## 761. *Griseofulvin. Part III.* The Structures of the Oxidation Products* $C_9H_9O_5Cl$ *and* $C_{14}H_{15}O_7Cl$.

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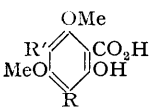
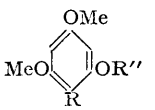
The oxidation products $C_9H_9O_5Cl$ and $C_{14}H_{15}O_7Cl$ are shown to be 3-chloro-2-hydroxy-4 : 6-dimethoxybenzoic (I; R = Cl, R′ = H) and 7-chloro-2-hydroxy-4 : 6-dimethoxycoumaran-3-one-2-β-butyric acid (IV) respectively, as originally suggested by Oxford, Raistrick, and Simonart (*Biochem. J.*, 1939, **33**, 240).

THE monobasic acid $C_9H_9O_5Cl$, m. p. 222° (decomp.), obtained as one product in the potassium permanganate oxidation of griseofulvin was shown by Oxford, Raistrick, and Simonart (*Biochem. J.*, 1939, **33**, 240) to give a purple ferric chloride colour typical of a salicylic acid, and to be derived from 2-hydroxy-4 : 6-dimethoxybenzoic acid. 3-Chloro-2-hydroxy-4 : 6-dimethoxybenzoic acid (I; R = Cl, R′ = H) was preferred to the 5-chloro-structure (I; R = H, R′ = Cl) on the basis of the negative Millon reaction which was taken to indicate the presence of an *oo*′-disubstituted phenol (Vaubel, *Z. angew. Chem.*, 1900, **13**, 1127). In view of the unreliability of the Millon test when applied to derivatives of sali-



(I)



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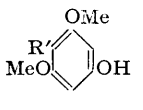


(III)

cylic acid, a formal proof of the orientation of the substituents in the griseofulvin oxidation product was essential. This has been achieved by decarboxylation of the acid $C_9H_9O_5Cl$ to a chlorophenol whose structure has been established as 2-chloro-3 : 5-dimethoxyphenol (II; R = Cl, R″ = H) by two unambiguous syntheses.

In the first synthesis, nitration of 3 : 5-dimethoxyphenol gave the steam-volatile 2-nitro-compound (II; R = $NO_2$, R″ = H) from which the corresponding amine was readily obtained by reduction, its structure as a 2-aminophenol being confirmed by the formation of a 2-methylbenzoxazole.

Attempts to convert the 2-aminophenol directly into the 2-chlorophenol by the Sandmeyer reaction were unsuccessful. Reduction of 3 : 5-dimethoxy-2-nitrophenyl acetate (II; R = $NO_2$, R″ = Ac) gave only 2-acetamido-3 : 5-dimethoxyphenol (II; R = NHAc, R″ = H) but reduction of the benzyl ether of 3 : 5-dimethoxy-2-nitrophenol gave

* Part II, preceding paper.

the amine (II; R = $NH_2$, R″ = $CH_2Ph$). By the Sandmeyer reaction the latter yielded the benzyl ether of 2-chloro-3 : 5-dimethoxyphenol, identical with the benzyl ether of the chlorophenol obtained from the griseofulvin oxidation product, $C_9H_9O_5Cl$.

In the second synthesis, methyl 4-hydroxy-2 : 6-dimethoxybenzoate (III; R′ = $CO_2Me$) (Pfeffer and Fischer, *Annalen*, 1912, **389**, 207) with sulphuryl chloride in chloroform afforded methyl 3-chloro-4-hydroxy-2 : 6-dimethoxybenzoate. Hydrolysis and decarboxylation were effected in one stage by treatment with concentrated sulphuric acid; the resultant 2-chloro-3 : 5-dimethoxyphenol (II; R = Cl, R″ = H) was identical with the chlorophenol obtained by decarboxylation of the acid $C_9H_9O_5Cl$.

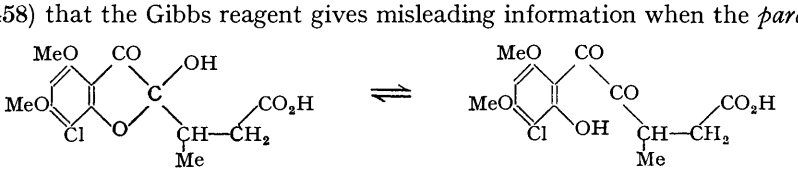
Moreover, methyl 3-chloro-4-hydroxy-2 : 6-dimethoxybenzoate is distinct from the methyl ester of the griseofulvin oxidation product $C_9H_9O_5Cl$.

Attempts to monochlorinate 4-hydroxy-2 : 6-dimethoxybenzoic acid yielded only the 3 : 5-dichloro-derivative, which gave 2 : 6-dichloro-3 : 5-dimethoxyphenol on decarboxylation. All attempts to prepare 4-chloro-3 : 5-dimethoxyphenol (III; R′ = Cl) from the corresponding 4-amino-compound failed. The amine was obtained, (*a*) by catalytic reduction of the 4-nitrophenol which in turn was isolated *inter alia* from the benzene-soluble portion of the non-steam-volatile product from the nitration of 3 : 5-dimethoxyphenol, and (*b*) by reduction of 3 : 5-dimethoxy-4-nitrosophenol prepared by the nitrosation of 3 : 5-dimethoxyphenol (cf. Weidel and Pollak, *Monatsh.*, 1900, **21**, 15).

The unambiguous syntheses of 2-chloro-3 : 5-dimethoxyphenol demonstrate conclusively that the acid $C_9H_9O_5Cl$ is 3-chloro-2-hydroxy-4 : 6-dimethoxybenzoic acid (I; R = Cl, R′ = H) as suggested by Oxford *et al.*; but the colour reactions frequently used in orientating phenols can often be misleading. This is borne out by our experience with the two series of compounds obtained by chlorination of 2-hydroxy-4 : 6-dimethoxybenzoic acid and 3 : 5-dimethoxyphenol respectively. Chlorination of 2-hydroxy-4 : 6-dimethoxybenzoic acid (or its silver salt) in carbon tetrachloride solution afforded 3-chloro-2-hydroxy-4 : 6-dimethoxybenzoic acid, identical with the griseofulvin oxidation product, and the isomeric 5-chloro-2-hydroxy-4 : 6-dimethoxybenzoic acid (I; R = H, R′ = Cl) together with small amounts of 3 : 5-dichloro-2-hydroxy-4 : 6-dimethoxybenzoic acid (I; R = R′ = Cl).

Treatment of 3 : 5-dimethoxyphenol in chloroform with sulphuryl chloride gave the steam-volatile 2- and the non-volatile 4-chloro-3 : 5-dimethoxyphenol (III; R′ = Cl). The 4-chlorophenol was also obtained by decarboxylation of 5-chloro-2-hydroxy-4 : 6-dimethoxybenzoic acid.

3 : 5-Dichloro-2-hydroxy-4 : 6-dimethoxybenzoic acid gave a negative Millon reaction but the 5-chloro-compound (I; R = H, R′ = Cl) gave the yellow colour normally obtained with salicylic acids with a free *o*-position. Both acids rapidly gave a strong blue colour with the 2 : 6-dibromoquinone-chloroimide reagent (Gibbs, *J. Biol. Chem.*, 1927, **72**, 649); the 3-chloro-compound (I; R = Cl, R′ = H) also gave a positive reaction with the reagent but rather surprisingly the colour developed considerably more slowly than was the case with the first two substances. Similarly, both 2- and 4-chloro-3 : 5-dimethoxyphenol gave positive reactions with the reagent, the colour developing more rapidly with the latter compound. There is an increasing body of evidence (Davidson, Keane, and Nolan, *Sci. Proc. R. Dublin Soc.*, 1943, **23**, 143; Calam, Clutterbuck, Oxford, and Raistrick, *Biochem. J.*, 1947, **41**, 458) that the Gibbs reagent gives misleading information when the *para*-position




to a hydroxyl group is blocked by a chlorine substituent, and the above results strongly emphasise this conclusion.

Oxford *et al.* (*loc. cit.*), considered the monobasic acid $C_{14}H_{15}O_7Cl$, the second product of the permanganate oxidation of griseofulvin, to be 7-chloro-2-hydroxy-4 : 6-dimethoxy-

coumaran-3-one-2-β-butyric acid (IV*a*), although little evidence was presented in support of this and the mechanism of its formation from griseofulvin was left obscure. The ready formation of the neutral lactone A, $C_{14}H_{13}O_6Cl$, m. p. 220° (Part II), suggested the presence of a γ-hydroxy-carboxylic acid grouping; in addition Oxford *et al.* showed that the acid contained two methoxyl groups and considered that the slow formation of a precipitate with Brady's reagent indicated the presence of a carbonyl group. The presence of these functional groupings has been confirmed. Additional evidence for the carboxyl group comes from the positive hydroxamic acid reaction and from the formation of a methyl ester. Warming with excess of alkali, followed by back-titration, failed to disclose the presence of a second acidic grouping. Although all attempts to prepare a carbonyl derivative failed, the presence of a keto-group is demonstrated by the infra-red spectrum of the methyl ester (Fig. 1) which clearly shows bands at 1735 cm.$^{-1}$ arising from the saturated ester (Grove and Willis, *J.*, 1951, 877) and at 1705 cm.$^{-1}$ attributed to the coumaranone ring. The infra-red spectrum of the acid $C_{14}H_{15}O_7Cl$ (Fig. 1) shows only one intense band (at 1710 cm.$^{-1}$) in the 6-μ region, the characteristic >C=O frequencies of the carboxylic acid residue and the coumaranone ring overlapping. A doublet with maxima at 2640 and 2540 cm.$^{-1}$ in the spectrum of this acid is attributed to –OH stretching vibrations in the carboxyl group.

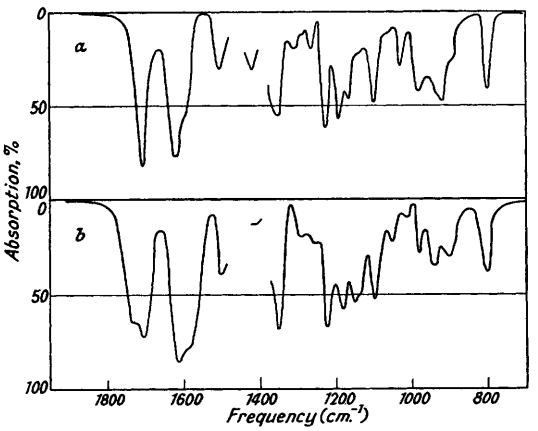



FIG. 1.

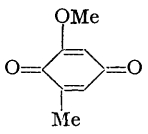
*a*, *7-Chloro-2-hydroxy-4:6-dimethoxycoumaran-3-one-2-β-butyric acid.*

*b*, *Methyl 7-chloro-2-hydroxy-4:6-dimethoxycoumaran-3-one-2-β-butyrate.*

Both the acid and its methyl ester show alcoholic hydroxyl absorption in the region of 3350 cm.$^{-1}$ (acid, 3333; ester, 3356 cm.$^{-1}$). Six of the seven oxygen atoms in this acid are contained in the carboxyl, two methoxyl, carbonyl, and hydroxyl groupings, leaving one unaccounted for, presumably in an ether linkage. Microhydrogenation showed the absence of ethylenic bonds. The ultra-violet absorption curve (Fig. 2) retains the main chromophoric system of griseofulvin and is characteristic of a compound in which phloroglucinol and carbonyl chromophores are conjugated. The acid $C_{14}H_{15}O_7Cl$ therefore contains an aromatic ring and it follows from what is known about the oxygen function that it must contain one further ring. A Kühn–Roth oxidation shows the presence of one *C*-methyl group. All the evidence set out above can be quoted in support of structure (IV*a*). Final proof of this structure (or of a similar one in which the methyl group is attached to the α- instead of to the β-carbon atom) comes from the oxidation with periodic acid, in which 1 mol. of periodate is consumed and the acid $C_{14}H_{15}O_7Cl$ is split quantitatively into 3-chloro-2-hydroxy-4:6-dimethoxybenzoic acid and (+)-methylsuccinic acid, identical with an authentic specimen.* (+)-Methylsuccinic acid is also isolated when the acid $C_{14}H_{15}O_7Cl$ is

* Comparison of the infra-red spectra of the natural and the synthetic (+)-methylsuccinic acids was carried out by Mr. T. A. Kletz, Imperial Chemical Industries Limited, Billingham Division, using a reflecting microscope attached to an infra-red spectrometer. A considerable difference existed between the spectrum of a sublimed sample of (+)-methylsuccinic acid and that of a specimen crystallised from a solvent. Similar differences existed in the spectra of specimens of (±)-methylsuccinic acid prepared by sublimation and crystallisation, and these also differed slightly from the corresponding spectra of the optically active material. This matter has been further investigated in these laboratories by Dr. L. A. Duncanson and is the subject of a separate communication (*J.*, 1952, 1753).

oxidised with chromic acid; the formation of 3-chloro-2-hydroxy-4 : 6-dimethoxybenzoic acid when the compound is oxidised with permanganate in acetone has already been reported (Part II).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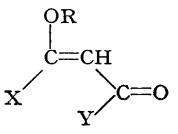
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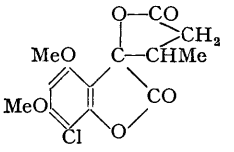
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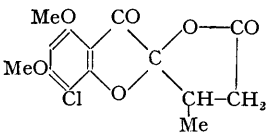
(VIII)

3-Methoxy-2 : 5-toluquinone (V), obtained by chromic oxide oxidation of griseofulvin (Part II), contains the enolic ether system (VI) which is believed to be present in griseofulvin (Part I) and is attacked in the permanganate oxidation. It follows that the keto-group at position 2 in 3-methoxy-2 : 5-toluquinone must arise by the rupture of a bond or bonds attaching the carbon atom at this position to the rest of the griseofulvin molecule and that the carbonyl group at position 5 in the quinone corresponds to the carboxyl group in the acid $C_{14}H_{15}O_7Cl$. The methyl group must therefore be in the β-position to the carboxyl group

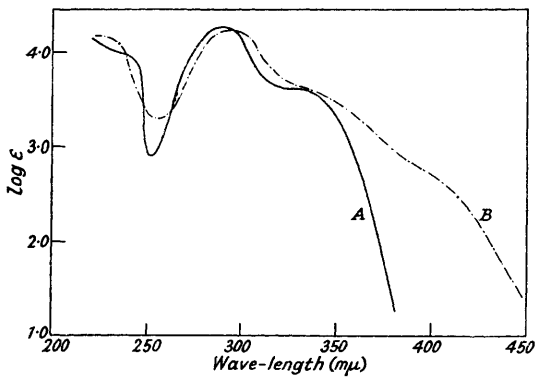


FIG. 2. *Ultra-violet absorption of 7-chloro-2-hydroxy-4 : 6-dimethoxycoumaran-3-one-2-β-butyric acid :*

*A, in methanol;*
*B, in 0·1N-sodium hydroxide.*

in this acid which has the 7-chloro-2-hydroxy-4 : 6-dimethoxycoumaran-3-one-2-β-butyric acid structure (IV*a*) proposed by Oxford *et al.*

Structure (IV) should be capable of showing tautomerism and although the compound exists in the cyclic semiacetal form (IV*a*) (cf. Erdtman, *Research*, 1950, **3**, 63) in the solid state (as shown by the alcoholic hydroxyl absorption in the infra-red spectrum) and in methanol solution (as shown by the ultra-violet absorption spectrum), the presence of the open chain form (IV*b*) is demonstrated by the absorption curve in alkaline solution (Fig. 2, band near 420 mμ characteristic of −CO·CO−). 7-Chloro-2-hydroxy-4 : 6-dimethoxycoumaran-3-one-2-β-butyric acid is stable to acid hydrolysis but in boiling 3N-sodium hydroxide undergoes benzilic acid rearrangement to a dilactone (VII) isomeric with lactones A and B (VIII) (Part II).

## EXPERIMENTAL

M. p.s are corrected. Microanalyses are by Drs. Weiler and Strauss, Oxford, and by Mr. W. Brown.

*Nitration of* 3 : 5-*Dimethoxyphenol.*—To a well-stirred solution of 3 : 5-dimethoxyphenol (25 g.) in acetic acid (100 ml.) and acetic anhydride (60 ml.) at −5° to 0°, there was added dropwise (½ hour) nitric acid (*d*, 1·42; 16 g.) in acetic acid (50 ml.). After 2 hours at room temperature the mixture was poured over cracked ice (500 g.). The combined crude product from two experiments was steam-distilled. The first litre of distillate, deep orange and containing most of the acetic acid, was discarded and distillation continued until no more solid crystallised from the distillate (60 l.). The 3 : 5-dimethoxy-2-nitrophenol (13·0 g., 20%) which separated at 0° was collected and recrystallised from ethanol in yellow needles or plates, m. p. 130° (Found : C, 48·2, 48·3; H, 4·7, 4·5; N, 7·2, 7·2. $C_8H_9O_5N$ requires C, 48·2; H, 4·6; N, 7·0%). 3 : 5-Dimethoxy-2-nitrophenol formed an orange solution in aqueous sodium hydroxide and gave a

brownish colour with ferric chloride in ethanol. The *acetate* formed pale yellow prisms, m. p. 123°, from ethanol (Found : C, 50·1; H, 4·7; N, 5·9. $C_{10}H_{11}O_6N$ requires C, 49·8; H, 4·6; N, 5·8%). The O-*benzyl ether* formed pale yellow needles, m. p. 100°, from ethanol (Found : C, 62·5; H, 5·3; N, 4·75. $C_{15}H_{15}O_5N$ requires C, 62·3; H, 5·2; N, 4·8%).

The pitch-like residue from the steam-distillation was separated from the residual aqueous liquor by decantation, dried by the repeated addition and evaporation of benzene, and then extracted for 6 hours with benzene in a Soxhlet apparatus. The benzene-insoluble material (30 g.) was discarded. The dried benzene-soluble extract was concentrated to 300 ml. and chromatographed on alumina (pH 4; grade II). The following bands (from bottom to top) were obtained and eluted with benzene : (i) A yellow band which passed rapidly down the column and furnished on recovery yellow needles (0·30 g.), m. p. 256° (decomp.), of 2 : 6-dimethoxy-*p*-benzoquinone (Found : C, 57·5; H, 5·1. Calc. for $C_8H_8O_4$ : C, 57·1; H, 4·8%). (ii) A band, colourless in daylight but showing a pale blue fluorescence in ultra-violet light, which yielded only gum (20 mg.). (iii) A yellow band giving, on recovery, 3·5 g. of a yellow solid, m. p. 125—170°, which was separated by fractional crystallisation from benzene into 3 : 5-dimethoxy-2-nitrophenol, m. p. 128—130° (1·05 g.), and 3 : 5-*dimethoxy*-4-*nitrophenol* (1·2 g., 2%), lemon yellow needles (from benzene), m. p. 168° (Found : C, 48·2; H, 4·7; N, 6·8. $C_8H_9O_5N$ requires C, 48·2; H, 4·6; N, 7·0%) [*acetate*, almost colourless prisms (from ethanol), m. p. 101° (Found : C, 50·1; H, 4·7; N, 5·8. $C_{10}H_{11}O_6N$ requires C, 49·8; H, 4·6; N, 5·8%)]. The remaining bands were eluted with benzene + 1% of methanol and the substances isolated from them will be reported later.

3 : 5-*Dimethoxy*-4-*nitrosophenol*.—3 : 5-Dimethoxyphenol (2·0 g.), in methanol (2 ml.) and concentrated hydrochloric acid (2 ml.), at 0°, was treated in portions, with shaking, with sodium nitrite (1·0 g.) in water (4 ml.). When addition was complete the 4-nitroso-compound was collected, washed with water, and crystallised from acetic acid, forming brick-red needles (1·0—1·6 g.), decomp. above 220°. (Weidel and Pollak, *loc. cit.*, give decomp. 222°.)

2-*Amino*-3 : 5-*dimethoxyphenol*.—3 : 5-Dimethoxy-2-nitrophenol (3·5 g.) in ethanol (200 ml.) was reduced by shaking it with hydrogen at room temperature in the presence of a Raney nickel catalyst (0·5 g.) (absorption : 3 mols. in 2 hours). After filtration and evaporation of the solvent under reduced pressure in nitrogen, the brown product was sublimed at 120°/$10^{-3}$ mm., yielding colourless crystalline 2-*amino*-3 : 5-*dimethoxyphenol*, decomp. 150° after darkening at 142° (Found : C, 57·1; H, 6·6; N, 8·3. $C_8H_{11}O_3N$ requires C, 56·8; H, 6·55; N, 8·3%). It slowly darkened, and was rapidly oxidised in solution in ethanol in the presence of air. The hydrochloride formed colourless needles, m. p. 205° (decomp.), from 15% hydrochloric acid (Weidel and Pollak, *loc. cit.*, give m. p. 205—206°) (Found : C, 42·8; H, 6·3. Calc. for $C_8H_{11}O_3N,HCl,H_2O$ : C, 42·95; H, 6·3%).

The neutral ON-*diacetyl* derivative, obtained by heating 2-amino-3 : 5-dimethoxyphenol (0·2 g.) under reflux with acetic anhydride (2 ml.) for 1 hour, formed colourless prisms (0·18 g.), m. p. 93°, from ethanol (Found : C, 57·2; H, 5·9; N, 4·8, 5·1; OMe, 21·2; Ac, 32·2%; *M*, 226. $C_{12}H_{15}O_5N$ requires C, 56·9; H, 6·0; N, 5·5; 2OMe, 24·5; 2Ac, 34·0%; *M*, 253). (The triacetyl compound $C_{14}H_{17}O_6N$ requires C, 56·9; H, 5·8; N, 4·75; 3Ac, 43·7%; *M*, 295.) The diacetyl derivative was insoluble in both dilute hydrochloric acid and in sodium hydroxide.

4 : 6-*Dimethoxy*-2-*methylbenzoxazole*.—2-Amino-3 : 5-dimethoxyphenol (0·18 g.) and acetic anhydride (2 ml.) were heated under reflux for 24 hours, excess of acetic acid removed, and the residual oil heated at 220° for several minutes (distillation of volatile material and darkening of residue). The ethereal solution of the residue was extracted with 3N-hydrochloric acid, and the extract basified, affording a brown oil which crystallised from light petroleum (b. p. 60—80°) in colourless needles (43 mg.), m. p. 78°, of 4 : 6-*dimethoxy*-2-*methylbenzoxazole* (Found : C, 62·4; H, 5·8; N, 7·5. $C_{10}H_{11}O_3N$ requires C, 62·1; H, 5·7; N, 7·3%). The neutral ethereal fraction gave on recovery the above diacetyl compound, m. p. 93° (10 mg.).

4-*Amino*-3 : 5-*dimethoxyphenol*.—(*a*) 3 : 5-Dimethoxy-4-nitrophenol (0·7 g.) in ethanol (40 ml.) absorbed 3 mols. of hydrogen at room temperature in the presence of a Raney nickel catalyst (18 hours). After removal of the catalyst and evaporation under reduced pressure in nitrogen the product was sublimed at 130°/$10^{-3}$ mm., giving a colourless sublimate (0·45 g.) of 4-*amino*-3 : 5-*dimethoxyphenol*, which crystallised from ethyl acetate in needles, m. p. in a sealed tube 165—180° (decomp.) (Found : C, 56·9; H, 6·7. $C_8H_{11}O_3N$ requires C, 56·8; H, 6·55%). The hydrochloride, decomp. from 200°, was obtained in colourless needles from 15% hydrochloric acid (Weidel and Pollak, *loc. cit.*, give m. p. 171—173°) (Found : C, 43·0; H, 6·1; Cl, 15·8. Calc. for $C_8H_{11}O_3N,HCl,H_2O$ : C, 42·95; H, 6·3; Cl, 15·8%).

The ON-*diacetyl* derivative, insoluble in both dilute sodium hydroxide solution and dilute

hydrochloric acid, formed colourless plates, m. p. 169°, from ethanol (Found : C, 56·9; H, 5·9; N, 5·0, 5·0; Ac, 36·5. $C_{12}H_{15}O_5N$ requires C, 56·9; H, 6·0; N, 5·5; 2Ac, 34·0%).

(*b*) 3 : 5-Dimethoxy-4-nitrosophenol (1·0 g.) in 30% aqueous ammonia (10 ml.) was filtered from insoluble material and saturated with hydrogen sulphide until the dark red colour was discharged (½ hour) and thereafter for ½ hour. The colourless needles of the 4-aminophenol were collected (800 mg.), decomposing above 170°. The diacetyl derivative formed colourless needles (from methanol), m. p. 167°, not depressed by the material obtained as in (*a*) above.

*Reduction of 3 : 5-Dimethoxy-2-nitrophenyl Acetate.*—(a) *With Raney nickel.* The acetate (5·5 g.) in ethanol (500 ml.) was hydrogenated as described above for the reduction of 3 : 5-dimethoxy-2-nitrophenol. Removal of the solvent left a greenish-brown solid (3·0 g.), m. p. 110—112°, immediately soluble in 3N-sodium hydroxide and only slowly dissolving in 3N-hydrochloric acid; on sublimation at 150°/$10^{-3}$ mm. followed by crystallisation from ether, it gave colourless prisms of 2-*acetamido*-3 : 5-*dimethoxyphenol*, m. p. 112—114° (Found : C, 57·1; H, 6·4; N, 7·0. $C_{10}H_{13}O_4N$ requires C, 56·8; H, 6·2; N, 6·6%). It gave a green colour with ferric chloride. Acetylation under reflux with acetic anhydride yielded the diacetate, m. p. 93°, identical with the diacetate obtained above from 2-amino-3 : 5-dimethoxyphenol.

(b) *With Adams's catalyst.* The acetate (0·8 g.) in ethanol (55 ml.) in the presence of Adams's platinum oxide catalyst (0·08 g.) slowly absorbed 120 ml. of hydrogen during 10 hours and reduction then ceased (Calc. : $3H_2$, 250 ml.). After evaporation under reduced pressure in nitrogen, the dark solid residue was taken up in ether and extracted with 3N-hydrochloric acid.

The ethereal layer yielded, on recovery and crystallisation from ethanol, unchanged starting material, m. p. 123° (0·20 g.). The acid washings were cooled to 0°, neutralised with ice-cold 3N-sodium hydroxide and extracted with ether. The dark brown solid obtained on recovery was now insoluble in 3N-hydrochloric acid and crystallised from light petroleum (b. p. 60—80°) (charcoal) in prisms, m. p. 108—111° (60 mg.), not depressed by admixture with 2-acetamido-3 : 5-dimethoxyphenol.

*Attempted Sandmeyer Reactions.*—(a) *With 2-amino-3 : 5-dimethoxyphenol.* The diazonium hydrochloride and sulphate were obtained in solution in the usual way but attempts, using a wide variety of experimental conditions, to obtain 2-chloro-3 : 5-dimethoxyphenol by the action of cuprous chloride were unsuccessful.

(b) *With 4-amino-3 : 5-dimethoxyphenol.* Diazotisation followed by treatment with cuprous chloride gave only 2 : 6-dimethoxy-*p*-benzoquinone as yellow needles, m. p. 256° (decomp.) (Found : C, 57·1; H, 5·1; OMe, 37·2. Calc. for $C_8H_8O_4$ : C, 57·1; H, 4·8; 2OMe, 36·9%). This gave an intense purple colour with concentrated sulphuric acid. Diazotisation in the presence of stannous chloride (cf. Kozlov, *J. Gen. Chem., U.S.S.R.*, 1937, **7**, 1635), but not cupric sulphate, largely prevented the formation of the quinone but no 4-chloro-3 : 5-dimethoxyphenol was obtained on treatment of the diazo-solutions with cuprous chloride.

*Reduction of the Benzyl Ether* (II; R = $NO_2$, R″ = $CH_2Ph$).—To a boiling solution of the benzyl ether (3·00 g.) in ethanol (100 ml.) and water (120 ml.) was added in portions during 10 minutes, sodium hydrosulphite (dithionite) (15 g.) until the liquid was almost colourless. After removal of most of the ethanol under reduced pressure, the residual oily emulsion was rendered alkaline by the addition of 3N-sodium hydroxide (25 ml.) and extracted with ether. The ethereal extract was washed with 3N-sodium hydroxide and water and on recovery gave a pale brown oil which did not crystallise. On the addition of concentrated hydrochloric acid to the oil, the *hydrochloride* of the amine (II; R = $NH_2$, R″ = $CH_2Ph$) separated and was collected, washed with 3N-hydrochloric acid, and dried over solid sodium hydroxide (2·32 g., 76%). The hydrochloride tended to colour in air; after two crystallisations from 3N-hydrochloric acid (charcoal) it formed long colourless needles, m. p. 200—202° (decomp.) (rapid heating) (Found : C, 61·0; H, 5·6; Cl, 13·2; N, 5·5. $C_{15}H_{18}O_3NCl$ requires C, 60·9; H, 6·1; Cl, 12·0; N, 4·7%). The oily amine obtained on basification of the hydrochloride was insoluble in sodium hydroxide and gave a purple colour with ferric chloride solution. The *acetyl* derivative, formed when the free amine (100 mg.) was heated with acetic anhydride (0·5 ml.) and acetic acid (0·5 ml.) at 100° for 30 seconds, crystallised from aqueous ethanol in colourless plates (110 mg.), m. p. 146—147° (Found : C, 67·5, 67·8; H, 6·4, 6·3; $C_{17}H_{19}O_4N$ requires C, 67·8; H, 6·4%). The NN-*diacetyl* derivative was obtained when the free amine (0·2 g.) was heated under reflux with acetic anhydride (1 ml.) for 5 minutes. It formed colourless plates (231 mg.) (from aqueous ethanol), m. p. 111—112° (Found : C, 66·5; H, 6·2; N, 4·0. $C_{19}H_{21}O_5N$ requires C, 66·5; H, 6·2; N, 4·1%).

*Sandmeyer Reaction of the Amine* (II; R = $NH_2$, R″ = $CH_2Ph$).—The amine hydrochloride (590 mg., 2 millimols.) in N-hydrochloric acid (6 ml.) was diazotised in the usual way by the

addition of sodium nitrite (152 mg., 2·2 millimols.) in water (0·6 ml.). After filtration from a little dark amorphous material, the clear solution was added to a vigorously stirred solution of cuprous chloride in concentrated hydrochloric acid (3 ml., 3 millimols.) at 0° and the mixture allowed to warm to room temperature. Ether was then added and stirring continued for 15 minutes, after which the ether was replaced by benzene and the temperature raised to 80°. Stirring was continued for 1 hour, the benzene being replaced at intervals of 15 minutes. The combined organic extracts (40 ml.) were washed with water and, after removal of the solvent, the residue was taken up in ether and washed with 3N-sodium hydroxide and water. The dark oil obtained on recovery was distilled at 130°/$10^{-3}$ mm.; the pale yellow distillate (42 mg.) crystallised from ether in colourless prisms of *benzyl 2-chloro-3 : 5-dimethoxyphenyl ether*, m. p. 71—72° (21 mg.), identical by mixed m. p. and infra-red absorption spectrum with the benzyl ether of the chlorophenol obtained by decarboxylation of the griseofulvin oxidation product, $C_9H_9O_5Cl$ (see below) (Found : C, 64·6; H, 5·4; Cl, 12·6. $C_{15}H_{15}O_3Cl$ requires C, 64·6; H, 5·4; 12·7%).

*Decarboxylation of the Griseofulvin Oxidation Product* $C_9H_9O_5Cl$ (*3-Chloro-2-hydroxy-4 : 6-dimethoxybenzoic Acid*).—The acid (0·1 g.) was heated in redistilled quinoline (5 ml.) with copper chromite (0·1 g.), whilst a stream of carbon dioxide-free nitrogen was passed through the apparatus. Evolution of carbon dioxide was complete in 45 minutes at a bath temperature of 160° (total $CO_2$ evolved = 1·04 mols.). After removal of the catalyst, the filtrate was poured into 3N-hydrochloric acid (25 ml.) and extracted continuously with ether for some hours. The ethereal extract was washed with 3N-hydrochloric acid, concentrated to 20 ml., and extracted first with saturated sodium carbonate solution and then with 3N-sodium hydroxide. The sodium hydroxide extract was acidified with concentrated hydrochloric acid and re-extracted with ether. Evaporation left a tar which partly solidified and then sublimed at 70°/10 $^2$ mm. The colourless sublimate (30 mg.; m. p. 54°) crystallised from light petroleum (b. p. 60—80°) in needles of *2-chloro-3 : 5-dimethoxyphenol*, m. p. 60°, identical with a synthetic specimen (Found : C, 51·2; H, 5·1; Cl, 18·9. $C_8H_9O_3Cl$ requires C, 50·9; H, 4·8; Cl, 18·8%). The carbonate extract on acidification and working up in a similar manner yielded a further 20 mg. of the same phenol, m. p. 58°. 2-Chloro-3 : 5-dimethoxyphenol gave a blue colour with ferric chloride and positive Millon's and Gibbs's reactions.

The benzyl ether (II; R = Cl, R″ = $CH_2Ph$) was prepared by treatment of the phenol (1·88 g.) with benzyl bromide (1·75 g.) and anhydrous potassium carbonate (2·3 g.) in boiling acetone (20 ml.) for 2½ hours, and the crude product (2·5 g.) after two crystallisations from ether was obtained in colourless prisms, m. p. 71—72°, identical with the material obtained from the Sandmeyer reaction on the amine (II; R = $NH_2$, R″ = $CH_2Ph$).

*Decarboxylation of 5-Chloro-2-hydroxy-4 : 6-dimethoxybenzoic Acid.*—The acid (179 mg.) was decarboxylated by the method described above, 0·83 mol. of carbon dioxide being evolved in 1 hour at 180°. The ethereal extract was extracted with 3N-sodium hydroxide and after acidification, recovery, and sublimation the colourless sublimate (35 mg., m. p. 115—140°) was fractionally crystallised from benzene, giving unchanged starting material, needles (5 mg.), m. p. 138—140°, and 4-chloro-3 : 5-dimethoxyphenol, prisms (10 mg.), m. p. 130—132°, not depressed by mixture with the specimen obtained by the direct chlorination of 3 : 5-dimethoxyphenol.

*Methyl Ester of the Griseofulvin Oxidation Product* $C_9H_9O_5Cl$ (*Methyl 3-Chloro-2-hydroxy-4 : 6-dimethoxybenzoate*).—The acid $C_9H_9O_5Cl$ (70 mg.), suspended in ether (50 ml.), was treated with diazomethane (from 5 g. of nitrosomethylurea) in ether (50 ml.) and kept overnight. The solid was collected, combined with the solid obtained by concentration of the ethereal mother-liquors, and successively extracted with saturated sodium hydrogen carbonate solution and 3N-sodium hydroxide. Acidification of the former yielded a trace of the acid $C_9H_9O_5Cl$, identified by a mixed m. p. determination. The sodium hydroxide extract on acidification yielded *methyl 3-chloro-2-hydroxy-4 : 6-dimethoxybenzoate*, colourless needles (40 mg.) (from ethyl methyl ketone), m. p. 186—187° (Found : C, 48·5; H, 4·3; Cl, 14·05. $C_{10}H_{11}O_5Cl$ requires C, 48·8; H, 4·5; Cl, 14·4%). The neutral material crystallised from light petroleum (b. p. 60—80°) in colourless needles (15 mg.), m. p. 126—127° undepressed on admixture with methyl 3-chloro-2 : 4 : 6-trimethoxybenzoate (Calam and Oxford, *J.*, 1939, 280).

*Methyl 4-hydroxy-2 : 6-dimethoxybenzoate*, prepared from 2 : 4 : 6-trihydroxybenzoic acid in 60% overall yield by the method of Pfeffer and Fischer (*loc. cit.*), formed colourless needles (from aqueous methanol), m. p. 184—186° (Found : C, 56·8; H, 5·8; OMe, 41·0. Calc. for $C_{10}H_{12}O_5$ : C, 56·6; H, 5·7; 3OMe, 43·9%).

*Methyl 3-Chloro-4-hydroxy-2 : 6-dimethoxybenzoate.*—The above ester (200 mg.) in dry chloroform (35 ml.) was treated with sulphuryl chloride (135 mg.) in chloroform (1 ml.) and set aside

overnight. After being washed with water, the dried chloroform solution was concentrated, yielding the 3-*chloro*-compound (200 mg.), m. p. 153—154°, colourless needles from benzene (Found: C, 49·0; H, 4·5; Cl, 14·1. $C_{10}H_{11}O_5Cl$ requires C, 48·8; H, 4·5; Cl, 14·4%). This chloro-ester was distinct (mixed m. p. and infra-red spectrum) from the methyl ester of the griseofulvin oxidation product $C_9H_9O_5Cl$ described above.

2-*Chloro*-3 : 5-*dimethoxyphenol*.—The above chloro-compound (100 mg.) was dissolved in ice-cold sulphuric acid (0·1 ml.) and the solution, after being kept at 30° for 2 hours, was poured on crushed ice (500 mg.). The diluted reaction mixture was then heated at 100° for several minutes, cooled, and extracted with ether. The recovered gum was sublimed at 75°/0·1 mm., affording 2-chloro-3 : 5-dimethoxyphenol which crystallised from light petroleum (b. p. 40—60°) in colourless needles (60 mg.), m. p. 60—61° undepressed on admixture with the phenol, m. p. 60°, obtained from griseofulvin.

3 : 5-*Dichloro*-4-*hydroxy*-2 : 6-*dimethoxybenzoic Acid*.—(*a*) 4-Hydroxy-2 : 6-dimethoxybenzoic acid (396 mg.), m. p. 177° (decomp.), prepared by hydrolysis of the corresponding ester described above, was suspended in dry carbon tetrachloride (20 ml.) and treated with chlorine (150 mg.) in carbon tetrachloride (5 ml.). After 3 hours, the solid was collected, extracted with hot carbon tetrachloride (15 ml.), and filtered from unchanged starting material (180 mg.). On cooling, the carbon tetrachloride extract furnished the 3 : 5-*dichloro*-derivative as colourless needles (110 mg.) m. p. 125° (Found: C, 41·1; H, 3·3; Cl, 25·8. $C_9H_8O_5Cl_2$ requires C, 40·45; H, 3·0; Cl, 26·6%).

(*b*) 4-Hydroxy-2 : 6-dimethoxybenzoic acid (180 mg.), suspended in carbon tetrachloride (30 ml.), was heated under reflux with chlorine (250 mg.) in carbon tetrachloride (15 ml.) until complete solution was effected. By next morning the solution had deposited the 3 : 5-dichloro-compound in colourless needles (150 mg.), m. p. 125°.

2 : 6-*Dichloro*-3 : 5-*dimethoxyphenol*.—(*a*) 3 : 5-Dichloro-4-hydroxy-2 : 6-dimethoxybenzoic acid (100 mg.) in quinoline (2 ml.) was heated at 125—130° with copper chromite (40 mg.) in a stream of nitrogen. The evolution of carbon dioxide was complete in 1 hour. The cooled quinoline solution was poured into 3N-hydrochloric acid and extracted with ether. Recovery of the ethereal extract gave 2 : 6-*dichloro*-3 : 5-*dimethoxyphenol* (60 mg.), colourless needles [from benzene–light petroleum (b. p. 60—80°)], m. p. 147—149° (Found: C, 43·3; H, 3·9; Cl, 31·4. $C_8H_8O_3Cl_2$ requires C, 43·05; H, 3·6; Cl, 31·8%).

(*b*) 3 : 5-Dichloro-4-hydroxy-2 : 6-dimethoxybenzoic acid (100 mg.) in 2N-hydrochloric acid (2 ml.) was heated under reflux in a stream of nitrogen. Evolution of carbon dioxide was complete in 1 hour. The dichlorophenol which had begun to crystallise from the reaction was collected after ½ hour and crystallised as above, forming colourless needles (60 mg.), m. p. 147—149°.

*Chlorination of* 3 : 5-*Dimethoxyphenol*.—3 : 5-Dimethoxyphenol (2·0 g.), in dry chloroform (15 ml.) was vigorously shaken while sulphuryl chloride (1·75 g.) in chloroform (10 ml.) was added dropwise. After 48 hours, the chloroform was removed by distillation and the residual brown gum was distilled in steam (500 ml. of distillate). The volatile colourless oil, recovered by ether-extraction, crystallised from light petroleum in long needles (300 mg.), m. p. 58—59°, undepressed on admixture with 2-chloro-3 : 5-dimethoxyphenol prepared above.

Crystallisation of the non-steam volatile fraction from benzene afforded 4-*chloro*-3 : 5-*dimethoxyphenol* as small colourless needles (240 mg.), m. p. 132—133° (Found: C, 51·0; H, 4·85; Cl, 18·95. $C_8H_9O_3Cl$ requires C, 50·9; H, 4·8; Cl, 18·8%). The *benzyl ether* crystallised from ether in colourless plates, m. p. 97—98° (Found: C, 64·6; H, 5·5; Cl, 12·4. $C_{15}H_{15}O_3Cl$ requires C, 64·6; H, 5·4; Cl, 12·7%). The 4-chloro-phenol readily sublimed at 110—120° (oil-bath) and 0·1 mm. Hg and gave a positive reaction with the Gibbs reagent, the blue colour developing much more rapidly than with the above 2-chloro-derivative.

The residue, obtained by recovery of the benzene mother-liquors from crystallisation of the 4-chloro-compound, partly distilled at 110° (oil-bath) and 0·1 mm. Hg, affording a further 650 mg. of the 2-chloro-compound. The residue from the distillation sublimed at 120° (oil-bath)/0·1 mm., giving the 4-chloro-derivative (200 mg.).

*Chlorination of* 2-*Hydroxy*-4 : 6-*dimethoxybenzoic Acid*.—(*a*) 2-Hydroxy-4 : 6-dimethoxybenzoic acid (2·0 g.) was dissolved in 1% sodium hydroxide solution (40 ml.), excess of silver nitrate (1·8 g.) in water added, and the white precipitate collected, washed with water, and dried to constant weight at 100—106° (2·54 g., 82%). The dried silver salt was suspended in dry carbon tetrachloride (30 ml.), and chlorine in carbon tetrachloride (10 ml., 0·67 g. of chlorine) added dropwise under anhydrous conditions. The mixture was heated cautiously on the water-bath and then under reflux for 3 hours. The hot solution was filtered and the solid material washed with sodium hydrogen sulphite solution (1%) and then with sodium hydrogen

carbonate solution (1%). The white solid, which was precipitated on acidification of the bicarbonate washings, was collected (1·35 g.); it had m. p. 190—220° and recrystallised from ethyl acetate, giving 3-chloro-2-hydroxy-4 : 6-dimethoxybenzoic acid as colourless needles, m. p. 220—223° (decomp.) undepressed on admixture with the acid $C_9H_9O_5Cl$ obtained from griseofulvin (Part II) (Found : C, 46·65; H, 4·2. Calc. for $C_9H_9O_5Cl$ : C, 46·5; H, 3·9%).

On cooling, the carbon tetrachloride filtrate deposited unchanged starting material, m. p. and mixed m. p. 150—157° (0·233 g.). Evaporation of the solution gave a yellow solid, m. p. 110—130° (0·42 g.), which was acidic and gave a purple colour in ethanol with ferric chloride. It was taken up in ether (20 ml.) and extracted with saturated sodium hydrogen carbonate solution (3 × 10 ml.). Acidification of this extract gave a white solid, m. p. 125—132°, which resisted further purification by crystallisation. Conversion into the silver salt and recovery followed by two crystallisations from water furnished colourless needles of *5-chloro-2-hydroxy-4 : 6-dimethoxybenzoic acid*, m. p. 139—140° (0·2 g.) (Found : C, 46·7; H, 4·0; Cl, 15·05; OMe, 27·0%; equiv., 240. $C_9H_9O_5Cl$ requires C, 46·5; H, 3·9; Cl, 15·2; 2OMe, 26·7%; equiv., 233). This acid gave a positive Gibbs reaction, a yellow colour with Millon's reagent, and a deep purple colour with ferric chloride in ethanol. On methylation in methanol solution with ethereal diazomethane, methyl 3-chloro-2 : 4 : 6-trimethoxybenzoate, m. p. 126°, was obtained, identical with a synthetic specimen.

(*b*) 2-Hydroxy-4 : 6-dimethoxybenzoic acid (2·0 g.) was suspended in carbon tetrachloride (20 ml.), and a solution of chlorine in the same solvent (10·8 ml., 0·72 g. of chlorine) slowly added at room temperature. The mixture was heated under reflux for 3 hours, then filtered hot, and the insoluble material (1·22 g.; m. p. 120—180°) extracted with boiling ethyl acetate (18 ml.). The ethyl acetate-insoluble fraction (0·28 g.) furnished on crystallisation from a larger volume of ethyl acetate colourless needles of 3-chloro-2-hydroxy-4 : 6-dimethoxybenzoic acid, m. p. 220° (decomp.). The ethyl acetate-soluble fraction, after extensive fractional crystallisation first from ethyl acetate and then from benzene, gave a further small fraction of 3-chloro-2-hydroxy-4 : 6-dimethoxybenzoic acid (50 mg.), unchanged 2-hydroxy-4 : 6-dimethoxybenzoic acid (26 mg.), a colourless substance, m. p. 175—180° (0·15 g.), and a large fraction (0·46 g.), m. p. 125—140°, which consisted mainly of 5-chloro-2-hydroxy-4 : 6-dimethoxybenzoic acid.

The colourless material, m. p. 175—180°, was sublimed at 140°/$10^{-4}$ mm. and then crystallised three times from benzene; it formed long colourless needles of *3 : 5-dichloro-2-hydroxy-4 : 6-dimethoxybenzoic acid*, m. p. 181—182° (Found : C, 40·6; H, 3·2; Cl, 27·4. $C_9H_8O_5Cl_2$ requires C, 40·5; H, 3·0; Cl, 26·6%). 3 : 5-Dichloro-2-hydroxy-4 : 6-dimethoxybenzoic acid gave a deep purple colour with ferric chloride in ethanol and a negative Millon reaction. With the Gibbs reagent a deep blue colour rapidly developed.

On cooling, the carbon tetrachloride mother-liquor from the reaction mixture deposited crystalline material (0·7 g.; m. p. 120—130°) from which 3 : 5-dichloro-2-hydroxy-4 : 6-dimethoxybenzoic acid (27 mg.), 2-hydroxy-4 : 6-dimethoxybenzoic acid (50 mg.), and impure 5-chloro-2-hydroxy-4 : 6-dimethoxybenzoic acid (0·25 g.; m. p. 125—140°) were obtained after fractional crystallisation from ethyl acetate and benzene.

*7-Chloro-2-hydroxy-4 : 6-dimethoxycoumaran-3-one-2-β-butyric Acid* (IV), prepared by the oxidation of griseofulvin with zinc permanganate as described in Part II, was fractionally crystallised from ethyl acetate. The first fraction which contained traces of 3-chloro-2-hydroxy-4 : 6-dimethoxybenzoic acid was rejected. Subsequent fractions were combined and recrystallised from ethyl methyl ketone–light petroleum (b. p. 60—80°) in needles, m. p. 190° (decomp.), and were sufficiently pure for degradative work although alkaline hydrolysis (see below) revealed the presence, in some batches only, of traces of 7-chloro-4 : 6-dimethoxycoumaranone-2-β-butyric acid. Further purification raised the m. p. to 204° (decomp.) (Found : C, 51·0; H, 4·7; OMe, 18·0; *C*-Me, 3·7%; equiv., 323. Calc. for $C_{14}H_{15}O_7Cl$ : C, 50·8; H, 4·6; 2OMe, 18·8; 1*C*-Me, 4·5%; equiv., 330·5), $[\alpha]_D^{19}$ −12° (*c*, 0·98 in acetone). Ultra-violet absorption max. : 331, 288, ~244 mμ, log ε 3·63, 4·28, 3·95 in methanol; ~405, ~330, 296, ~231 mμ, log ε 2·64, 3·63, 4·26, 4·18 in 0·1N-sodium hydroxide. It gave no colour with ferric chloride. It did not couple with diazotised amines and the Millon and the Gibbs reaction were negative. The hydroxamic acid test for the carboxyl group was positive.

Contrary to the findings of Oxford *et al.*, pure specimens of the acid did not reduce Fehling's solutions or ammoniacal silver nitrate. It did not form derivatives on treatment with Brady's reagent or with phenylhydrazine and was recovered unchanged after attempted reaction with semicarbazide and with hydroxylamine. The acid was recovered unchanged after 1 hour's heating under reflux with 3N-sulphuric acid.

The acid (72·0 mg.) in 0·1032N-sodium hydroxide (5·00 ml.) was heated on the steam-bath

for 1 hour, cooled, and back-titrated with 0·0865N-hydrochloric acid (phenolphthalein). 3·78 ml. of acid were required, whence 1·83 ml. sodium hydroxide were used (Calc. for $1CO_2H$ : 2·08 ml.).

The *methyl* ester, obtained by reaction with 10% methanolic hydrochloric acid, sublimed at $120°/10^{-4}$ mm. in prisms, m. p. 54—58°, not raised by further sublimation (Found : C, 52·3; H, 5·1; Cl, 10·1; OMe, 27·8. $C_{15}H_{17}O_7Cl$ requires C, 52·3; H, 5·0; Cl, 10·3; 3OMe, 27·1%). The methyl ester did not couple with diazotised nitraniline. It was insoluble in water and neutral to the Universal indicator but dissolved slowly in sodium carbonate and more rapidly in sodium hydroxide giving yellow solutions. It did not give a precipitate with Brady's reagent.

*Periodate Oxidation of* 7-*Chloro*-2-*hydroxy*-4 : 6-*dimethoxycoumaranone*-2-β-*butyric Acid.*—To the acid (311 mg.) in water (60 ml.), 0·092N-sodium periodate (160 ml.) and concentrated hydrochloric acid (2 ml.) were added. After 48 hours at room temperature, the precipitated solid was collected and washed with water, and the combined filtrate and washings were made up to 250 ml. (Titration of a 25 ml. portion of this solution indicated the consumption of 1·05 mols. periodate.) The precipitate (176 mg., 80%) crystallised from ethyl acetate in colourless needles (110 mg.), m. p. and mixed m. p. with 3-chloro-2-hydroxy-4 : 6-dimethoxybenzoic acid, 216—218° (decomp.).

The aqueous filtrate was continuously extracted with ether for 14 hours. The gum, obtained on recovery, sublimed at $100—103°/10^{-3}$ mm. as a yellow solid which was crystallised from benzene and resublimed (20 mg.). Crystallisation from water gave needles, m. p. 108—110° undepressed on admixture with synthetic (±)- and (+)-methylsuccinic acids and with the product of the chromic oxide oxidation of the acid $C_{14}H_{15}O_7Cl$ (see below). The acid had $[\alpha]_D^{19}$ +9·7° (*c*, 1·48 in water) (Found : C, 45·4; H, 6·1. Calc. for $C_5H_8O_4$ : C, 45·4; H, 6·1%). The infra-red spectrum of a sublimed specimen was identical with that of a sublimed specimen of authentic (+)-methylsuccinic acid.

*Chromic Oxide Oxidation of* 7-*Chloro*-2-*hydroxy*-4 : 6-*dimethoxycoumaranone*-2-β-*butyric Acid.*—The acid (400 mg.) in acetic acid (2 ml.) was heated on the water-bath with chromic oxide (1·0 g.) in acetic acid (8 ml.) and water (2 ml.). After 10 minutes the reaction mixture was diluted with water and continuously extracted with ether, affording on recovery a greenish solid. Re-extraction with ether gave a colourless solid which sublimed at $60—80°/10^{-3}$ mm. and, after crystallisation from benzene and resublimation, had m. p. 108—110° (30 mg.). The product did not depress the m. p.s of synthetic (±)- or (+)-methylsuccinic acids or the m. p. of the product obtained in the periodate oxidation (above) (Found : C, 45·3; H, 6·1%; equiv., 64·9. Calc. for $C_5H_8O_4$ : C, 45·4; H, 6·1%; equiv., 66). Available quantities of the product were insufficient to determine the optical rotation; however, the infra-red absorption spectrum was identical with that of sublimed synthetic (+)-methylsuccinic acid.

*Alkaline Hydrolysis of* 7-*Chloro*-2-*hydroxy*-4 : 6-*dimethoxycoumaranone*-2-β-*butyric Acid.*—The acid (0·50 g.) in 3N-sodium hydroxide (15 ml.) was heated under reflux for 2 hours in nitrogen. After acidification with 3N-sulphuric acid, the solution was steam-distilled but no volatile acid was detected in the distillate. The colourless solid (0·35 g.) which separated from the residual solution at 0°, was taken up in ether (solution A), and the insoluble material (20 mg.) rejected. Continuous ether-extraction of the filtrate and recovery furnished a gum which was redissolved in ether (solution B).

Solution A was extracted in turn with 1% aqueous sodium hydrogen carbonate and N-sodium hydroxide, and, on removal of the ether, the neutral fraction remained as a crystalline solid (74 mg.), m. p. 150—155°. After three crystallisations from ethanol it formed colourless prisms of a *dilactone* (VII), m. p. 165—168°. The compound was neutral to the Universal Indicator, and gave no colour with ferric chloride [Found : C, 53·9, 53·9; H, 4·1, 4·4; OMe, 19·5%; *M* (Rast), 298; equiv. (by lactone titration), 153. $C_{14}H_{13}O_6Cl$ requires C, 53·7; H, 4·2; 2OMe, 19·8%; *M*, 312·6; equiv., 156 (for a dilactone)]. Ultra-violet absorption max. : 290 mμ, (log ε 3·25) in methanol. The infra-red spectrum showed two bands in the >C=O region at 1815 and 1795 $cm.^{-1}$ ascribed to five-membered ring $\Delta^{\beta\gamma}$-lactone and unconjugated five-membered ring lactone, respectively.

The bicarbonate extract, after acidification and extraction with ether, furnished a solid (60 mg.), m. p. 165—175° (decomp.), separated by fractional crystallisation from ethyl methyl ketone–light petroleum (b. p. 60—80°) into unchanged starting material and the dibasic acid $C_{13}H_{13}O_6Cl$ (Part II), m. p. 190—198° (decomp.), identified by comparison of the infra-red spectra [Found : C, 51·4; H, 4·5; OMe, 9·8%; equiv., 159. Calc. for $C_{13}H_{13}O_6Cl$ : C, 51·9; H, 4·3; 1OMe, 10·3%; equiv., 150 (dibasic acid)].

The sodium hydroxide extract after acidification, extraction with ether, and recovery, afforded a *phenol* (2 mg.), crystallising in colourless needles, m. p. 205—208° (decomp.), from ethyl

acetate and giving a green colour with ferric chloride in ethanol. This substance was not investigated further.

Solution B after extraction with sodium hydrogen carbonate, recovery, and recrystallisation from ethanol yielded needles, m. p. and mixed m. p. with lactone A, $C_{14}H_{13}O_6Cl$ (Part II), 220° (10 mg.). The bicarbonate extract, after acidification and recovery, as before, furnished a mixture (50 mg.), m. p. 158—164° (decomp.), of acidic substances, including starting material, which resisted purification.

A similar yield of the dilactone $C_{14}H_{13}O_6Cl$ was obtained when the time of heating under reflux was 6 hours, but in this case the small acidic fractions appeared homogeneous and consisted of the dibasic acid $C_{13}H_{13}O_6Cl$ only.

Only starting material was recovered when 7-chloro-2-hydroxy-4 : 6-dimethoxycoumaranone-2-β-butyric acid was warmed with 40% sodium hydroxide solution for 10 minutes at 60° and then set aside for 48 hours at room temperature.

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