828. *The Chemistry of Fungi. Part XLIII.*[1] *The Synthesis of Aposclerotaminic Acid.*

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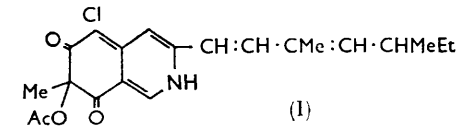
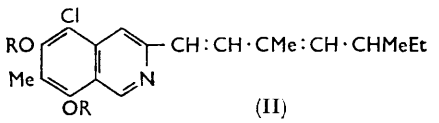
Aposclerotaminic acid (5-chloro-6,8-dihydroxy-7-methylisoquinoline-3-carboxylic acid) has been synthesised by standard procedures.

Our studies [2-5] concerning the fungal metabolite, sclerotiorin, have shown that reductive aromatisation of sclerotioramine (I) forms aposclerotioramine (II; R = H) by extrusion of the acetoxyl residue. Ozonolysis of di-*O*-acetylaposclerotioramine (II; R = OAc) and subsequent hydrolysis of the product furnishes aposclerotaminic acid which we have formulated as 5-chloro-6,8-dihydroxy-7-methylisoquinoline-3-carboxylic acid (III). Confirmation of this structure is now provided by the synthesis of (III).



(I)



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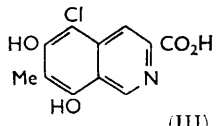

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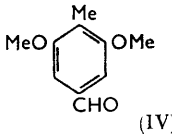

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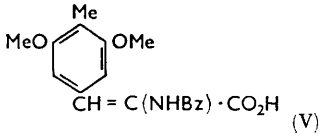

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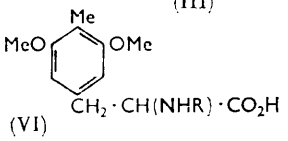

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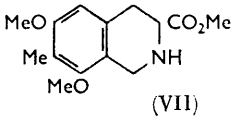

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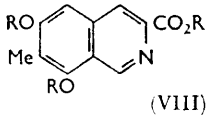

(VIII)

3,5-Dimethoxy-4-methylbenzaldehyde (IV) (prepared by the Rosenmund reduction of the corresponding acid chloride) was converted into α-benzamido-3,5-dimethoxy-4-methylcinnamic acid (V) by way of the corresponding oxazolone. Hydrogenation of the cinnamic

[1] Part XLII, Mehta and Whalley, *J.*, 1963, 3777.
[2] Eade, Page, Robertson, Turner, and Whalley, *J.*, 1957, 4913.
[3] Fielding, Graham, Robertson, Travers, and Whalley, *J.*, 1957, 4931.
[4] Fielding, Robertson, Travers, and Whalley, *J.*, 1958, 1814.
[5] Holker, Ross, Staunton, and Whalley, *J.*, 1962, 4150.

acid (V) readily furnished *N*-benzoyl-β-(3,5-dimethoxy-4-methylphenyl)alanine (VI; R = Bz) which was hydrolysed by acid to β-(3,5-dimethoxy-4-methylphenyl)alanine (VI; R = H). Cyclisation of (VI; R = H) with formalin followed by esterification of the crude product gave 1,2,3,4-tetrahydro-6,8-dimethoxy-3-methoxycarbonyl-7-methylisoquinoline (VII), which readily formed 6,8-dimethoxy-3-methoxycarbonyl-7-methylisoquinoline (VIII; R = Me) on dehydrogenation. The infrared spectrum (in $CHCl_3$) of (VIII; R = Me) on dehydrogenation. The infrared spectrum (in $CHCl_3$) of (VIII; R = Me) showed absorption at $\nu_{max.}$ 1709 (aromatic ester) cm.$^{-1}$, whilst the n.m.r. spectrum has singlets at τ 8·33 (aromatic *C*-Me, 3 protons), 6·68 (OMe; 6 protons), 6·58 (methyl group in ester; 3 protons), 3·69 (C-5 proton), 2·22 (C-1 proton), and 1·23 (C-4 proton).

A compound identical with (VIII; R = Me) was obtained from the di-*O*-acetate of aposclerotaminic acid by dechlorination with hydriodic acid to give the dechloro-compound (VIII; R = H), followed by esterification with methanol and hydrogen chloride, and subsequent *O*-methylation with methyl iodide and potassium carbonate in acetone. Under more forcing methylation conditions the methiodide of (VIII; R = Me) was the principal product. The total synthesis of aposclerotaminic acid was completed by using compound (VIII; R = Me), derived from sclerotiorin, as a relay. Thus, demethylation of (VIII; R = Me) with hydriodic acid gave (VIII; R = H), which was smoothly chlorinated with sulphuryl chloride to give aposclerotaminic acid.

The synthesis of aposclerotaminic acid provides collateral evidence for the structure previously derived for sclerotiorin and its derivatives.

Certain experiments with 3,5-dimethoxybenzaldehyde which were performed as a preliminary to the main synthesis are reported.

Experimental

β-(3,5-*Dimethoxy*-4-*methylphenyl*)*alanine Hydrochloride*.—Prepared from 3,5-dimethoxy-4-methylbenzoic acid (10 g.) by the use of thionyl chloride, 3,5-dimethoxy-4-methylbenzoyl chloride (9·5 g.) in xylene (50 ml.) containing 5% palladium–barium sulphate (1 g.) was heated under reflux (stirring) in a current of hydrogen. After evolution of the theoretical quantity of hydrogen chloride the solution was filtered and evaporated to yield an oil. Purification from light petroleum (b. p. 60—80°) gave 3,5-*dimethoxy*-4-*methylbenzaldehyde* (7·5 g.) in needles, m. p. 98° [Found: C, 67·0; H, 6·7; OMe, 34·7. $C_8H_6O(OMe)_2$ required C, 66·7; H, 6·7; OMe, 34·5%]. The 2,4-*dinitrophenylhydrazone* formed red needles, m. p. 259°, from ethyl acetate [Found: C, 53·4; H, 4·5; N, 15·3; OMe, 17·2. $C_{14}H_{10}N_4O_4(OMe)_2$ requires C, 53·3; H, 4·5; N, 15·6; OMe, 17·2%]. The *semicarbazone* separated from ethanol in needles, m. p. 211° (Found: C, 55·6; H, 6·3; N, 17·6. $C_{11}H_{15}N_3O_3$ requires C, 55·7; H, 6·4; N, 17·7%).

Prepared from the previous aldehyde (3 g.) in the usual manner 4-(3,5-dimethoxy-4-methylbenzylidene)-2-phenyl-5-oxazolone (3·8 g.) separated from acetone in orange plates, m. p. 202° [Found: C, 70·7; H, 5·2; N, 4·6; OMe, 19·4. $C_{17}H_{11}NO_2(OMe)_2$ requires C, 70·6; H, 5·3; N, 4·3; OMe, 19·2%]. A solution of this oxazolone (2 g.) in acetone (100 ml.) containing 2% aqueous sodium hydroxide (100 ml.) was refluxed for 2½ hr., and the acetone removed under reduced pressure. Addition of concentrated hydrochloric acid to the cooled hydrolysate gave α-*benzoyl*-3,5-*dimethoxy*-4-*methylcinnamic acid* (1·8 g.) which formed needles, m. p. 229°, from aqueous alcohol [Found: C, 67·1; H, 5·7; N, 4·2; OMe, 18·1. $C_{17}H_{13}NO_3(OMe)_2$ requires C, 66·9; H, 5·6; N, 4·1; OMe, 18·2%].

Hydrogenation of this cinnamic acid (2 g.) in acetic acid (50 ml.) containing 5% palladium–charcoal (1 g.) and 2N-hydrochloric acid (5 drops) was completed in 3 hr. The resultant N-*benzoyl*-β-(3,5-*dimethoxy*-4-*methylphenyl*)*alanine* (1.9 g.) separated from ethyl acetate in needles, m. p. 182° [Found: C, 66·2; H, 6·2; N, 4·3; OMe, 18·1. $C_{17}H_{15}NO_3(OMe)_2$ requires C, 66·5; H, 6·2; N, 4·1; OMe, 18·1%]. Hydrolysis of this *N*-benzoate (2 g.) in boiling acetic acid (50 ml.) containing 20% hydrochloric acid (100 ml.) was complete in 48 hr. The cooled hydrolysate was filtered and extracted with ether, and the aqueous hydrolysate evaporated to dryness *in vacuo*. Crystallisation of the residue from 1% hydrochloric acid gave the *hydrochloride* of β-(3,5-dimethoxy-4-methylphenyl)alanine (1·5 g.), m. p. 238° [Found: C, 52·5; H, 6·5; N, 4·9; OMe, 22·0. $C_{10}H_{12}ClNO_2(OMe)_2$ requires C, 52·4; H, 6·5; N, 5·1; OMe, 22·5%].

6,8-*Dihydroxy*-3-*methoxycarbonyl*-7-*methylisoquinoline*.—A solution of the previous hydrochloride (1·2 g.) in water (10 ml.) was neutralised with 0·6N-sodium hydroxide, followed by the addition of an excess (20 ml.) of reagent and 40% formalin (4 ml.). The mixture was kept at 30° in an atmosphere of nitrogen for 14 days. The solid obtained by removal of the solvent *in vacuo* was dissolved in methanol (100 ml.), and the solution was saturated with hydrogen chloride and refluxed for 6 hr. Next day the reaction mixture was diluted with water, basified by the addition of excess of sodium carbonate, and extracted with ether. Evaporation of the ether furnished 1,2,3,4-tetrahydro-6,8-dimethoxy-3-methoxycarbonyl-7-methylisoquinoline as an oil (0·9 g.), which was characterised as the *picrate*, yellow needles, m. p. 208° (decomp.), from ethanol [Found: C, 49·0; H, 4·4; N, 11·3; OMe, 18·9. $C_{17}H_{13}N_4O_8(OMe)_2$ requires C, 48·6; H, 4·5; N, 11·3; OMe, 18·8%].

Regenerated from the picrate with ammonia solution, this tetrahydroisoquinoline (0·5 g.) in *p*-cymene (50 ml.) containing 30% palladium–charcoal (1 g.) was refluxed in a stream of carbon dioxide during 6 hr. The cooled mixture was diluted with benzene and filtered, and the clarified solution extracted with excess of 2N-hydrochloric acid. Basification of this acidic extract followed by isolation of the precipitate with chloroform and purification by way of the picrate furnished 3-*methoxycarbonyl*-6,8-*dimethoxy*-7-*methylisoquinoline* (0·15 g.), which separated from benzene–light petroleum (b. p. 60—80°) in cubes, m. p. 154°, and from methanol in prisms, m. p. 168° (Found: C, 65·0; H, 6·1: N, 5·5. $C_{14}H_{15}NO_4$ requires C, 64·5; H, 5·8; N, 5·4%).

Dechloroaposclerotaminic Acid.—A solution of di-*O*-acetylaposclerotaminic acid (2 g.) in hydriodic acid (S.G. 1·7) (40 ml.) was refluxed for 20 hr., and then evaporated to dryness *in vacuo*. After washing with water (50 ml.), purification of the product from 2N-sulphuric acid gave dechloroaposclerotaminic acid in yellow prisms (1·2 g.), m. p. 270° (decomp.).

A suspension of this acid (1 g.) in methanol (15 ml.) was saturated with hydrogen chloride and maintained at the boiling point for 5 hr. Removal of the solvent under reduced pressure followed by treatment of the residue with excess of sodium hydrogen carbonate solution gave *methyl dechloroaposclerotaminate* in yellow needles (1·1 g.), m. p. 270° (decomp.), from methanol (Found: C, 61·3; H, 4·6; N, 5·8; Cl, 0. $C_{12}H_{11}NO_4$ requires C, 61·8; H, 4·8; N, 6·0%). The *di*-O-*benzoate* separated from ethanol in prisms, m. p. 238° [Found: C, 70·7; H, 4·4; N, 3·4; OMe, 6·7. $C_{25}H_{16}NO_5(OMe)$ requires C, 70·7; H, 4·3; N, 3·2; OMe, 7·0%].

Sulphuryl chloride (110 mg.) was added to a solution of methyl dechloroaposclerotaminate (200 mg.) in acetic acid (300 ml.). After 24 hr. the solvent was removed *in vacuo* and a solution of the oily residue in methanol (15 ml.) was saturated with hydrogen chloride and then refluxed during 15 min. Methyl aposclerotaminate (100 mg.), m. p. and mixed m. p. 240° (decomp.), having the requisite infrared spectrum, separated from the cooled mixture. This ester was further characterised by conversion into methyl di-*O*-acetylaposclerotaminate, m. p. and mixed m. p. 184°, having the requisite infrared spectrum.

Prepared quantitatively by the acetic anhydride–pyridine method, *methyl di*-O-*acetyldechloroaposclerotaminate* formed prisms, m. p. 176°, from methanol [Found: C, 60·5; H, 5·1; N, 4·5; OMe, 9·8. $C_{15}H_{12}NO_5(OMe)$ requires C, 60·6; H, 4·8; N, 4·4; OMe, 9·8%].

Methylation of methyl dechloroaposclerotaminate (0·6 g.) in boiling acetone (30 ml.) containing methyl iodide (4 ml.) and potassium carbonate (5 g.) during 1½ hr. gave methyl di-*O*-methyldechloroaposclerotaminate (0·12 g.) in stout prisms, m. p. 168°, from methanol [Found: C, 64·7; H, 5·3; OMe, 33·0. Calc. for $C_{11}H_6NO(OMe)_3$: C, 64·4; H, 5·8; N, 5·4; OMe, 35·6%]. This ester was identical with the synthetic specimen of 3-carboxymethoxy-6,8-dimethoxy-7-methylisoquinoline and had the requisite infrared, ultraviolet, and n.m.r. spectra.

The *picrate* formed yellow plates from acetic acid, m. p. 238° (decomp.) (Found: N, 11·3. $C_{14}H_{15}O_4N{\cdot}C_6H_3O_7N_3$ requires N, 11·4%).

Demethylation of methyl dechlorodi-*O*-methylaposclerotaminate (60 mg.) with boiling hydriodic acid (5 ml.) and acetic acid (0·5 ml.) during 3 hr. gave dechloroaposclerotaminic acid (50 mg.), which was characterised by conversion into methyl di-*O*-acetyldechloroaposclerotaminate, m. p. and mixed m. p. 176°, and having the requisite infrared spectrum.

Methylation of methyl dechloroaposclerotaminate by the methyl iodide–potassium carbonate method in boiling acetone during 5 hr. gave the *methiodide* of methyl dechlorodi-*O*-methylaposclerotaminate as the principal product, yellow needles, m. p. 198° (decomp.), from methanol [Found: C, 44·1; H, 4·8; OMe, 21·3; I, 31·2. $C_{12}H_9NOI(OMe)_3$ requires C, 44·6; H, 4·5; OMe, 23·1; I, 31·5%]: the compound contained ionic iodine.

The same methiodide (m. p., mixed m. p., and infrared spectrum) was obtained in high yield

when methyl dechlorodi-*O*-methylaposclerotaminate was refluxed during 4 hr. in methanol containing methyl iodide.

N-*Acetyl*-β-(3,5-(*dimethoxyphenyl*)*alanine*.—Prepared by the hydrolysis of 2-acetyl-4-(3,5-dimethoxybenzylidene)-5-oxazolone (4 g.) in acetone (38 ml.) with boiling water (15 ml.), *α-acetamido-3,5-dimethoxycinnamic acid* formed needles (3·2 g.), m. p. 198°, from hot water [Found: 58·6; H, 5·7; N, 5·2; OMe, 23·1. $C_{11}H_9NO_3(OMe)_2$ requires C, 58·9; H, 5·7; N, 5·3; OMe, 23·4%].

Hydrogenation of this acid (1 g.) in acetic acid (100 ml.) containing 10% palladium–charcoal (0·3 g.) at 90 lb./sq. in., at 100° was complete in 5 hr. After removal of the acetic acid N-*acetyl*-β-(3,5-*dimethoxyphenyl*)*alanine* separated from hot water in plates (0·9 g.), m. p. 157° [Found: C, 58·1; H, 6·6; N, 5·2; OMe, 23·0. $C_{11}H_{11}NO_3(OMe)_2$ requires C, 58·4; H, 6·4; N, 5·2; OMe, 23·2%].

β-(3,5-*Dimethoxyphenyl*)*alanine*.—Prepared from 3,5-dimethoxybenzaldehyde (1·5 g.), sodium acetate (1 g.), hippuric acid (1·6 g.), and acetic anhydride (10 ml.) on the steam-bath during 2 hr., 4-(3,5-*dimethoxybenzylidene*)-2-*phenyl*-5-*oxazolone* separated from acetone in yellow needles (2·4 g.), m. p. 151° [Found: C, 69·8; H, 5·0; N, 4·6; OMe, 19·6. $C_{16}H_9NO_2(OMe)_2$ requires C, 69·9; H, 4·9; N, 4·5; OMe, 20·1%].

Hydrolysis of this oxazolone (1 g.) in acetone (50 ml.) with boiling 1% sodium hydroxide (50 ml.) during 1½ hr., followed by isolation in the usual manner gave *α-benzamido-3,5-dimethoxycinnamic acid* (0·9 g.) in needles, m. p. 206°, from aqueous alcohol [Found: C, 66·0; H, 5·2; N, 4·4; OMe, 18·7. $C_{16}H_{11}NO_3(OMe)_2$ requires C, 66·1; H, 5·2; N, 4·3; OMe, 19·0%].

Hydrogenation of this cinnamic acid (0·5 g.) in acetic acid (50 ml.) containing 10% palladium–charcoal (0·15 g.) occurred during 4 hr., to yield N-*benzoyl*-β-(3,5-*dimethoxyphenyl*)*alanine* in needles (0·4 g.), m. p. 175°, from aqueous alcohol [Found: C, 65·3; H, 6·0; N, 4·3; OMe, 18·9. $C_{16}H_{13}NO_3(OMe)_2$ requires C, 65·6; H, 5·8; N, 4·3; OMe, 18·9%].

Hydrolysis of this alanine (1 g.) in boiling 20% hydrochloric acid (50 ml.) occurred during 20 hr., in an atmosphere of nitrogen. The cold hydrolysate was filtered and extracted with ether, and the aqueous phase evaporated to dryness *in vacuo*. Purification of the crystalline residue from 2% aqueous hydrochloric acid gave β-(3,5-*dimethoxyphenyl*)*alanine hydrochloride* (0·5 g.) in needles, m. p. 230° [Found: C, 50·5; H, 5·9; N, 5·3; OMe, 24·1. $C_9H_9NO_2(OMe)_2$; HCl requires C, 50·5; H, 6·2; H, 5·4; OMe, 23·8%].

Unless otherwise stated infrared spectra were determined in a Nujol mull using a Perkin-Elmer 521 spectrophotometer: n.m.r. spectra were determined in deuteriochloroform solution with a Varian A-60 spectrometer with tetramethylsilane as an internal standard.

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