The Biosynthesis of Fungal Metabolites. Part III.[1] Structure of Shamixanthone and Tajixanthone, Metabolites of *Aspergillus variecolor*

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The structures of shamixanthone and tajixanthone, optically active metabolites of *Aspergillus variecolor*, are shown to be (1*R*,2*S*)-1,11-dihydroxy-2-isopropenyl-5-methyl-8-(3-methylbut-2-enyl)- (IV) and (1*R*,2*S*)-8-[(2*S*)-2,3-epoxy-3-methylbutyl]-1,11-dihydroxy-2-isopropenyl-5-methyl-2,3-dihydro-1*H*-pyrano[3,2-*a*]xanthen-12-one (V) respectively. These structures were established by detailed analyses of the ^{1}H and ^{13}C n.m.r. spectra of the metabolites and their derived compounds as well as by chemical degradation of the substituted dihydrobenzopyran system. The absolute configurations were assigned by application of the Horeau asymmetric synthesis.

THE isolation of six optically active xanthones from cultures of *Aspergillus variecolor* (Curzi) (*A. stellatus*) has been reported [2] and structures (I), (II), and (III) have been proposed for three of these; shamixanthone ($C_{25}H_{26}O_5$), tajixanthone ($C_{25}H_{26}O_6$), and ajamxanthone ($C_{25}H_{24}O_4$) respectively.[3] In view of the apparent novelty of these structures we have carried out a re-examination of the same strain of *A. variecolor* † and have isolated a number of metabolites, two of which are clearly identical with shamixanthone and tajixanthone. We now report investigations leading to new structures (IV) and (V) respectively for these metabolites.

† We are grateful to Dr. A. Kamal for supplying this strain of *A. variecolor*.

[1] Part II, J. R. Hadfield, J. S. E. Holker, and D. N. Stanway, *J. Chem. Soc.* (*C*), 1967, 751.

The presence of a *peri*-hydroxyxanthone chromophore in these compounds is indicated by the identical u.v. spectra, $\lambda_{max.}$ 392, 292, 275, 270sh, 256, and 242 nm (ε 5100, 7400, 27,400, 25,400, 18,800, and 20,800), i.r. bands at 3240—3100 and 1642 cm^{-1}, and a singlet at τ −2·58 (1H, exchangeable with D_2O), in their ^{1}H n.m.r. spectra. Furthermore, both compounds give typical iron(III) chelate colourations. This structural conclusion represents the limit of our agreement with previous workers.[3]

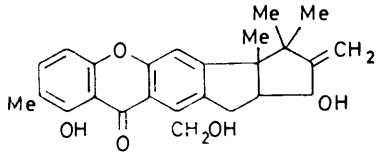
The presence of a secondary alcoholic function in both

[2] A. Kamal, S. A. Husain, R. Noorani, M. Murtaza, J. H. Qureshi, and A. A. Qureshi, *Pakistan J. Sci. Ind. Res.*, 1970, **13**, 251.

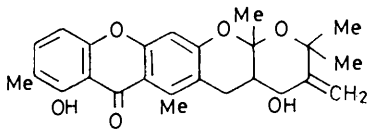
[3] A. Kamal, S. A. Husain, and A. A. Qureshi, *Pakistan J. Sci. Ind. Res.*, 1971, 90 and 104.

metabolites is established by the ^1H n.m.r. spectra which show a hydroxylic proton, exchanged with D_2O, at τ 4·98 (d, J 3·9 Hz) with the corresponding C*H*OH signal at τ 4·61 (q, J 3·9 and 2·8 Hz, collapsing to d, J 2·8 Hz after D_2O exchange). The O–H stretching at 3515 cm^{-1} in the i.r. spectrum of shamixanthone is independent of concentration and this high frequency suggests that the group is not hydrogen bonded. It is unusual to find a secondary alcohol which does not form an intermolecular hydrogen bond in concentrated solution and indicates that the hydroxy-group must be in a sterically crowded environment. Methylation (VIII) which had typical ^1H n.m.r. signals at τ 7·93 (Me·CO) and 3·02 (methine proton shifted 1·55 p.p.m. to low field compared with parent alcohol).

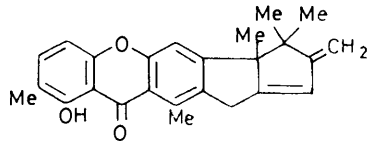
Two isolated olefinic double bonds were shown to be present in shamixanthone by oxidation with one mol. equiv. of monoperphthalic acid to give a monoepoxide, and with excess of reagent, a diepoxide. The ^1H n.m.r. spectra and m.p.s of the monoepoxide and tajixanthone were identical, establishing the structural relationship between the two metabolites. This was confirmed by conversion of tajixanthone into shamixanthone by reaction with triphenylphosphine selenide



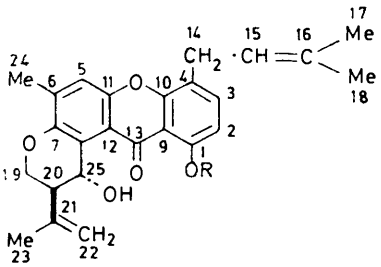
(I)



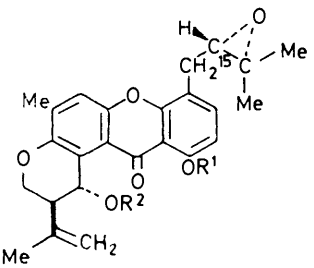
(II)



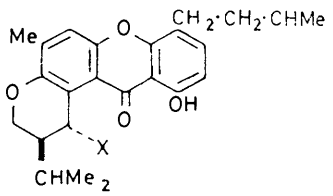
(III)



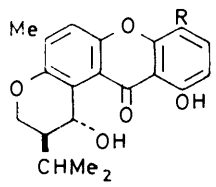
(IV) R = H
(VI) R = Me



(V) $R^1 = R^2 = H$
(VII) R^1 = Me, R^2 = H
(VIII) R^1 = Me, R^2 = Ac



(X) X = OH
(XI) X = H



(IX) R = $CH_2 \cdot CH{=}CMe_2$
(XII) R = $CH_2 \cdot CH(O){-}CMe_2$ (*S*-configuration)
(XIII) R = $CH_2 \cdot CH(OH) \cdot CHMe_2$ (*S*-configuration)
(XIV) R = $CH_2 \cdot CO \cdot CHMe_2$
(XV) R = $CH_2 \cdot CH(OH) \cdot C(OMe)Me_2$ (*S*-configuration)
(XVI) R = $CH_2 \cdot CO \cdot C(OMe)Me_2$

of shamixanthone and tajixanthone with methyl iodide and potassium carbonate in acetone gave the monomethyl ethers (VI) and (VII) respectively, and the secondary alcoholic O–H stretch was now at 3470 cm^{-1} in the i.r. spectrum of (VI). Since this is at lower frequency than in the parent (IV) and is unchanged on dilution, the hydroxy-group must now be intramolecularly hydrogen bonded. The change from a non-bonded hydroxy-group in the parent metabolites to an intramolecularly bonded group in the methyl ethers suggests that the OH group is relatively close to the xanthone carbonyl and becomes hydrogen bonded when the original strong hydrogen bond of the *peri*-phenolic hydroxy-group is removed by methylation. Acetylation of tajixanthone methyl ether gave the acetate and trifluoracetic acid, a reaction which effects the conversion of an epoxide into an alkene.[4]

Hydrogenation of shamixanthone with chlorotristriphenylphosphinerhodium(I) catalyst in benzene[5] gave the dihydro-derivative (IX), with palladium–carbon in ethyl acetate, the tetrahydro-derivative (X), and with platinum in ethyl acetate, a mixture of the tetrahydro-derivative (X) and the tetrahydrodeoxy-derivative (XI). The formation of the latter compound indicates that the secondary alcoholic hydroxy-group is benzylic and this is confirmed by the position of the signal of the methine proton (τ 4·61) in the ^1H n.m.r. spectrum of shamixanthone. Hydrogenation of tajixanthone with a platinum catalyst in ethyl acetate gave a mixture of the dihydro-derivative (XII) and the

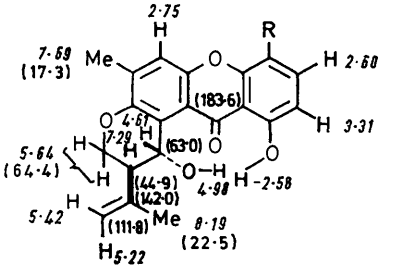
[4] D. L. J. Clive and C. V. Denyer, *J.C.S. Chem. Comm.*, 1973, 253.

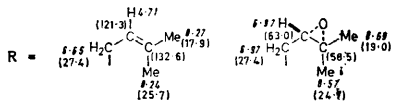
[5] J. A. Osborn, F. H. Jardine, J. F. Young, and G. Wilkinson, *J. Chem. Soc.* (*A*), 1966, 1711.

tetrahydro-derivative (XIII) in which the epoxide ring had been cleaved to a secondary alcohol, shown by oxidation with chromic oxide to the ketone (XIV). It is noteworthy that neither in this oxidation nor in similar attempted oxidations of shamixanthone (IV) and its tetrahydro-derivative (X) has it been possible to oxidise the benzyl alcohol group. These observations, and the hydrogen bonding studies discussed earlier, suggest that the benzyl alcohol group must be in a ring system and substituted *ortho* to the xanthone carbonyl group. Oxidation would then be difficult since the hypothetical derived ketones would have strong steric and electronic repulsions between the introduced and xanthone carbonyl groups. Hydrogenation of tajixanthone with palladium-carbon in methanol gave compound (XV), in which saturation of the isopropenyl double bond was accompanied by addition of the elements of methanol to the epoxide ring. Presumably a trace of acid in the catalyst facilitates this addition. Oxidation of compound (XV) with chromic oxide gave the methoxy-ketone (XVI).

TABLE 1

N.m.r. chemical shifts [^{13}C in parentheses (δ); ^{1}H in italics (τ)]. Solutions in $CDCl_3$





	Shamixanthone	Tajixanthone
Aromatic C–H	(135·8)	(136·4)
	(118·7)	(118·7)
	(109·3)	(109·7)
Aromatic C–O	(159·2)	(159·7)
	(152·1)	(152·3)
	(151·5)	(151·4)
	(148·8)	(149·0)
Aromatic C–C	(137·6)	(137·8)
	(120·5)	(120·6)
	(118·4)	(114·8)
	(116·4)	(116·4)
	(108·7)	(108·7)

The principal structural features in shamixanthone and tajixanthone were deduced initially from correlations of 100 MHz ^{1}H and proton-noise decoupled 25·2 MHz ^{13}C n.m.r. spectral data summarised in Table 1. The assignments of individual signals in the spectra were made from expected chemical shift values, comparisons between the ^{1}H and ^{13}C spectra of the metabolites and their derivatives, and off-resonance decoupling of the ^{13}C spectra.

The main conclusions from these studies are summarised as follows. (*a*) The spectra of shamixanthone (IV) and tajixanthone (V) differ only in the signals which must be assigned to an aromatic prenyl substituent in the former compound and the corresponding epoxide in the latter.

(*b*) The signals due to the isopropenyl residue in both metabolites are replaced in dihydrotajixanthone (XII) by the characteristic signals of an isopropyl group with non-equivalent methyls, *i.e.* τ 9·02 and 8·98 (J 6·5 Hz) in the ^{1}H spectrum and at δ 20·4 and 21·2 in the ^{13}C spectrum. The CH fragment of the isopropyl group occurs at τ 8·32 (^{1}H) and δ 25·2 (^{13}C) in the corresponding spectra.

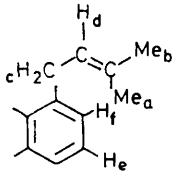
(*c*) The aromatic methyl substituent has the same chemical shifts in all the above derivatives, *i.e.* τ 7·68 (^{1}H) and δ 17·3 (^{13}C).

(*d*) The ^{13}C spectra show twelve aromatic carbon atoms of which three only are C–H carbons. In the ^{1}H spectra the corresponding three protons occur as an *ortho*-coupled pair and another showing a small coupling (J 0·9 Hz) which is discussed below. ^{13}C Chemical shifts show that four of the remaining nine aromatic carbon atoms carry oxygen substituents.

(*e*) The signal at lowest field in the ^{13}C spectra, *i.e.* δ 183·6 is assigned to the xanthone carbonyl group.

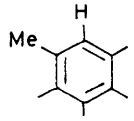
TABLE 2a

^{1}H Couplings in shamixanthone (IV)

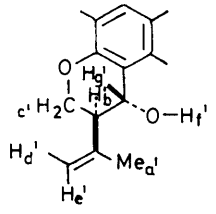


J_{cd} 7·7, J_{ef} 8·5, J_{ad} *ca.* 0·6 Hz

J_{bd} 1·2, J_{cf} *ca.* 0·5 Hz



J 0·9 Hz

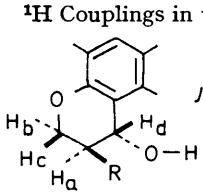


$J_{b'c'}$ 3·0, $J_{f'g'}$ 3·9, $J_{b'g'}$ 2·8, $J_{a'd'}$ *ca.* 0·8, $J_{a'e'}$ 1·4, $J_{b'e'}$ *ca.* 0·6, $J_{b'd'}$ 1·4, $J_{d'e'}$ 1·4 Hz

The many couplings in the ^{1}H spectrum of shamixanthone were elucidated by extensive decoupling experiments using a degassed solution in acid free $CDCl_3$. These couplings, which are summarised in Table 2a, were vital in the following structural deductions.

TABLE 2b

^{1}H Couplings in tetrahydroshamixanthone (X)



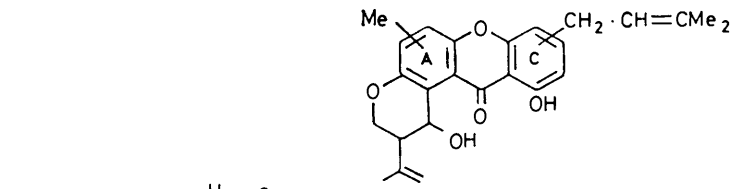
J_{bc} 11·0, J_{ab} 2·8, J_{ac} 3·2, J_{ad} 2·8 Hz

(*a*) In addition to the usual *vic*- and allylic couplings in the *C*-prenyl residue which serve to confirm this structural feature, the methylene protons of this residue show an allylic coupling (J 0·5 Hz) with the lower field *ortho*-coupled aromatic proton. Hence the prenyl group is located in a tetrasubstituted aromatic ring.

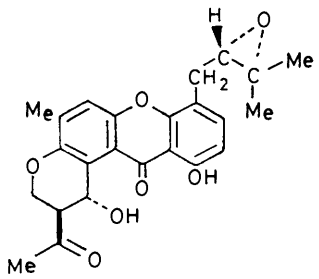
(*b*) The protons of the aromatic methyl substituent are coupled (J 0·9 Hz) with the aromatic proton, τ 2·75, referred to above. Since this proton shows no further coupling, the methyl substituent is located in a pentasubstituted aromatic ring.

(*c*) The fragment $-O{\cdot}CH_2CHRCH(CH)-$ is established by chemical shift considerations, hydroxylic proton exchange with D_2O discussed earlier, and *vic*-couplings. Although the methylene protons of this fragment have the same chemical shift at room temperature (τ 5·64), they begin to separate at low temperature, and in the hydrogenated derivatives (IX), (X), and (XII) they are non-equivalent, showing the typical AB portion of an ABX spectrum (τ 5·74 and 5·58 respectively in tetrahydroshamixanthone). This suggests that the fragment $-O{\cdot}CH_2CHRCH(OH)-$ is in a cyclic system as part of a substituted dihydrobenzopyran ring. Furthermore, since the *vic*-couplings of this system are all small (Table 2b) (J 2·8, 3·2, and 2·8 Hz respectively), they correspond to *gauche*-conformations of the individual protons, and the substituent R in this system must therefore have a *pseudo*-axial conformation.

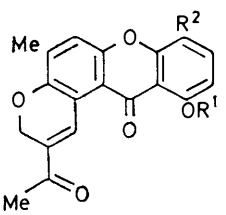
(*d*) In the spectrum of shamixanthone the isopropenyl group shows the usual *gem*-coupling of the $H_2C{=}C<$ protons (J 1·4 Hz) and allylic couplings between these and the methyl group (J *ca.* 0·8 and 1·4 Hz respectively). However, each of the $H_2C{=}C<$ protons is also coupled with the high field proton of the $-O{\cdot}CH_2CH(R)CH(OH)-$ fragment (J 1·4 and *ca.* 0·6 Hz respectively). Hence, the substituent R in this fragment is defined as the isopropenyl group in shamixanthone and as isopropyl in the tetrahydro-derivative (Table 2b).



(XVII)



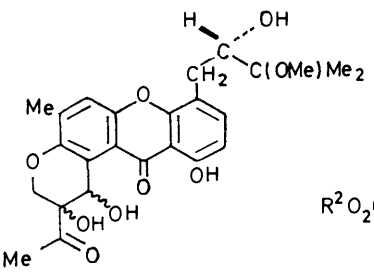
(XVIII)



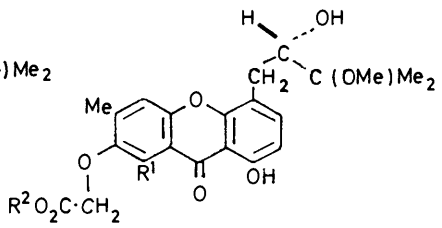
(XIX) $R^1 = H$, $R^2 = CH_2 \cdot CH(O)CMe_2$ (*S*-configuration)

(XX) $R^1 = Me$, $R^2 = CH_2 \cdot CH(O)CMe_2$ (*S*-configuration)

(XXI) $R^1 = Me$, $R^2 = CH_2 \cdot CH(OH) \cdot C(OMe)Me_2$ (*S*-configuration)



(XXII)



(XXIII) $R^1 = CHO$, $R^2 = H$

(XXIV) $R^1 = H$, $R^2 = Me$

At this stage the partial structure (XVII) can be advanced for shamixanthone, the only features to be decided, apart from stereochemical considerations, being the positions of the aromatic methyl and prenyl substituents in rings A and C respectively. The latter was established from the following considerations. (*a*) Tajixanthone and tetrahydroshamixanthone gave negative Gibb's tests under the modified conditions of King *et al.*[6] The 4-position of the 1-hydroxyxanthone system must therefore be substituted by the prenyl

[6] F. E. King, T. J. King, and L. C. Manning, *J. Chem. Soc.*, 1957, 563.

residue. It is interesting that shamixanthone slowly developed a positive reaction with the Gibb's reagent. Although the reason for this is not clear it does not invalidate the general conclusion.

(*b*) No loss of 56 mass units was seen in the mass spectrum of shamixanthone. *o*-Prenyl phenols characteristically show this loss.[7]

(*c*) The ^{1}H n.m.r. spectrum of *O*-methyltajixanthone (VII) in 75% C_6D_6–$CDCl_3$ showed an upfield shift of 0·36 p.p.m. for the methoxy-protons, compared with the spectrum in $CDCl_3$. This indicates that at least one of the positions *ortho* to the methoxy-group is unsubstituted.[8]

The position of the aromatic methyl substituent in ring A of the metabolites was established by the following reaction sequence, which also provides confirmatory evidence for the substituted dihydrobenzopyran ring. Oxidation of tajixanthone with osmium tetraoxide and sodium periodate gave the methyl ketone (XVIII) (τ 7·69 and $\nu_{max.}$ 1710 cm^{-1}). Since this compound is a β-hydroxyketone it was readily dehydrated with potassium hydroxide in methanol to the αβ-unsaturated ketone (XIX) ($\nu_{max.}$ 1660—1640 cm^{-1}) which had ^{1}H n.m.r. signals at τ 7·50 (Me·CO) and 1·09 (t, J 1·5 Hz, –CH=C–CO–, allylic coupling to ring methylene group). The very low field chemical shift of this vinyl proton is due to the deshielding effect of the xanthone carbonyl group. Reaction of compound (XIX) with osmium tetraoxide, decomposition of the resultant osmate ester with hydrogen sulphide, and subsequent addition of methanol, gave the *vic*-diol (XXII), in which the elements of methanol had added across the epoxide ring. Oxidation of this compound with periodic acid in ether [9] gave the formyl-acid (XXIII), formed by cleavage of the *vic*-diol followed by oxidative fission of the resultant α-diketone. The structure of this compound was established by its ^{1}H n.m.r. spectrum in $(CD_3)_2CO$ which showed signals at τ 5·37 (–O·CH_2·CO_2H) and —0·61 (Ar·C*H*O). The corresponding methyl ester, prepared with diazomethane, was decarbonylated with chlorotristriphenylphosphinerhodium(I) in benzene [10] to give the product (XXIV), in which the chemical shifts of the two ring A aromatic protons were readily differentiated since the signal at τ 2·58 showed the usual 0·9 Hz coupling with the aromatic methyl substituent and that at τ 2·76, which must be due to the newly introduced proton, showed no coupling greater than *ca.* 0·3 Hz which represented the spectral resolution obtained [spectrum in $(CD_3)_2CO$]. The absence of *m*-coupling between these two protons defines the position of the methyl substituent in ring A and provides the final piece of evidence which establishes structures (IV) and (V) for shamixanthone and tajixanthone respectively, apart from stereochemical assignments.

The mass spectra of the metabolites accord with the proposed structures and the principal fragmentation patterns of structural interest are summarised in Schemes 1 and 2. Since both shamixanthone and tajixanthone show relatively abundant ions due to losses of C_5H_8 and C_5H_9, these must occur principally from the substituted dihydrobenzopyran residues. It is probable that rearrangement of the parent ions occur leading to *O*-prenyl aldehydes, as illustrated for shamixanthone in Scheme 1. Subsequent losses would then be as expected for prenyl ethers, leading to ions *a* and *b* from shamixanthone and *a*′ and *b*′ from tajixanthone. Ions *a* and *a*′ further fragment by loss of CO, presumably from the aldehyde groups, giving ions *c* and *c*′ respectively. Other fragmentations of both parent ions are similar and include losses of H_2O, presumably from the secondary alcohol portions, giving ions *d* and *d*′ respectively. A series of fragmentations of particular interest in tajixanthone is represented by ions *h*′, *i*′, and *j*′ due to successive losses of CH_3·$\dot{C}O$, C_2H_4, and H_2O respectively from the parent. Ion *j*′ represents the base peak of this spectrum. This fragmentation pattern is characteristic of *C*-prenyl epoxides [11] and is thought to arise by initial pinacol type rearrangement to give ion *g*′, followed by fragmentations as shown in Scheme 2. In the case of shamixanthone the interesting loss of Me· from ions *c* and *d* to give ions *e* and *f* respectively is typical of an aromatic prenyl substituent with ether oxygen in the *o*-position.[11] The mass spectra of the methyl ethers (VI) and (VII) are characterised by losses of C_5H_9 giving base peaks at *m/e* 351 and 367 respectively.

The relative and absolute stereochemistry of shamixanthone and tajixanthone must now be considered. In shamixanthone (IV) there are two chiral centres, C(20) and C(25), and in tajixanthone the additional centre at C(15).* Furthermore, since tajixanthone has been converted into shamixanthone both compounds have the same absolute stereochemistry at C(20) and C(25). The absolute configuration at C(15) in tajixanthone was readily established by the method of Boar and Damps.[13] Thus, methylation of the αβ-unsaturated ketone (XIX) gave the methyl ether (XX) which was converted into the dimethoxy-alcohol (XXI) by acid-catalysed methanolysis. This compound was treated with (±)-α-phenylbutyric anhydride, as in the Horeau asymmetric synthesis,[14] to give an excess of (—)-α-phenylbutyric acid (optical yield 33%). Hence, tajixanthone has the (15*S*)-configuration, as shown in structure (V). The absolute configuration at C(25) was also established by the same method. Thus,

* The numbering of the non-aromatic carbon atoms in these metabolites accords with the system devised for the biogenetically related compound, arugosin C.[12]

[7] J. A. Ballantine, D. J. Francis, C. H. Hassall, and J. L. C. Wright, *J. Chem. Soc.* (*C*), 1970, 1175.
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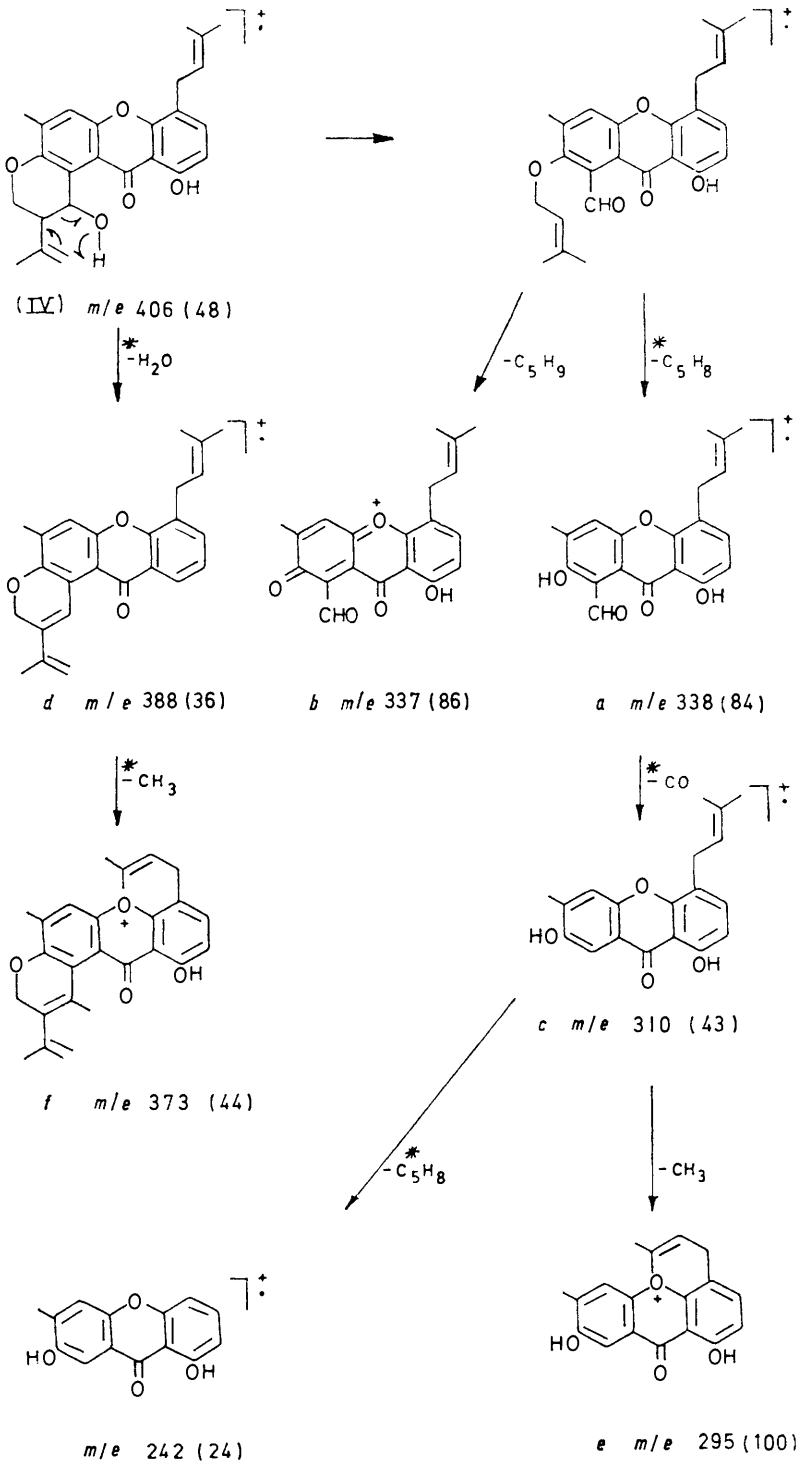
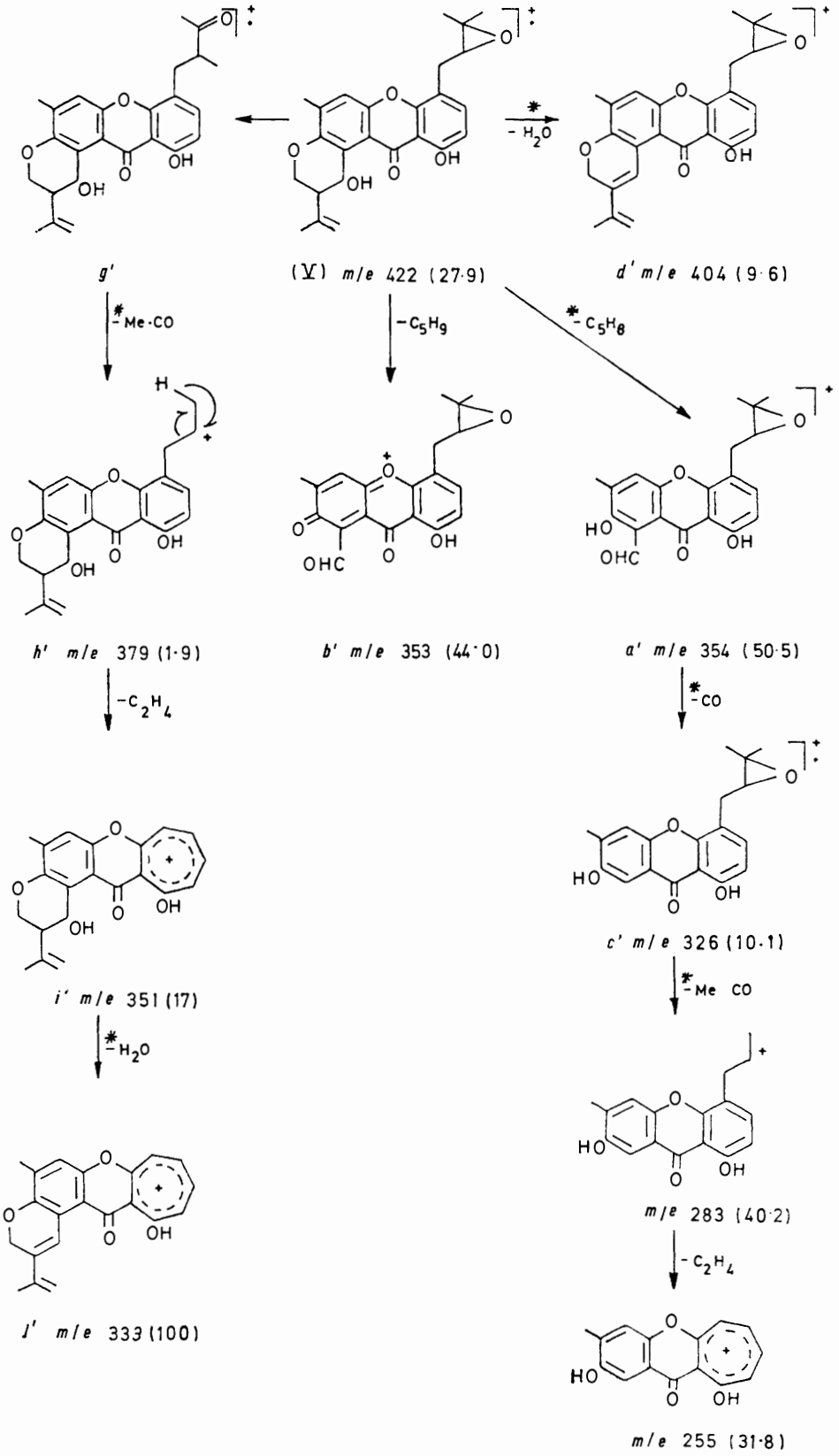


SCHEME 1 Principal mass spectral fragmentations of shamixanthone; relative abundance of ions in parentheses. Metastable ions are indicated



SCHEME 2 Principal mass spectral fragmentations of tajixanthone

treatment of *o*-methylshamixanthone with (±)-α-phenylbutyric anhydride gave an excess of (+)-α-phenylbutyric acid (optical yield 20%). On the assumption that the phenyl group is the largest substituent at C(25) in this chiral alcohol, then shamixanthone, and corresponding tajixanthone, have the (25*R*)-configuration as shown in structures (IV) and (V) respectively.

The relative configuration at C(20) in shamixanthone and tajixanthone is suggested by the preferred axial conformation of the isopropyl substituent in the hydrogenated derivatives, (IX), (X), and (XII). If this substituent and the adjacent hydroxy-group at C(25) were *cis*-related there would seem to be no valid reason for this conformational preference. On the other hand, a *trans*-relationship of these two groups would be better accommodated in a diaxial rather than a diequatorial conformation with its attendant *vic*-nonbonded interactions. Hence, it seems probable that the isopropenyl and hydroxy-substituents are *trans*-related in the dihydropyran rings of shamixanthone and tajixanthone as shown in the structures (IV) and (V) respectively.

The structures of minor metabolites of *A. variecolor* and the biogenesis of all these compounds are being investigated and will be reported subsequently.

EXPERIMENTAL

Unless otherwise stated, i.r. absorption spectra were measured with a Perkin-Elmer model 125 instrument for KBr discs, u.v. spectra with a Unicam SP 800 instrument for solutions in ethanol, ^{1}H n.m.r. spectra with a Varian HA-100 instrument for solutions in acid-free deuteriochloroform containing tetramethylsilane as internal standard, ^{13}C n.m.r. spectra with a Varian XL-100-15 FT spectrometer for similar solutions, and optical rotations with an ETL-NPL automatic polarimeter for solutions in chloroform. Mass spectra were measured at 70 eV with an A.E.I. MS12 spectrometer and accurate masses with an MS9 instrument. Thin-layer chromatography (t.l.c.) was performed using silica gel G.F. (Merck). M.p.s were determined with a Kofler hot-stage instrument. Spectral data marked with an asterisk are listed in Supplementary Publication No. SUP 20992 (12 pp., 1 microfiche).*

Isolation of Shamixanthone (IV) *and Tajixanthone* (V).—*Aspergillus variecolor*, I.M.I. strain 112543, was grown from a spore suspension in static culture for 15 days at 25° in flat vessels (*ca.* 1 l capacity), each containing Czapex–Dox medium (500 ml). The dried mycelium (*ca.* 7—8 g^{-1}) was ground and continuously extracted with light petroleum (b.p. 40—60°). The resulting dark oil was triturated with warm methanol and the methanol-soluble fraction evaporated to give a yellow solid which was fractionated by preparative t.l.c. using benzene–ether (95 : 5 v/v) as developing solvent. Shamixanthone (IV) was eluted with ethyl acetate from the band with R_F 0·60, and crystallised from methanol to give yellow needles (40—60 mg^{-1}), m.p. 154—156°, $[\alpha]_D^{24}$ +11·9° (*c* 1·92), $\nu_{max.}$* $\lambda_{max.}$* (Found: C, 73·8; H, 6·4. Calc. for $C_{25}H_{26}O_5$: C, 73·9; H, 6·5%). Tajixanthone (V) was similarly eluted from the band with R_F 0·35 and crystallised from methanol to give yellow needles (60—80 mg^{-1}), m.p. 158—159°, $[\alpha]_D^{24}$ −5·6° (*c* 2·44), $\nu_{max.}$* $\lambda_{max.}$* (Found: C, 71·1; H, 6·2. Calc. for $C_{25}H_{26}O_6$: C, 71·3; H, 6·6%).

Methylation of Shamixanthone and Tajixanthone.—On reaction with methyl iodide and anhydrous potassium carbonate in acetone, shamixanthone (60 mg) gave (1R,2S)-1-*hydroxy*-2-*isopropenyl*-11-*methoxy*-5-*methyl*-8-(3-*methylbut*-2-*enyl*)-2,3-*dihydro*-1H-*pyrano*[3,2-a]*xanthen*-12-*one* (VI), needles (45 mg) from acetone–light petroleum (b.p. 60—80°), m.p. 170—171°, $[\alpha]_D^{24}$ +22·3° (*c* 1·34), $\nu_{max.}$* $\lambda_{max.}$* τ* (Found: C, 74·2; H, 6·8. $C_{26}H_{28}O_5$ requires C, 74·3; H, 6·7%).

Under similar conditions, tajixanthone (100 mg) gave (1R,2S)-8-[(2S)-2,3-*epoxy*-3-*methylbutyl*]-1-*hydroxy*-2-*isopropenyl*-11-*methoxy*-5-*methyl*-2,3-*dihydro*-1H-*pyrano*[3,2-a]-*xanthen*-12-*one* (VII), needles (72 mg) from methanol–ether, m.p. 176—177°, $[\alpha]_D^{24}$ +18·7° (*c* 2·0), $\nu_{max.}$* $\lambda_{max.}$* τ,* δ_C* (Found: C, 71·3; H, 6·2. $C_{26}H_{28}O_6$ requires C, 71·6; H, 6·4%). Acetylation of O-methyltajixanthone (VII) (100 mg) with acetic anhydride–pyridine under the usual conditions gave the *monoacetate* (VIII) as a gum (86 mg), $\nu_{max.}$* *m/e*.*

Epoxidation of Shamixanthone.—Shamixanthone (50 mg) was treated with 1 mol. equiv. of monoperphthalic acid (23 mg) in ether (50 ml) at 0° for 4 days. The ether solution was then extracted with saturated sodium hydrogen carbonate solution (3 × 20 ml), washed with water (3 × 20 ml), and dried ($MgSO_4$). After removal of the solvent, the residue was recrystallised from methanol to give tajixanthone (42 mg), m.p. and mixed m.p. 158—159°, with identical i.r. and ^{1}H n.m.r. spectra.

Treatment of shamixanthone (50 mg) with an excess of monoperphthalic acid (60 mg) under similar conditions, gave the diepoxide (38 mg), yellow needles from methanol, m.p. 193—194°, $[\alpha]_D^{24}$ −46·0° (*c* 0·56), $\nu_{max.}$* $\lambda_{max.}$* τ.*

Conversion of Tajixanthone into Shamixanthone.—Tajixanthone (100 mg) and triphenylphosphine selenide (242 mg) in dichloromethane (30 ml) were stirred with trifluoroacetic acid (27 mg) for 12 h at room temperature. After filtration through Celite and removal of the solvent, the residue was purified by t.l.c. using ether–benzene (5 : 95 v/v) as developing solvent. The band, R_F 0·60, was removed and extracted with ethyl acetate to give shamixanthone, yellow needles (70 mg) from methanol, m.p. and mixed m.p. 154—156°, with identical i.r. and ^{1}H n.m.r. spectra.

Hydrogenation of Shamixanthone.—(*a*) Shamixanthone (148 mg) in dry benzene (20 ml) was stirred under hydrogen for 24 h with chlorotristriphenylphosphinerhodium(I) (160 mg) in dry benzene (10 ml). After removal of the solvent, the residue was purified by t.l.c. to give *dihydroshamixanthone* (IX), yellow needles from methanol (90 mg), m.p. 163—165°, $\nu_{max.}$* $\lambda_{max.}$* τ,* *m/e** (Found: M^+, 408·190. $C_{25}H_{28}O_5$ requires M, 408·194).

(*b*) Shamixanthone (150 mg) in ethyl acetate (50 ml) was hydrogenated at room temperature and atmospheric pressure for 3 days in the presence of Adams catalyst (100 mg) to give a mixture of two products which were separated by t.l.c., using ether–benzene (1 : 9 v/v) as eluting solvent. The band, R_F 0·56, gave *tetrahydroshamixanthone* (X), as yellow needles (80 mg) from methanol, m.p. 141—142°, $[\alpha]_D^{24}$ +11·6 (*c* 1·07), $\nu_{max.}$* $\lambda_{max.}$* τ* (Found: C, 73·1;

* For details of Supplementary Publications, see Notice to Authors No. 7 in *J.C.S. Perkin I*, 1972, Index issue.

H, 7·4. $C_{25}H_{30}O_5$ requires C, 73·3; H, 7·4%). The band, R_F 0·80, gave *tetrahydrodeoxyshamixanthone* (XI), yellow prisms (35 mg) from light petroleum (b.p. 40—60°), m.p. 78—82°, $[\alpha]_D^{24}$ −73·5° (*c* 1·2), $\nu_{max.}$,* $\lambda_{max.}$,* τ* (Found: C, 76·2; H, 7·6. $C_{25}H_{30}O_4$ requires C, 76·1; H, 7·7%).

(*c*) Shamixanthone (140 mg) in ethyl acetate (60 ml) was hydrogenated at room temperature and atmospheric pressure for 24 h in the presence of palladium–carbon (10%; 32 mg) to give tetrahydroshamixanthone (130 mg), yellow needles from methanol, m.p. and mixed m.p. 141—142°, with identical i.r. and ^{1}H n.m.r. spectra.

Hydrogenation of Tajixanthone.—(*a*) Tajixanthone (120 mg) in ethyl acetate (100 ml) was hydrogenated at room temperature and atmospheric pressure for 24 h in the presence of Adams catalyst (100 mg) to give a mixture of two products which were separated by t.l.c., using ether–benzene (1 : 9 v/v) as eluting solvent. The band, R_F 0·10, gave *tetrahydrotajixanthone* (XIII) as yellow needles (32 mg) from acetone–light petroleum (b.p. 60—80°), m.p. 185—187°, $[\alpha]_D^{24}$ −50·9 (*c* 1·75), $\nu_{max.}$,* $\lambda_{max.}$,* τ,* *m/e** (Found: C, 69·7; H, 7·0%; M^+, 426·204. $C_{26}H_{30}O_6$ requires C, 70·4; H, 7·1%; *M*, 426·204). The band, R_F 0·24, gave *dihydrotajixanthone* (XII) as yellow hexagonal prisms (40 mg) from acetone–light petroleum (b.p. 60—80°), m.p. 179—180°, $[\alpha]_D^{24}$ −4·9° (*c* 2·6), $\nu_{max.}$,* $\lambda_{max.}$,* τ,* δ_C* (Found: C, 70·7; H, 6·6. $C_{25}H_{28}O_6$ requires C, 70·7; H, 6·6%).

Oxidation of tetrahydrotajixanthone (50 mg) with Jones reagent at 0° gave (1R,2S)-1,11-*dihydroxy*-2-*isopropyl*-5-*methyl*-8-(3-*methyl*-2-*oxobutyl*)-2,3-*dihydro*-1H-*pyrano*[3,2-a]*xanthen*-12-*one* (XIV), yellow needles (20 mg) from methanol, m.p. 163—164°, $[\alpha]_D$ +18·4° (*c* 1·29), $\nu_{max.}$,* $\lambda_{max.}$,* τ* (Found: C, 70·5; H, 6·8. $C_{25}H_{28}O_6$ requires C, 70·7; H, 6·6%).

(*b*) Tajixanthone (120 mg) in methanol (40 ml) was hydrogenated at room temperature and atmospheric pressure for 48 h in the presence of 5% palladium–carbon (20 mg) to give (1R,2S)-1,11-*dihydroxy*-8-[(2S)-2-*hydroxy*-3-*methoxy*-3-*methylbutyl*]-2-*isopropyl*-5-*methyl*-2,3-*dihydro*-1H-*pyrano*[3,2-a]*xanthen*-12-*one* (XV), as yellow needles (120 mg), m.p. 156—158°, $[\alpha]_D^{24}$ −86·0° (*c* 0·70), $\nu_{max.}$,* $\lambda_{max.}$,* τ,* *m/e** (Found: M^+, 456·216. $C_{26}H_{32}O_7$ requires *M*, 456·216).

Oxidation of this compound (100 mg) with Jones reagent at 0° gave (1R,2S)-1,11-*dihydroxy*-2-*isopropyl*-8-(3-*methoxy*-3-*methyl*-2-*oxobutyl*)-5-*methyl*-2,3-*dihydro*-1H-*pyrano*[3,2-a]*xanthen*-12-*one* (XVI) which was purified by t.l.c. and crystallised from light petroleum (b.p. 60—80°) to give yellow needles (60 mg), m.p. 156—157°, $[\alpha]_D^{24}$ +19·6° (*c* 3·0), $\nu_{max.}$,* $\lambda_{max.}$,* τ,* *m/e** (Found: M^+, 454·196. $C_{26}H_{30}O_7$ requires *M*, 454·196).

Osmium Tetraoxide–Periodate Cleavage of Tajixanthone.—Tajixanthone (100 mg) in 25% aqueous dioxan (50 ml) was treated with osmium tetraoxide (20 mg) in dioxan (5 ml) and sodium metaperiodate (150 mg). After stirring for 24 h the mixture was poured into water and the product isolated in ethyl acetate. (1R,2S)-2-*Acetyl*-8-[(2S)-2,3-*epoxy*-3-*methylbutyl*]-1,11-*dihydroxy*-5-*methyl*-2,3-*dihydro*-1H-*pyrano*[3,2-a]*xanthen*-12-*one* (XVIII) separated from methanol in yellow needles (90 mg), m.p. 177—179°, $\nu_{max.}$,* $\lambda_{max.}$,* τ* (Found: C, 67·8; H, 6·0. $C_{24}H_{24}O_7$ requires C, 67·9; H, 5·7%).

Dehydration of the Ketol (XVIII).—The ketol (100 mg) in methanol (50 ml) was heated under reflux for 10 min with 10% methanolic potassium hydroxide (50 ml) and then poured into water (200 ml). The mixture was extracted with ethyl acetate and the extract washed successively with water, 2% sulphuric acid, water, and then dried. Evaporation of the solvent gave 2-*acetyl*-8-[(2S)-2,3-*epoxy*-3-*methylbutyl*]-11-*hydroxy*-5-*methyl*-3H-*pyrano*[3,2-a]*xanthen*-12-*one* (XIX) (90 mg), yellow needles, m.p. 192—194° (from methanol), $\nu_{max.}$,* $\lambda_{max.}$,* τ,* δ_O* (Found: C, 70·5; H, 5·8. $C_{24}H_{22}O_6$ requires C, 70·9; H, 5·5%).

Prepared with methyl iodide and anhydrous potassium carbonate, the *methyl ether* (XX) formed pale yellow needles, m.p. 205—207° (from methanol), τ* (Found: C, 71·1; H, 5·7. $C_{25}H_{24}O_6$ requires C, 71·4; H, 5·7%).

Hydroxylation of the αβ-Unsaturated Ketone (XIX).—This compound (400 mg) in dry tetrahydrofuran (40 ml) was treated overnight with osmium tetraoxide (300 mg) in tetrahydrofuran (10 ml). After saturating the suspension with hydrogen sulphide the solid was removed and the clear solution evaporated. The residue gave 2-*acetyl*-1,2,11-*trihydroxy*-8-[(2S)-2-*hydroxy*-3-*methoxy*-3-*methylbutyl*]-5-*methyl*-2,3-*dihydro*-1H-*pyrano*[3,2-a]*xanthen*-2-*one* (XXII), yellow rods (300 mg), m.p. 195—198°, from methanol, $\nu_{max.}$,* $\lambda_{max.}$,* τ,* *m/e** (Found: C, 63·0; H, 6·0%; M^+, 472·171. $C_{25}H_{28}O_9$ requires C, 63·5; H, 6·0%; *M*, 472·173).

Periodate Cleavage of the Diol (XXII).—50% Periodic acid in water (0·7 ml) was added to a vigorously stirred solution of the diol (139 mg) in ether (25 ml). After 90 min the pale yellow ether layer was separated, washed with water, dried ($MgSO_4$), and evaporated to a yellow oil (120 mg) which was crystallised from acetone–light petroleum (b.p. 60—80°) to give the 2-*carboxymethoxy*-1-*formyl*-8-*hydroxy*-5-[(2S)-2-*hydroxy*-3-*methoxy*-3-*methylbutyl*]-3-*methylxanthone* (XXIII) as small yellow rods (85 mg), m.p. 112—115°, $\nu_{max.}$,* $\lambda_{max.}$,* τ,* *m/e** (Found: C, 61·5; H, 5·6%; M^+, 444·144. $C_{23}H_{24}O_9$ requires C, 62·2; H, 5·4%; *M*, 444·142).

The Decarbonylation Product (XXIV).—The formyl-acid (XXIII) (75 mg) was treated with diazomethane in ether and the resultant crude methyl ester in benzene (25 ml) heated under reflux for 4 h in an atmosphere of nitrogen with chlorotristriphenylphosphinerhodium(I) (100 mg). After removal of the solvent the crude oil was purified by preparative t.l.c., the band at R_F 0·40 (benzene–ether, 50 : 50) being eluted to give an oil (20 mg) which separated from methanol in pale yellow needles of 8-*hydroxy*-5-[(2S)-2-*hydroxy*-3-*methoxy*-3-*methylbutyl*]-2-*methoxycarbonylmethoxy*-3-*methylxanthone* (XXIV), m.p. 136—138°, $\nu_{max.}$,* $\lambda_{max.}$,* τ,* *m/e** (Found: M^+, 430·159. $C_{23}H_{26}O_8$ requires *M*, 430·159).

Methanolysis of the Methyl Ether (XX).—This compound (100 mg) in hot methanol (60 ml) was treated with 72% perchloric acid (2 ml) on a steam-bath for 5 min. The solution was then poured into water (200 ml), and the product isolated in ethyl acetate and purified by t.l.c. The band, R_F 0·3 (benzene–acetone 3 : 1), separated from methanol in pale yellow needles (85 mg) of 2-*acetyl*-8-[(2S)-2-*hydroxy*-3-*methoxy*-3-*methylbutyl*]-11-*methoxy*-5-*methyl*-3H-*pyrano*[3,2-a]*xanthen*-12-*one* (XXI), m.p. 229—231°, τ* (Found: C, 68·5; H, 6·2. $C_{26}H_{28}O_7$ requires C, 69·0; H, 6·2%).

Determinations of Absolute Stereochemistry.—(*a*) The methanolate (XXI) (90 mg, 0·2 mmol) was treated with (±)-α-phenylbutyric anhydride (187 mg, 0·6 mmol) in pyridine (3 ml) for 3 days at room temperature. Water (20 ml) was added and after warming to 100° for 20 min

the resultant α-phenylbutyric acid (99 mg) was isolated (in ethyl acetate), $[\alpha]_D$ −6·4° (*c* 0·98), optical yield, 33%.

(*b*) *O*-Methylshamixanthone (VI) (82 mg, 0·2 mmol) was treated with (±)-α-phenylbutyric anhydride (187 mg, 0·6 mmol) as above for 10 days at room temperature to give α-phenylbutyric acid (115 mg), $[\alpha]_D$ +3·8° (*c* 0·90), optical yield 20%.

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