidly until 9 atoms of oxygen had been used, but the permanganate solution constituting the tenth portion was decolourised very slowly. After keeping overnight, the manganese oxide precipitate was collected and washed with pyridine and water, and the combined filtrate and washings concentrated under diminished pressure to 20—30 c.c. Extraction with ether, and evaporation of the solvent, left a trace of a viscous oil with an odour of rue. The aqueous solution was acidified, and extracted six times with ether (20 c.c. each time). After drying over sodium sulphate, the ether was slowly distilled through a fractionating column; the residue was a pungent-smelling liquid, largely immiscible with water, and acid to litmus. This was shaken with water (5 c.c.), and the upper layer separated (0·15 g.) and converted into the *p*-phenylphenacyl ester, m. p. 67°, alone or mixed with authentic *p*-phenylphenacyl *n*-octoate (cf. Drake and Bronitsky, *J. Amer. Chem. Soc.*, 1930, **52**, 3715; 1932, **54**, 2059). The only other identifiable product of the oxidation was oxalic acid.

Dihydroflavoglaucin Dimethyl Ether (?)—A solution of dihydroflavoglaucin (1·8 g.) in acetone (50 c.c.) was refluxed for 2 hours and 20 minutes with the addition of seven successive equivalent quantities of methyl sulphate (1 c.c.) and aqueous sodium hydroxide (4 c.c. of 10%) at intervals of 20 minutes. An excess of sodium hydroxide was added, the acetone removed in a vacuum, and the product isolated by means of ether as a viscous oil (1·3 g.), b. p. 190—195°/0·01 mm. (ferric reaction, negative). The 2 : 4-dinitrophenylhydrazone was an oil which could not be crystallised. Oxidation of the *product* (0·8 g.) with slightly more than the theoretical quantity of permanganate gave unchanged material (0·5 g.) and a small quantity of liquid acids.

A mixture of the oil (0·24 g.), alcohol (10 c.c.), hydroxylamine hydrochloride (0·07 g.), sodium acetate (0·14 g.), and water (3 c.c.) was refluxed for 10 hours. A crystalline product (80 mg.) separated on the addition of water and cooling; this crystallised from aqueous alcohol in colourless needles, m. p. 78°. The crystals retained microscopic droplets of oil which could not be removed by recrystallisation, and the analytical results were unsatisfactory (Found: C, 73·5; H, 9·9; MeO, 15·8. $C_{21}H_{35}O_3N$ requires C, 72·2; H, 10·0; 2MeO, 17·8%).

Demethylation of the supposed dimethyl ether with aluminium bromide in benzene solution gave a highly coloured product, insoluble in alkali and in light petroleum, from which no crystalline substance could be isolated.

Dihydroflavoglaucin Monomethyl Ether.—Auroglaucin monomethyl ether (3·2 g.) in alcohol (50 c.c.) was shaken for 2 hours with hydrogen at 2—3 atms. in the presence of palladised strontium carbonate (1 g. of 2%). The volume of hydrogen absorbed corresponded to $3·8H_2$, and the colour of the solution changed from orange to pale yellow. Evaporation of the filtered alcoholic solution left a yellow oil (deep yellowish-brown coloration with alcoholic sodium hydroxide; permanent dark green coloration with alcoholic ferric chloride), which was characterised as its 2 : 4-*dinitrophenylhydrazone*, prepared by the use of Brady's reagent. The derivative was nearly insoluble in alcohol and crystallised from glacial acetic acid in small, deep red needles, m. p. 193° (Found: C, 62·3; H, 7·3; N, 11·3. $C_{26}H_{36}O_6N_4$ requires C, 62·4; H, 7·2; N, 11·2%).

Oxidation of Dihydroflavoglaucin Monomethyl Ether by Hydrogen Peroxide.—The conditions specified by Dakin (*Amer. Chem. J.*, 1909, **42**, 477) were modified owing to the insolubility of the substance in aqueous alkalis. The monomethyl ether (1 part), 20% sodium hydroxide solution (equivalent to 3 parts NaOH), and perhydrol (1½ parts of H_2O_2) were mixed with enough alcohol to form a homogeneous solution. On heating to 30—35° in an atmosphere of nitrogen, a vigorous reaction set in, and the temperature rose spontaneously to 60—65° and a solid separated. After being maintained at 60° for ½ hour, the mixture was cooled, the liquid rapidly filtered, and the precipitate washed with a little alcohol. The combined filtrates were immediately methylated by the addition of methyl sulphate equivalent to the sodium hydroxide originally present and by heating on the steam-bath. After two further additions of equivalent quantities of methyl sulphate and sodium hydroxide with continued heating, an excess of sodium hydroxide was added, and most of the alcohol removed in a vacuum. The oil so liberated was taken up in ether, and the solution shaken twice with dilute aqueous sodium hydroxide, dried, and evaporated. The dark yellow residue was distilled, giving a pale yellow oil, b. p. 160—165°/0·02 mm. (Found: C, 71·9; H, 10·3. $C_{14}H_{22}O_3$ requires C, 70·6; H, 9·2. $C_{15}H_{24}O_3$ requires C, 71·4; H, 9·6%). It is possible that a methyl group has entered the aromatic nucleus and this may explain the curious inertness that the substance exhibits towards nitric acid.

The oil contained a trace of a ketone, presumed to be dihydroflavoglaucin dimethyl ether because it developed a red coloration with Brady's reagent. It was insoluble in alkali and gave no ferric reaction.

The precipitate from the oxidation process consisted of a sodium salt and this was decomposed with a slight excess of hydrochloric acid. The brown oily acid was separated from the aqueous layer and converted into its *p*-phenylphenacyl ester. This crystallised from methyl alcohol (norit) in colourless rhombic plates, m. p. 66—67° alone or mixed with an authentic specimen prepared in the same way from *n*-octoic acid (Found: C, 78·3; H, 7·7; side-chain Me by the Kuhn–Roth method, 4·9, 5·1. Calc. for $C_{22}H_{26}O_3$: C, 78·1; H, 7·7; 1Me, 4·4%).

Decahydroauroglaucin (*Tetrahydroflavoglaucin*).—A solution of auroglaucin (5 g.) in alcohol (100 c.c.) was shaken with hydrogen under 2—3 atms. in the presence of palladised strontium carbonate (3 g. of 2%). A volume equivalent to $4H_2$ was rapidly absorbed, and a fifth mol. more slowly, but measurable absorption was complete in 1 hour, and shaking was continued for 1 hour more. The orange solution became very pale yellow as the reduction proceeded. After filtration, the alcohol was removed in a vacuum in a stream of nitrogen. The dark residual oil solidified on cooling in ice in the presence of light petroleum, and the substance crystallised from light petroleum in colourless needles (3·6 g.), m. p. 85° (Found: C, 74·1; H, 10·5; side-chain Me, 10·9. $C_{19}H_{32}O_3$ requires C, 74·0; H, 10·4; 2·2Me, 10·7%). The substance became coloured on exposure to air, and rapidly decomposed in contact with aqueous sodium hydroxide. With alcoholic ferric chloride a very transient green colour was observed, and when treated with alcoholic sodium hydroxide it gave a dark yellowish-brown coloration which faded rapidly.

The non-crystalline residue from the reduction had a permanent green ferric reaction, and a ketonic group was present (2 : 4-dinitrophenylhydrazone). It was assumed to be largely dihydroflavoglaucin.

Triacetyl derivative. A mixture of tetrahydroflavoglaucin (0·2 g.), acetic anhydride (15 c.c.), and sodium acetate (0·6 g.) was boiled for 10 minutes and then poured into water; the product separated as an oil, which solidified on keeping. It crystallised from aqueous methyl alcohol in colourless needles, m. p. 70° (Found: C, 68·8; H, 8·6. $C_{25}H_{38}O_6$ requires C, 69·1; H, 8·5%. Found: Ac by oxidation, 52·0. 3Ac and 2·2 side-chain Me require Ac, 51·5%).

Oxidation of *triacetyl tetrahydroflavoglaucin* by means of chromic anhydride in acetic anhydride solution resulted in the formation of some liquid fatty acid (presumably *n*-octoic acid) and probably a neutral ketone, but no other product could be isolated.

Tetrahydroflavoglaucin Dimethyl Ether.—Tetrahydroflavoglaucin (1 g.) in acetone (15 c.c.) was shaken for 1 hour at a time at room temperature with three successive quantities of methyl sulphate (1 c.c.) and aqueous sodium hydroxide (4 c.c. of 10%). The colour of the alkaline solution, at first deep brown, changed to yellow. An excess of sodium hydroxide was added, and most of the acetone removed in a vacuum. Addition of water liberated an oil, which solidified on cooling, and was crystallised from dilute alcohol; m. p. 78° (0·84 g.). Recrystallised from light petroleum, it formed thin, colourless needles, m. p. 79° [Found: C, 74·9; H, 10·5; MeO, 18·3. $C_{21}H_{36}O_3$ requires C, 75·0; H, 10·7; 2MeO, 18·5%. Found: active hydrogen (Zerewitinoff), 4·9, 5·4/21° in pyridine. 1H requires 5·1%. Found: side-chain Me, 8·2, 8·4. 1·85 side-chain Me requires 8·3%]. The substance was insoluble in aqueous sodium hydroxide,

developed no coloration in alcoholic sodium hydroxide, and exhibited no ferric reaction. Oxidation under a variety of conditions gave liquid acidic products similar to those obtained in other cases and no trace of an aromatic acid could be isolated.

A solution of the substance (0·41 g.) in alcohol (25 c.c.) was optically inactive ($\alpha \pm 0{\cdot}0020°$). The ultra-violet absorption spectrum of the substance in light petroleum solution was examined. The strong absorption was indicated by the high dilution of the solutions required. The curve obtained for variation of the extinction coefficient with the wave-length shows a well-marked maximum at 3000 A. and a minimum at 2500—2460 A. The curve is typical of a benzene ring system, but the maximum and minimum show a shift of about 200 A. towards the longer wave-length. This shift is typical of quinol derivatives and readily differentiates them from resorcinol and catechol derivatives (Klingstedt, *Compt. rend.*, 1922, **175**, 365).

Tetrahydrodeoxyflavoglaucin.—Zinc dust was added in eight portions (0·2 g. each) to a boiling solution of dihydroflavoglaucin (0·3 g.) in acetic acid (25 c.c.), at intervals of 2 minutes. The yellow colour almost disappeared, and after a further 5 minutes' boiling, the excess of zinc was collected and washed with a little hot acetic acid; the product precipitated by the addition of water was crystallised from aqueous alcohol (norit) and then from light petroleum, forming colourless hair-like needles, m. p. 113° (Found: C, 78·4; H, 10·5. Calc. for $C_{19}H_{32}O_2$: C, 78·1; H, 10·9%). This substance (m. p. 111°) had been previously obtained by the catalytic (platinum-black) hydrogenation of flavoglaucin (Part I, *loc. cit.*, p. 84) and characterised by its bisphenylurethane, m. p. 161°. The product of the above reduction similarly afforded the bisphenylurethane, m. p. 161°.

The substance rapidly darkened and decomposed on exposure to the air (odour of *n*-octoic acid); the ferric reaction was a very transient green coloration. With aqueous alcoholic sodium hydroxide it developed a deep yellowish-brown coloration, rapidly fading to pale yellow, and a trace of a crystalline solid was precipitated (sodium octoate?).

Tetrahydrodeoxyflavoglaucin Dimethyl Ether.—Tetrahydrodeoxyflavoglaucin (0·7 g.) in acetone (20 c.c.) was shaken at room temperature for 1 hour at a time with three successive equivalent quantities of methyl sulphate (0·5 c.c.) and aqueous sodium hydroxide (2 c.c. of 10%). Excess of sodium hydroxide was then added, and the reaction completed on the steam-bath. The product isolated by means of ether was a yellow viscous oil (0·5 g.), b. p. 175—180°/0·02 mm. (Found: C, 78·6; H, 11·2. $C_{21}H_{36}O_2$ requires C, 78·8; H, 11·2%).

The *dimethyl* ether (0·4 g.), dissolved in acetic acid (2 c.c.), was treated drop by drop with nitric acid (1 c.c., *d* 1·42) at room temperature. The red oily product was apparently a mixture of a quinone with some nitro-derivative. On reduction and treatment of the acid solution with sodium nitrite a diazonium salt was produced. This coupled with β-naphthol to a red azo-compound that dissolved in sulphuric acid to a scarlet solution.

Comparative Diazo-coupling Tests.—These tests were made in aqueous alcoholic solutions in the usual way and in all cases where a positive result is stated the azo-compound was isolated in a crude condition and found to dissolve in concentrated sulphuric acid to a scarlet solution.

	Diazotised *p*-nitroaniline.		Diazotised sulphanilic acid	
Substance.	Na_2CO_3.	NaOH.	Na_2CO_3.	NaOH.
Auroglaucin, flavoglaucin, and derivatives	Negative	Negative	Negative	Negative
Tetrahydrodeoxyflavoglaucin	Coupling masked by intense colour of solution; none after colour fades, and none in acetic acid			
Quinoctophenone	Couples	Couples	Couples	Couples
Quinoctophenone 5-methyl ether	Very weak	Couples	Negative	Couples
4-*n*-Amylquinoctophenone (IV)	Negative	Very weak	Negative	Negative
5-*n*-Amyl-2-octylquinol	Coupling masked by intense colour of solution; none in acetic acid			
2-Methyl-5-*n*-propylquinol				
6-Allylquinacetophenone	Couples	Couples	Couples	Couples
6-*n*-Propylquinacetophenone (III)	Couples	Couples	Couples	Couples
3-*n*-Amylquinoctophenone 5-methyl ether (V) ...	Negative	Negative	Negative	Negative

A description of the new synthetic compounds mentioned above will be published shortly.

Comparative Bromination Tests.—The amounts of bromine absorbed by a number of substances were estimated under the following conditions. The solutions used were N/50-potassium bromide, N/10-sulphuric acid, N/50-potassium iodide (not accurately standardised), N/100-potassium bromate, N/100-sodium thiosulphate (standard). A mixture of carbon tetrachloride (25 c.c.) and 25 c.c. each of the sulphuric acid and potassium bromate solutions was shaken in a stoppered flask for ½ hour. After 1 hour, 25 c.c. of the potassium iodide solution were

added and the iodine set free was titrated with thiosulphate. Also 25 c.c. of a carbon tetrachloride solution of a known weight of the substance were added to the above mixture and kept for 1 hour. Addition of potassium iodide and titration with thiosulphate as in the blank gave the amount of unused bromine.

The substances, and the number of mols. of bromine absorbed per mol. in 1 hour, were:

Thymol: 2·9 mols. (Jost and Richter, *Ber.*, 1923, **56**, 120, describe the tribromo-derivative formed in presence of an excess of bromine).
Dihydroflavoglaucin: 1·5 mols. (1·7 in 6 hours).
Tetrahydrodeoxyflavoglaucin: 1·6 mols. (1·9 in 6 hours).
4-*n*-Amylquinoctophenone 5-methyl ether: 1·7 mols.
4-*n*-Amylquinoctophenone: 1·7 mols.

Quantitative Micro-hydrogenation of Flavoglaucin and Auroglaucin.—We are indebted to Professor I. M. Heilbron, F.R.S., and Dr. H. Jackson for the following results. In the presence of Adams's platinum oxide catalyst in acetic acid at 21·3°/764·7 mm. (4·5 hrs.) and 24·5°/760·7 mm. (2·75 hrs.) the hydrogen absorbed by flavoglaucin corresponded to 7·13 and 7·03 double bonds respectively. This is interpreted as reduction of the double bond and the benzene nucleus, together with reduction of carbonyl to methylene and removal of one hydroxyl in the benzene ring. With the same catalyst and auroglaucin at 23·9°/760·5 mm. (1 hr.) and 24·4°/759·5 mm. (3 hrs.), hydrogen corresponding to 9·69 and 9·65 double bonds was absorbed. Taking into account the three additional double bonds, the results are in agreement with those for flavoglaucin. With a palladium-black catalyst, auroglaucin consumed 5·92 H_2 (saturation of four double bonds and $CO \rightarrow CH_2$), but the result for flavoglaucin was anomalous (3·54, 3·54 H_2) and indicated partial further reduction or perhaps impurity of the specimen.

3 : 6-*Dimethoxyphthalic Anhydride.*—3 : 6-Dihydroxyphthalonitrile (35 g.) (Pai and Guha, *J. Indian Chem. Soc.*, 1934, **11**, 231) was methylated with excess of methyl sulphate and alkali. It was found impossible to carry the methylation to completion. The 3 : 6-dimethoxyphthalonitrile was hydrolysed to the acid by alkali fusion, which gave a better yield than the method of Graves and Adams (*J. Amer. Chem. Soc.*, 1923, **45**, 2446). A mixture of the nitrile (30 g.), potassium hydroxide (100 g.), and water (15 c.c.) was cautiously liquefied in a metal crucible and heated until the vigorous evolution of ammonia ceased (20—30 minutes). The diluted solution was added with stirring to a slight excess of 5N-sulphuric acid cooled in ice, and the acid liquors were heated on the steam-bath to complete the precipitation of the anhydride. The solid product (25 g.) consisted of a mixture of acid and anhydride and was recrystallised from acetic anhydride. The mother-liquors were concentrated and treated with charcoal, and a further quantity of 3 : 6-dimethoxyphthalic anhydride obtained (total yield, 22 g.; m. p. 259—260°).

3 : 6 : 6′-*Trimethoxy*-2-m-*toluoylbenzoic Acid* (XI).—The method of Walsh and Weizmann (*loc. cit.*) for preparing 3 : 6-dichloro-2′-methoxy-*m*-toluoylbenzoic acid was followed. A mixture of 3 : 6-dimethoxyphthalic anhydride (20 g.), *o*-tolyl methyl ether (14 g.), carbon disulphide (150 c.c.), and aluminium chloride (45 g.) was refluxed on the steam-bath for 12 hours. On working up, a dark semi-solid acidic product (24 g.) was obtained. This was dissolved in warm alcohol; the solution, on cooling, deposited the *acid* in a crystalline state. Recrystallised from alcohol, it formed pale yellow prisms (7 g.), m. p. 216°. A second recrystallisation from alcohol (norit) gave colourless, rhombic prisms, m. p. 218° (Found: C, 65·2; H, 5·4. $C_{18}H_{18}O_6$ requires C, 65·5; H, 5·5%). The remainder of the crude product consisted mainly of a mixture of this acid with partially demethylated acids.

5 : 8-*Dihydroxy*-3-*methoxy*-2-*methylanthraquinone* (X; R = H, R′ = Me).—When a solution of the above acid (2·4 g.) in concentrated sulphuric acid (15 c.c.) was maintained at 150—160° for 10 minutes, the deep red colour changed to violet-blue. The product (1·4 g.) was isolated by pouring into water and boiling to coagulate the precipitate, and separated from a dark-coloured impurity by solution in hot acetic acid. The diluted solution deposited red microscopic needles (1 g.), m. p. indefinite above 170°. A solution of this product (0·1 g.) in alcohol (5 c.c.) was refluxed for 2 hours with sodium ethoxide (0·2 g. of sodium in 10 c.c. of alcohol) and methyl sulphate (1 g.). The diluted solution deposited deep red needles, which were recrystallised from aqueous acetic acid; m. p. 194—195°, unchanged after sublimation at 170—180°/1 mm. (Found: C, 67·5; H, 4·4. $C_{16}H_{12}O_5$ requires C, 67·6; H, 4·2%).

The yellow solution in acetic acid exhibits a faint green fluorescence. The substance is very

slightly soluble in aqueous sodium carbonate, and soluble in aqueous sodium hydroxide to a magenta solution showing a green fluorescence; the colour slowly fades to pale red. The solution in sulphuric acid is violet-blue in thick layers, magenta in thin, and has a green fluorescence.

3 : 5 : 8-*Trimethoxy-2-methylanthraquinone* (X; R = R′ = Me).—A mixture of the above crude product (0·5 g.), acetone (10 c.c.), methyl iodide (2 g.), and potassium carbonate (2 g.) was refluxed for 6 hours; the colour adsorbed on the potassium carbonate had then almost completely disappeared. A bright yellow solid separated on addition of water; it crystallised from alcohol in long, yellow needles, m. p. 231° (Found: C, 69·1; H, 5·0. $C_{18}H_{16}O_5$ requires C, 69·2; H, 5·1%).

The yellow solution in acetic acid does not fluoresce. The colour in sulphuric acid is deep blue in thick layers, violet-blue in thin layers, with faint green fluorescence.

3 : 5 : 8-*Trihydroxy-2-methylanthraquinone* (X; R = R′ = H).—The crude condensation product (0·4 g.) was treated with concentrated sulphuric acid (10 c.c.) at 150—160° for 20 minutes; the dark-coloured solid isolated by pouring into water and boiling crystallised from aqueous alcohol in deep orange, microscopic needles (0·1 g.), m. p. 254°. Recrystallisation from alcohol and acetic acid, and sublimation at 180—190°/1 mm., did not alter the m. p. The sublimate formed deep red needles, slowly changing to deep orange on keeping (Found: C, 61·7; H, 4·1. $C_{15}H_{10}O_5,1{\cdot}2H_2O$ requires C, 61·7; H, 4·2%. Found in material dried at 110°: C, 64·1; H, 3·6. $C_{15}H_{10}O_5,0{\cdot}6H_2O$ requires C, 64·1; H, 3·9%).

The yellow solution in acetic acid fluoresces green. The substance is soluble in aqueous sodium carbonate to a wine-red solution with faint green fluorescence, fading to pale red; the caustic alkali solution is crimson with green fluorescence, fading to pale red. The solution in sulphuric acid is magenta both in thick and in thin layers.

The *triacetyl* derivative, obtained by the action of boiling acetic anhydride containing a little sodium acetate during 5 minutes, crystallised from aqueous acetic acid in yellow needles, m. p. 196° (Found: C, 63·8; H, 4·2. $C_{21}H_{16}O_8$ requires C, 63·6; H, 4·0%).

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