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XANTHONES OF THE GENTIANACEAE—II

FRASERA ALBICAULIS DOUGL. *EX* GRIESB.[1,2]

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Abstract—The root of *Frasera albicaulis* Dougl. *ex* Griesb. has been shown to contain fifteen xanthones, ten of which are previously unreported in nature. These compounds have been identified spectroscopically and in most cases by comparison with synthetic samples. They are shown to be based on the 1,3,5- and 1,3,7-trioxyxanthone systems with added oxygen atoms at 2 and/or 4, and represent a number of methylation patterns including 1,3-dihydroxy, 1-hydroxy, 2-hydroxy, and totally methylated materials.

Frasera albicaulis Dougl. *ex* Griesb., native to the mountains of the western US, is one of the smaller members of this genus, and differs considerably in its form from the tall-flowering species such as *F. caroliniensis* Walt. and *F. speciosa* Dougl. *ex* Griesb. Preliminary examination of the root extract showed the presence of a much more complicated group of yellow pigments than had been observed in the larger species.[2] The assumption on chemotaxonomic grounds that these were xanthones was supported by their chemical and spectroscopic properties.

Because of the number of compounds involved and because they tend to conform to consistent patterns, the analysis of their structures will be simplified by first considering some general features of the NMR spectra of such xanthones. As will become apparent, two general interpretational problems occurred repeatedly. The first was the identification of the pattern of signals resulting from a single oxygen substituent (in these cases a OMe group) on one of the aromatic rings of the xanthone system. The second was the reverse problem of locating a single proton on a ring bearing three O atoms.

Of the singly oxygenated rings, only those containing a substituent at 1,2, and 4 have been found in nature. In the 1-oxy case the low-field signal of the coupled group arises from H-3 and usually appears as a simple triplet with a splitting of 8–10 Hz as would be predicted from first-order considerations. The upfield portion may consist of a simple pair of doublets or may show further small splittings. The spread of the aromatic spectral region covering these signals is large, ca. 1 ppm.[2,3]

In the 2-oxy and 4-oxy cases with which we shall be concerned, the low-field signal arises from H-1, which is deshielded by the anisotropic field of the adjacent carbonyl. In 2-OMe compounds, this appears generally at τ (2·30) 2·35–2·45 as a narrow symmetric quartet with splittings of 1–3 Hz, comparable to those predicted on a first order basis for *meta* and *para* couplings. The gap between these and the major high-field signals is small, and the entire span of the two groups of strong signals is $\leqslant$0·45 ppm.[2] The 4-oxy compounds show at τ 2·12–2·25 (2·30) a much wider quartet with splittings of about 4 and 6 Hz, obviously the result of the non-ideal behavior of this ABC system. The span of the major high- and low-field regions is increased to $\geqslant$0·63 ppm.[2,4] In both cases small additional peaks associated with the ABC system appear, but they do not interfere with the interpretation of the major peaks.

Examination of models containing an oxygen at C-3 indicates that the low field peak somewhat resembles that of the 4-oxy compounds, but that the spread over the high- and low-field regions is larger, ca. 1·0 ppm.

With regard to trisubstituted rings the important case is that of the proton at C-2 vs. that at C-4. Examination of compounds of known structure in both the xanthone[2,5] and flavone[6] fields indicates that for a given set of OH and OMe substituents H-2 always appears at somewhat higher field (0·1–0·3 ppm) than does H-4. The exact position depends on the nature of the substituents on both rings, but in general H-2 is found at τ 3·5–3·7 and H-4 at τ 3·2–3·5. The identification of a single compound is thus generally possible, while the assignment of two isomers differing in this way is quite secure.

Chromatography of a large sample of *F. albicaulis* extract, from which no crystalline material could be obtained directly, yielded nine resolved fractions (Table 3), each of which showed only a single spot on TLC. From these were isolated by further fractionation fifteen xanthones, labeled A through O in approximate sequence of their increasing polarity on silica gel. These are considered below in the order of their appearance and are listed with their NMR spectra in Table 1.

TABLE 1. NMR SPECTRA OF *F. albicaulis* XANTHONES

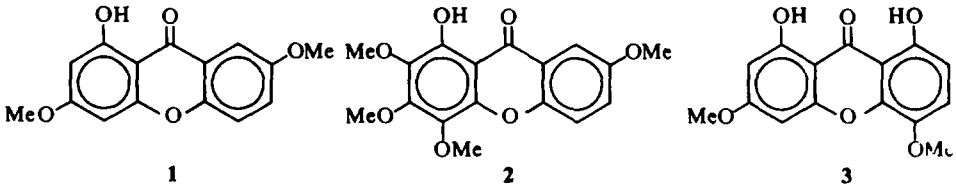
7-oxy	1	2	3	4	5,6	7	8	OMe	chel. OH	Note
A (1)	OH	3·70	OMe	3·58	2·68	OMe	2·41	6·08, 6·11	−2·94	*a*
B (2)	OH	OMe	OMe	OMe	2·63	OMe	2·43	5·91, 6·08, 6·12 (2)	−2·55	*a*
D (5)	OH	3·82	OH	OMe	2·68	OMe	2·45	6·25 (2)	−2·36	*b*
MeF (8)	OMe	OMe	OMe	3·30	2·75	OMe	2·38	6·02, 6·05, 6·11, 6·12		*a*
MeH, O (4)	OMe	3·56	OMe	OMe	2·68	OMe	2·38	6·00, 6·02, 6·10 (2)		*a*
J (16)	OMe	OH	OMe	OMe	2·67	OMe	2·34	5·88, 5·99, 6·03, 6·10		*a*
K (17)	OMe	OH	OMe	3·25	2·72	OMe	2·35	6·02 (2), 6·11		*a*
M (20)	OMe	3·68	OMe	3·52	2·78	OMe	2·30	6·08, 6·11 (2)		*a*
5-oxy	**1**	**2**	**3**	**4**	**5**	**6,7**	**8**			
C (3)	OH	3·75	OMe	3·58	OMe	2·82 3·39	OH	6·10, 6·13	−1·84, −1·23	*a*
MeE (6)	OMe	OMe	OMe	OMe	OMe	2·75	2·21	5·90, 5·96, 5·98, 6·07, 6·08		*a*
MeG, N (10)	OMe	3·57	OMe	OMe	OMe	2·80	2·25	5·98, 6·00, 6·04, 6·05		*a*
I (14)	OH	OMe	OMe	3·49	OMe	2·80	2·30	6·02, 6·07, 6·16	−2·62	*a*
L (19)	OMe	3·65	OMe	3·42	OMe	2·72	2·12	6·02, 6·11 (2)		*a*

[a] CH_2Cl_2 [b] DMSO

Fraction 1—*xanthone A.* Xanthone A, m.p. 169°, $C_{15}H_{12}O_5$, shows two OMe groups and a chelated OH in its NMR spectrum. Two doublets at τ 3·58 and 3·70 (J = 2·5 Hz) suggest from their position and coupling 1,3-dioxygenation, while the quartet at τ 2·41 and multiplet at τ 2·68 are those discussed above for 7(2)-oxygenation.

Direct comparison with the known methylgentisin,[7] 1-hydroxy-3,7-dimethoxyxanthone (**1**), showed the two compounds to be identical.

Fraction 2—xanthones B and C. Fraction 2 showed a broad melting range and was clearly a mixture. Rechromatography gave xanthone B, $C_{17}H_{16}O_7$, m.p. 119°, from the first fractions eluted. The NMR spectrum shows a chelated hydroxyl, four methoxyl groups, and the aromatic proton pattern indicative of a 7-OMe group. Direct comparison with 1-hydroxy-2,3,4,7-tetramethoxyxanthone, previously isolated from *F. carolinensis* Walt.,[2] confirmed the identification as **2**.

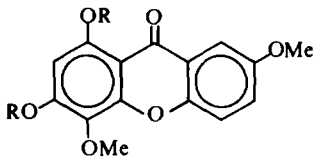


The later fractions from the column were mixtures of xanthones B and C, from which the latter could be isolated by selective extraction into aqueous hydroxide. Xanthones C, m.p. 189°, $C_{15}H_{12}O_1$, shows in its NMR two chelated OH groups and two OMe's The appearance of the aromatic protons as two pairs of doublets with *J*'s of 2·5 and 9 Hz suggested either 1,8-dihydroxy-3,5-dimethoxyxanthone (swerchirin[8]) or 1,8-dihydroxy-3,7-dimethoxyxanthone as the structure, and comparison with an authentic sample of swerchirin (**3**) from *Swertia chirata*[8] showed the former to be correct.

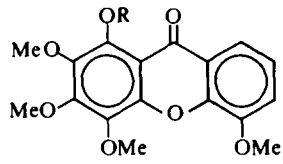
Fraction 3—xanthones D, E, F, G, H. This fraction also showed a broad melting range and a very complex NMR spectrum. One component, xanthone D, m.p. 246°, $C_{15}H_{12}O_6$, was easily separated, however, because of its insolubility in dichloromethane. Acetylation yielded a diacetate $C_{19}H_{16}O_8$, whose NMR spectrum showed two OMe as well as two acetoxyl peaks. The aromatic region shows the coupled set of three protons indicative of a 7-OMe substituent, as well as a singlet at τ 3·18. Total methylation of xanthone D yielded a product identified by NMR and direct comparison as 1,3,4,7-tetramethoxyxanthone (**4**), previously synthesized in connection with products H and O (see below).

One of the OH groups of xanthone D may be placed at position 1, since the NMR spectrum in DMSO shows a chelated hydroxyl signal at τ −2·36. The failure to give a positive Tollens test places one of the OMe groups at C-4, and the solubility of the compound in aqueous carbonate then locates the second OH group at C-3. Further evidence of this location is obtained from the shift in the UV spectrum of xanthone D in the presence of a trace of base (316, 375 mμ → 351, 400 mμ), which resembles closely that of 1,3-dihydroxy-7-methoxyxanthone (312, 370 mμ → 350, 390 mμ). In particular the band near 315 mμ shifts more in these 3-OH compounds than does that at the longer wavelength. Model 2-OH compounds, on the other hand (e.g. xanthone K: 323, 367 mμ → 340, 420 mμ), show a much larger shift for the long wavelength band. Thus xanthone D is 1,3-dihydroxy-4,7-dimethoxyxanthone (**5**).

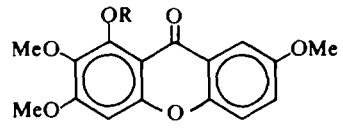
The mixture remaining after the removal of xanthone D could not be separated directly, but total methylation and chromatography on silica gel provided resolution into two fractions (i) and (ii). Each of these, however, still showed the properties of a mixture.



4: R = Me
5: R = H



6: R = Me
7: R = H



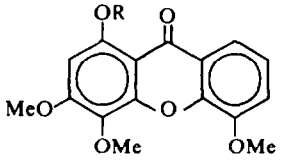
8: R = Me
9: R = H

Fraction (i) was ultimately resolved by preparative TLC on silica gel containing sulfuric acid[2] into two components, methyl xanthones E and F. Methyl xanthone E, $C_{18}H_{18}O_7$, m.p. 145°, shows in the NMR five OMe groups and the aromatic proton pattern of a 5 (or 4)-oxygenated ring. Direct comparison with 1,2,3,4,5-pentamethoxyxanthone[2] (**6**) proved the identity of the two molecules.

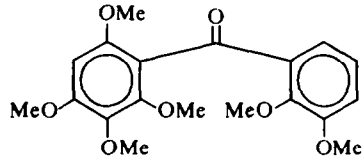
Methylxanthone F, m.p. 135°, $C_{17}H_{16}O_6$, shows four OMe groups, an aromatic singlet at τ 3·30, and the τ 2·38 quartet of a 7-oxygenated ring. The relatively low-field position of the singlet suggested that it arises from a proton at C-4, and direct comparison with 1,2,3,7-tetramethoxyxanthone[2] confirmed the structure as **8**.

The more polar fraction (ii) yielded on preparative TLC pure methylxanthone G, m.p. 177°, $C_{17}H_{16}O_6$. Four OMe signals are visible in the NMR spectrum, as well as aromatic signals very similar to those of **6** and a further singlet at τ 3·57. The high-field position of this singlet suggested H-2 as its source and indicated 1,3,4,5-trimethoxyxanthone (**10**) as the structure.

This new compound was synthesized by the condensation of 2,3-dimethoxybenzoic acid and 1,2,3,5-tetramethoxybenzene in the presence of trifluoroacetic anhydride (TFAA). The resulting benzophenone (**12**) was selectively demethylated to a mixture of products each containing a single OH group *ortho* to the carbonyl bridge. Ring closure with tetramethylammonium hydroxide in pyridine[2] gave a mixture of **10** and 1,2,3,5-tetramethoxyxanthone,[2] from which the former could be separated chromatographically. Comparison of the natural and synthetic materials assured their identity.



10: R = Me
11: R = H



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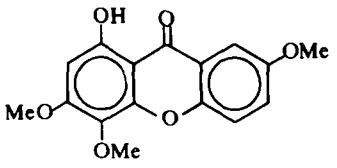
The final component of this mixture was present in relatively small amounts and could not be isolated in pure form. Inspection of the NMR spectrum of the mixture and consideration of the developing biosynthetic pattern suggested that it was probably 1,3,4,7-tetramethoxyxanthone (**4**). Subtraction of the peaks due to **10** from a NMR spectrum of fraction (ii) left a residual curve which was superimposable on that of an authentic specimen of **4** (xanthone O—see below).

The question remains as to the form in which the consistuents of this fraction actually occur in the plant. Selective demethylation of the natural or synthetic



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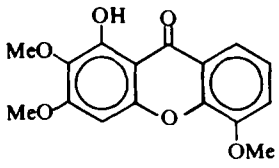

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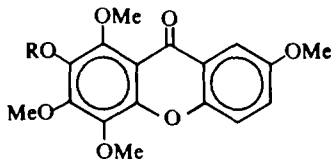


15: R = Me
16: R = H

compounds **6**, **8**, **10**, and **4** yielded the corresponding 1-OH products **7**,[2] **9**,[2] **11**, and **13**. All of these show the same mobility on TLC as the natural mixture, supporting —if not proving—the view that they are its constituents. In all cases the 1-OH compounds are more mobile than their methyl ethers, providing evidence that the natural compounds cannot contain 1-OMe systems. The possibility that they are 1,x-dihydroxy compounds cannot be entirely ruled out, but appears unlikely in view of the generally higher m.ps and lower solubilities of such substances, e.g. xanthone D. Thus it appears likely that the xanthones E, F, and G are in fact **7**, **9**, and **11**.

Although methyl xanthone H could arise by methylation of the xanthone D (**5**) known to be present in the original mixture, it was also isolated following methylation of a sample of fraction 3 which had been sublimed and exhaustively extracted with aqueous carbonate to remove **5**. Thus there is another, carbonate insoluble precursor, and xanthone H is presumably **13**.

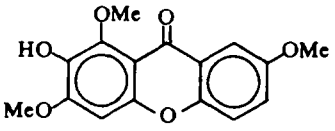
Fraction 4—*xanthone* I. Xanthone I, $C_{16}H_{14}O_6$, m.p. 188°, shows a chelated OH, three OMe peaks, and aromatic protons of the 5-OMe type, as well as an additional singlet at τ 3·49. The corresponding molecule, 1-hydroxy-2,3,5-trimethoxyxanthone (**14**), is known from studies on *Frasera caroliniensis*[2] and direct comparison proved the identity of the two samples.

Fraction 5—*xanthone J*. The NMR spectrum of xanthone J, m.p. 145°, $C_{17}H_{16}O_7$, reveals an unexpected feature. Only four OMe groups are present, but no signal appears in the τ −2 region despite the additional O atom, presumably present as a OH group. The aromatic protons indicated a substituent at C-7, and methylation yielded 1,2,3,4,7-pentamethoxyxanthone (**15**)[2] identical with that prepared by methylation of xanthone B.

Studies of the UV spectral shifts in base suggested that the OH group should be placed at C-2 or C-7, but to obtain conclusive proof single crystals of xanthone J were prepared and the structure studied by X-ray crystallographic methods.[9] These showed the compound to be 2-hydroxy-1,3,4,7-tetramethoxyxanthone (**16**), an unusual methylation pattern.

Fraction 6—*xanthone K*. Xanthone K, m.p. 192°, $C_{16}H_{14}O_6$, resembles the preceding compound in lacking a chelated OH signal in the NMR although it contains only three OMe groups. The aromatic protons appear as the familiar 7-oxy pattern, together with a singlet at τ 3·25 that suggested a proton at C-4. With the possibility of a OH group at C-3 excluded by the insolubility of xanthone K in sodium carbonate solutions, analogy with xanthone J suggests 2-hydroxy-1,3,7-trimethoxyxanthone (**17**) as the structure.

Synthetic **17** was prepared by the condensation of 2,6-dimethoxyhydroquinone and 2.5-dimethoxybenzoic acid in TFAA to give the dihydroxybenzophenone **18**.



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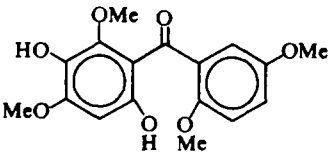

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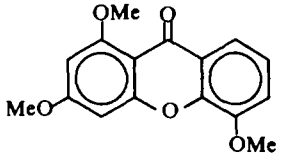
Basic ring closure led directly to **17** in 30% overall yield, and direct comparisons proved its identity with the natural material.

Fraction 7—xanthones L and M. This fraction was obtained only in very small amounts, and the crystalline material isolated from it had a broad melting range. The NMR showed a wide quartet at τ 2·12 and a smaller, narrow one at τ 2·30, indicating that a mixture of 5-oxy and 7-oxy compounds was present, the former predominating. In the high-field region there appear doublets at τ 3·42 and 3·65 (J = 2·5 Hz) and a weaker pair at τ 3·52 and 3·68 (J = 2·5 Hz). These resemble those shown by 1,3-dioxyxanthones (cf. xanthone A) and suggest the patterns of 1,3,5- and 1,3,7-trioxygenation. No chelated hydroxyl signal is visible in the spectrum, and the compounds are not retained by basic alumina, so the oxygenation is all present as alkoxyl groups. The OMe region is consistent with two groups of three OMe's and no other signals appear in the spectrum; thus the structures may be assigned as 1,3,5-trimethoxyxanthone (**19**) and 1,3,7-trimethoxyxanthone (**20**).

Comparison of a synthetic sample of **20** with fraction 7 showed that it has the same TLC behavior and that its NMR spectrum coincided with those portions of the mixed spectrum assigned to xanthone M.



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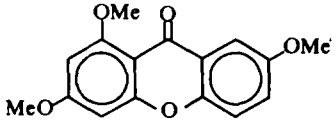

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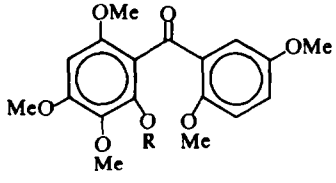


21: R = Me
22: R = H

Fraction 8—xanthone N. Xanthone N, m.p. 177°, $C_{17}H_{16}O_6$, is a neutral compound whose NMR spectrum is identical with that of methylxanthone G (**10**). The identity of the two was proved by direct comparison.

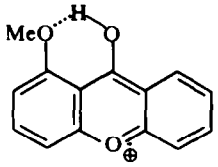
Fraction 9—xanthone O. Xanthone O, m.p. 188°, $C_{17}H_{16}O_6$, showed four OMe peaks in its NMR spectrum and four aromatic protons. Three of these correspond to the 7-OMe ring, and one, a singlet at τ 3·56, suggests a proton at C-2 and thus 1,3,4,7-tetramethoxyxanthone (**4**).

Condensation of 2,5-dimethoxybenzoic acid and 1,2,3,5-tetramethoxybenzene in TFAA yielded the benzophenone **21**. Selective demethylation afforded a 2-OH product that was assigned the structure **22** since this would arise by the expected cleavage of the most hindered OMe group.[2] In confirmation, cyclization in base gave **4**, clearly different from the previously synthesized alternative **8**, and identical with natural xanthone O.

During the liquid–liquid separation used in the isolation of the crude xanthone

mixture, a yellow crystalline residue separated. Hydrolysis of this in dilute acid gave a product that was predominantly swerchirin, but also contained smaller amounts of other xanthones, indicating that they also occur as glycosides.[2]

Chromatography of xanthones. The sequence of elution of these various products, as well as the separation of fraction 3(i) and 3(ii), points up the important effect of a 1-OMe group in determining the chromatographic polarity of these compounds on silica gel. It has long been known that 1-methoxyxanthones are more basic than their 1-OH counterparts,[10] but it has not been appreciated that as a result they are retained more strongly on this acid medium. The tendency of these colorless compounds to give yellow-orange, strongly fluorescent spots when adsorbed suggests that actual protonation occurs, leading to the stabilized ion **23**.



23

In general an increase in the number of OMe groups would be expected to increase the basicity of such a molecule and this effect is indeed observed. More important, however, is the effect of steric hindrance on the 1-OMe group. Thus the degree of retention is determined primarily by the substituent at C-2 and increases as this decreases in size (OMe < OH < H, e.g. **8** < **17** < **20**). Interference with the 1-OMe group causes it to rotate[9] and weakens the planar H—bond that contributes to the stability of 23. Because of the large effect of a proton *vs* a OMe group at C-2, it is quite easy to decide between these possibilities from the position of an unknown totally O-methylated compound on a TLC plate run against suitable standards. The same considerations would presumably also apply to other bulky C-2 substituents such as alkyl side chains.

Secondary effects arise from the position of additional OMe groups in the other ring and the addition of a OMe at C-4 to increase the crowding and mobility of a 1,2,3-trimethoxy compound. The importance of these is small (except for a 8-OMe group), but can be enhanced by the use of an acidified medium, e.g., the separation of **6** and **8**.

DISCUSSION

The xanthones identified in *Frasera albicaulis* include at least one O-methylated derivative of each of the hydroxylation patterns obtainable from the—at least conceptually—fundamental 1,3,5- and 1,3,7-trihydroxyxanthone systems by oxidation at C-2 and/or C-4. In this respect the species is more catholic than *F. caroliniensis*[2] and *Gentiana bellidifolia*,[5,11] which tend to favor oxygenation at one or the other of these positions.

The phloroglucinol ring in xanthones is generally considered to arise from acetate, and this has been confirmed for higher plants in one case.[12] Markham[5] has suggested that the isolation of only 1,3,4,5,8- and 1,3,4,7,8-pentaoxyxanthones from *G. bellidifolia*

TABLE 2. XANTHONES OF *F. albicaulis*

Subst.	1-OH	2-OH	Tot. Meth.	Other
1,3,7	A		M	
1,3,5			L	
1,2,3,7	F*	K		
1,2,3,5	I*			
1,3,4,7	H		O	D
1,3,4,5	G		N	*
1,2,3,4,7	B*	J		
1,2,3,4,5	E*			
1,3,5,8				C*

* Also found in *F. caroliniensis*.[2]

argues for oxidation following formation of the xanthone system since a 2,3,4,6-tetraoxybenzophenone would be expected to yield both 1,2,3- and 1,3,4-trioxyxanthones. unless the latter were energetically favored. This argument appears weak since it is entirely possible that an enzyme system would be as capable of discrimination in directing cyclization as in inserting oxygen. The isolation of 1,2,3-trioxy compounds from *F. carolinienis* shows that there is no absolute requirement that oxygen appear at C-4, and the simultaneous appearance of both patterns in *F. albicaulis* returns the question to a standoff.

The absence of 8-oxygenation in these compounds sets them apart from the other highly oxygenated xanthones of the Gentianaceae.[5,13] This difference is particularly germane to the biosynthesis of these compounds, since although the previously found 5,8- and 7,8-oxygenated molecules can be envisioned as arising by eliminative ring closure from the same 2,3,6-trioxy precursor,[14] the 5- and 7- substituted molecules cannot be so treated and favor an oxidative coupling route,[15,16] or the spiran rearrangement proposed by Gottlieb.[17] It remains to be seen whether these compounds actually represent different modes of cyclization or whether, as is more probable, there is a single path clouded by branching and/or adventitious oxidation. Thus swerchirin and its relatives could also arise from a basic 1,3,5-trioxyxanthone by a reasonable oxidation at the activated site *para* to the 5-oxy group, while the 7,8-dioxy compounds could derive similarly from a 1,3,7-trioxy precursor or its biosynthetic analog.

The types of methylation found among these products are much more varied than any previously encountered in this family. No examples of either 1-methoxy or totally O-methylated compounds had been observed earlier, and very few are known even from other families.[4,18–20] The 2-OH compounds J and K are particularly unusual, especially since they occur accompanied by the more common 1-OH isomers. A study of the biosynthetic sequence of oxidations and methylations leading to these products would be of considerable interest and is currently under way.

Preliminary examination of *F. montana* Muelford, a species very similar to *F. albicaulis* but found only in a restricted area of Idaho, demonstrated that both species afford the same complex display of xanthone spots on comparative TLC of their crude extracts and also yield similarly large amounts of non-xanthonic resin. Thus as in the case of the large species,[2] there is excellent agreement in chemistry among

species of similar morphology. Similarly, the two groups show strong, although less close, similarities in their products but significant differences from those produced by *Swertia* species.[2]

EXPERIMENTAL

All m.ps were taken on a Kofler micro hotstage and are corrected. NMR spectra were obtained on a Varian A-60 spectrometer. The numbers in parentheses refer to the multiplicity and relative areas of the peaks. UV spectra were taken on a Cary 14 spectrometer in 95% EtOH. Combustion analyses were performed by Dr. Alfred Bernhardt, Mulheim, Ruhr, West Germany. TLC was done on Silica Gel G from E. Merck, Darmstadt, using mixtures of EtOAc and hexane as solvents, while column separations were on Grace-Davidson 924 silica gel, 200–325 mesh, using the same solvents. Solutions were dried over $MgSO_4$ unless otherwise stated.

Isolation. The roots (1644 g) of *Frasera albicaulis* Dougl. *ex* Griesb., collected during late July at Satus Pass, Washington, were cut into lengths of approximately one inch and ground with 12 l. MeOH. After 15 min the deep yellow soln was decanted from the roots, which were washed twice more with additional solvent. The extracts were combined and concentrated by distilling off the excess MeOH until a distillate temp of 75–80° was obtained. Water (5% by vol) was added to the concentrated soln, which was then continuously extracted in a liquid–liquid extractor with 20% CH_2Cl_2 in pentane. The solvent was changed periodically and the extraction was continued until the extracts remained colorless. Evaporation of these extracts gave 27·36 g of crude product as a dark yellow resin. A solid material separated at the interface during the extraction and was filtered off, 2·36 g. An additional 4·37 g of resin (total yield 31·73 g, 1·95%) were obtained upon workup after the roots had soaked a further six weeks in MeOH.

Column chromatography of 12·78 g of extract on silica gel (column 1900 × 31 mm) gave nine primary fractions homogeneous by TLC. The initial solvent was 2:1 hexane–EtOAc, and the concentration of EtOAc was increased as the separation progressed as shown in Table 3.

TABLE 3.

Frac. No.	Components	EtOAC–hexane	Vol (ml)	Wt (g)
Forerun	—	1:2	1325	4·990
1	A	1:2	625	0·495
2	B, C	1:2	975	0·669
—	—	1:2	250	0·197
3	D, E, F, G, H	1:2	675	1·132
—	—	1:2	1650	0·641
4	I	1:2	2450	0·624
—	—	1:1	1875	0·187
5	J	1:1	1575	0·820
—	—	1:1	600	0·053
6	K	2:1	1725	0·254
—	—	2:1	1000	—*
7	L, M	2:1	325	0·033
—	—	2:1	1500	0·300
8	N	EtOAc	2500	0·135
9	O	EtOAc	3000	0·169

* Owing to a malfunction of the fraction collector most of this material and some of fraction 7 were lost.

Fraction 1. A light yellow crystalline solid (495 mg) was obtained after evaporation of solvent. Sublimation of 220 mg followed by crystallization from CH_2Cl_2–hexane gave light yellow needles of xanthone A suitable for analysis, first crop 20 mg, m.p. 169–170°, m.m.p. with 1-hydroxy-3,7-dimethoxyxanthone 168–170°. Comparison by NMR, UV, and TLC confirmed the identity; UV max: 238 (28.800), 259 (38.950), 308 (13.800), 396 (6250) mμ. (Found: C, 66·60; H, 4·50. $C_{15}H_{12}O_5$ requires: C, 66·17; H, 4·44%).

Fraction 2. Crystallization from CH_2Cl_2–hexane of the first material eluted in this fraction yielded bright yellow needles. A broad melting range and a complex NMR spectrum clearly indicated the material to be a mixture, however.

The entire fraction (670 mg) was rechromatographed on 150 g of silica gel. The first material eluted was crystallized from CH_2Cl_2–hexane to give 25 mg of xanthone B, m.p. 116–119°. Recrystallization gave material suitable for analysis, m.p. 118–119°, m.m.p. with 1-hydroxy-2,3,4,7-tetramethoxyxanthone[2] 116–118°. NMR, UV, and TLC confirmed the identification; UV max: 236 (29,950), 270 (33,970), 303 (10,700), 389 (5650) mμ. (Found: C, 61·50; H, 4·85. $C_{17}H_{16}O_7$ requires: C, 61·44; H, 4·85%).

A portion (230 mg) of the mixture recovered by stripping the column from which B had been isolated was taken up in CH_2Cl_2 and extracted three times with 10% NaOH. The aqueous layer was neutralized and extracted with ether, which was dried and evaporated to yield 52 mg of xanthone C. Crystallization from CH_2Cl_2–hexane gave bright yellow needles, first crop 40 mg, m.p. 188–190°, m.m.p. with swerchirin from *Swertia chirata* 186–188°. Identity confirmed by NMR, UV, and TLC; UV max: 237 (19,900), 254 (26,900), 278 (16,900), 336 (11,300) mμ. (Found: C, 62·64; H, 4·31. $C_{15}H_{12}O_6$ requires: C, 62·50; H, 4·20%).

The dichloromethane soln was washed with water, dried, and evaporated to give 148 mg of xanthone B. Crystallization from CH_2Cl_2–hexane yielded yellow needles, first crop 30 mg, m.p. 117–119°.

Fraction 3. The entire fraction (1·32 g) was sublimed in high vac and refluxed in CH_2Cl_2. The insoluble material filtered to give 122 mg of xanthone D. Recrystallization from THF gave bright yellow needles, m.p. 244–246°; UV max: 234 (25,600), 266 (26,600), 316 (9200), 376 (6140) mμ. (Found: C, 62·60; H, 4·54. $C_{15}H_{12}O_6$ requires: C, 62·50; H, 4·20%).

Acetylation of xanthone D with Ac_2O–pyridine yielded the diacetate, m.p. 170–173°: NMR (CH_2Cl_2): τ 2·46 (q, 1), 2·64 (m, 2), 3·28 (s, 1), 6·01 (s, 3), 6·12 (s, 3), 7·62 (s, 3), 7·66 (s, 3); UV max: 245, 354, 360 mμ. (Found: C, 61·78; H, 4·33. $C_{19}H_{16}O_8$ requires: C, 61·29; H, 4·33%).

Methylation with Me_2SO_4 and NaH in THF gave the neutral dimethyl ether, m.p. 187–189°, m.m.p. with 1,3,4,7-tetramethoxyxanthone 186·5–188°; NMR (CH_2Cl_2): τ 2·42 (q, 1), 2·72 (m, 2), 3·61 (s, 1), 6·04 (s, 3), 6·06 (s, 3), 6·15 (s, 6); UV max: 236, 260, 310, 368 mμ. (Found: C, 64·50; H, 5·21. $C_{17}H_{16}O_6$ requires: C, 64·55; H, 5·10%).

After removal of xanthone D a portion of the material (809 mg) was methylated with 0·6 ml of Me_2SO_4 and excess NaH in 20 ml of THF. After 4 hr 1 ml of Me_2SO_4 and 2 drops of water were added and the mixture allowed to stand overnight. The soln was added to 100 ml of 10% NaOH and heated on the steam bath 1 hr. The basic soln was cooled and extracted with CH_2Cl_2, which was washed with Claisen's alkali and water, dried, and the solvent evaporated to give 650 mg of methylated products.

The mixture was separated by preparative TLC to give fractions (i), 365 mg, and (ii), 138 mg, with an intermediate fraction of 85 mg.

Fraction 3(i). Fraction (i; 200 mg) was separated by preparative TLC on silica gel plates containing H_2SO_4.[2] Elution with 1:1 EtOAc–hexane separated the mixture into a dark orange band at the front and a trailing yellow band at the rear. The bands were scraped from the plates and extracted with CH_2Cl_2–MeOH. These solns were washed with $NaHCO_3$, dried, and passed through basic alumina to remove acidic impurities. Crystallization from CH_2Cl_2–hexane gave 101 mg of methylxanthone E from the orange band and 49 mg of methylxanthone F from the yellow. Methylxanthone E had m.p. 144–145°, m.m.p. with 1,2,3,4,5-pentamethoxyxanthone[2] 142–144°; NMR, UV, and TLC confirmed the identity; UV max: 247 (33,200), 253 (38,500), 294 (10,100), 357 (5050) mμ. (Found: C, 62·39; H, 5·33. $C_{18}H_{18}O_7$ requires: C, 62·42; H, 5·24%).

Direct comparison of methylxanthone F, m.p. 134–135°, and 1,2,3,7-tetramethoxyxanthone[2] showed their identity, m.m.p. 134–136°; UV max: 243 (32,100), 258 (32,700), 280 (11,150), 312 (11,600), 355 (6600) mμ. (Found: C, 64·36; H, 5·12. $C_{17}H_{16}O_6$ requires: C, 64·55; H, 5·10%).

Fraction 3(ii). A portion of fraction (ii), 95 mg, was subjected to preparative TLC and the front edge of the band was removed. Extraction of the silica gel yielded 22 mg of methylxanthone G. Recrystallization from CH_2Cl_2–hexane gave 10 mg of white needles, m.p. 176–177°, m.m.p. with synthetic 1,3,4,5-tetramethoxyxanthone, 176–177°; UV max: 240 sh (30,600), 248 (43,200), 308 (16,300), 351 (6000) mμ. (Found: C, 64·72; H, 5·12. $C_{17}H_{16}O_6$ requires: C, 64·55; H, 5·10%).

Fraction 4. The material from fraction 4 (624 mg) was rechromatographed on a short column of silica gel. Crystallization of the eluted material from CH_2Cl_2–hexane gave xanthone I, light yellow needles, first crop 236 mg, m.p. 188–190°, m.m.p. with an authentic sample of 1-hydroxy-2,3,5-trimethoxyxanthone[2] 186–190°; UV max: 221 (20,300), 243 (30,900), 253 sh (29,200), 295 sh (19,150), 303 (15,500), 370 (4050) mμ. (Found: C, 63·51; H, 4·75. $C_{16}H_{14}O_6$ requires: C, 63·57; H, 4·67%).

Fraction 5. Sublimation followed by crystallization from CH_2Cl_2–hexane yielded bright yellow prisms of xanthone J, first crop 434 mg. Recrystallization produced material suitable for analysis, m.p. 145–146°; UV max: 241 (29,200), 267 (27,700), 290 (9360), 320 (6750), 379 (6000) mμ. (Found: C, 61·42; H, 4·95. $C_{17}H_{16}O_7$ requires: C, 61·44; H, 4·85%).

Methylation with MeI and K_2CO_3 in acetone for 2 hr gave the methyl ether, white crystals from CH_2Cl_2–hexane, m.p. 124–125°, m.m.p. with authentic 1,2,3,4,7-pentamethoxyanthone[2] 123–125°; UV max: 241, 262, 289, 364 mμ. (Found: C, 62·65; H, 5·54. $C_{18}H_{18}O_7$ requires: C, 62·42; H, 5·24%).

Fraction 6. The crude material (253 mg) was sublimed, taken up in CH_2Cl_2, and extracted with Claisen's alkali. The basic soln was neutralized and extracted with CH_2Cl_2, which was dried and evaporated to give 156 mg of a yellow-orange solid residue. Preparative TLC on silica gel followed by crystallization from CH_2Cl_2–hexane gave 52 mg of xanthone K, light yellow crystals m.p. 190–191·5°, m.m.p. with 2-hydroxy-1,3,7-trimethoxyxanthone 190–191°. Direct comparison by NMR, UV, and TLC confirmed the identification; UV max: 226 sh (24,200), 246 (36,100), 256 sh (28,400), 283 (12,400), 332 (12,520), 368 (7530) mμ. (Found: C, 63·31; H, 4·85. $C_{16}H_{14}O_6$ requires: C, 63·57; H, 4·67%).

Fraction 7. Fraction 7 (35 mg) was passed through a short column of basic alumina. Originally a dark yellow oil, it yielded 10 mg of white crystalline product. This was homogeneous by TLC, but had a broad melting range and a mixed NMR.

Fraction 8. Sublimation of the crude product gave 95 mg of light yellow-brown solid. Preparative TLC on silica gel followed by passage through basic alumina yielded 75 mg of a white crystalline residue. Crystallization from CH_2Cl_2–hexane gave white needles of xanthone N, first crop 43 mg, m.p. 176–177°, m.m.p. with 1,3,4,5-tetramethoxyxanthone 176–177°. Direct comparison by NMR, UV, and TLC verified the identity. UV max: 240 sh (36,600), 248 (43,200), 308 (16,300), 351 (6000) mμ. (Found: C, 64·74; H, 5·02. $C_{17}H_{16}O_6$ requires: C, 64·55; H, 5·10%).

Fraction 9. A portion of fraction 9 (124 mg) was passed through a short column of basic alumina to yield 89 mg of a white crystalline residue which was recrystallized from CH_2Cl_2–hexane to give fine white needles of xanthone O, first crop 65 mg, m.p. 187–188·5°, m.m.p. with 1,3,4,7-tetramethoxyxanthone 186–188°; UV max: 337 (26,000), 259 (39,800), 309 (10,300), 367 (7720) mμ. (Found: C, 64·72; H, 5·50. $C_{17}H_{16}O_6$ requires: C, 64·55; H, 5·10%).

3,7-Dimethoxy-1-hydroxyxanthone (**1**).[7] 1,3-Dihydroxy-7-methoxyxanthone[18] (164 mg) was refluxed with 200 mg of anhyd K_2CO_3, 0·60 ml MeI, and 10 ml of acetone for 1·5 hr. The acetone was evaporated, and the residue was taken up in CH_2Cl_2 and extracted with Claisen's alkali. Neutralization of the extract, extraction with CH_2Cl_2, and crystallization from CH_2Cl_2–hexane gave 86 mg of light yellow needles, m.p. 167·5–169·0° (lit.[7] 167°); UV max: 238 (28,800), 259 (38,950), 308 (13,800), 396 (6520) mμ. Methylation in the same fashion for 8·5 hr yielded 1,3,7-trimethoxyxanthone m.p. 172–174° (lit.[21] 171–173°).

2-Hydroxy-2',3,3',4,6-pentamethoxybenzophenone. 2,3-Dimethoxybenzoic acid (1·0 g, 0·0055 moles) and 1,2,3,5-tetramethoxybenzene[22] (1·0 g, 0·0051 moles) were left in 5 ml trifluoroacetic anhydride for 23 days at room temp. The reaction mixture was poured into water and extracted with CH_2Cl_2, which was washed with 10% NaOH, dried, and evaporated to give a bright red oil. Attempts to crystallize the product failed. No further purification was attempted, and the mixture of benzophenone **12** and tetramethoxybenzene was dissolved in 15 ml of anhyd ether and treated with $AlCl_3$ (1·0 g). After standing overnight, the reaction showed (TLC) two main components, with only a trace of starting material. The mixture was poured into water, heated on the steam bath, and extracted with CH_2Cl_2 which was then extracted with Claisen's alkali. The Claisen's soln was neutralized and extracted with CH_2Cl_2, which yielded a red oily solid after evaporation. This was chromatographed on a short silica gel column and the yellow eluent gave 550 mg of crystalline material after the removal of solvent. TLC again showed two spots, presumed to be the two benzophenones 2-hydroxy-2',3,3',4,6-pentamethoxybenzophenone and 2'-hydroxy-2,3,3',4,6-pentamethoxybenzophenone.

1,3,4,5-Tetramethoxyxanthone (**10**). The mixture of isomeric benzophenones was refluxed in pyridine (10 ml) and 10% tetramethylammonium hydroxide in water (10 ml) for 24 hr. TLC showed two products, one corresponding to 1,2,3,5-tetramethoxyxanthone and identical to an authentic sample[2] in its properties. The reaction was poured into dil HCl and extracted with CH_2Cl_2. This extract was washed with Claisen's alkali and water, passed through a short column of basic alumina, and separated by preparative TLC. The product was extracted from the adsorbent with MeOH, sublimed, and crystallized from CH_2Cl_2–hexane to give 123 mg of white needles, m.p. 175–177°; NMR (CH_2Cl_2) τ 2·25 (q, 1), 2·80 (m, 2), 3·57 (s, 1), 5·99 (s, 3), 6·01 (s, 3), 6·06 (s, 3), 6·10 (s, 3); UV max 352, 310, 248, 240 sh mμ.

1-Hydroxy-3,4,5-trimethoxyxanthone (**11**). 1,3,4,5-Tetramethoxyxanthone (43 mg), ether (20 ml), and

an excess of $AlCl_3$ were allowed to stand at room temp for 8 days. The reaction mixture was poured into dil HCl, warmed on the steam bath, and extracted with CH_2Cl_2 which was then extracted with Claisen's alkali. Acidification of the basic soln yielded a light yellow flocculent ppt that was extracted into ether. Evaporation of the solvent gave 22 mg of crystalline residue, and crystallization from CH_2Cl_2–hexane gave yellow needles, first crop 11·5 mg, m.p. 196–201°. Recrystallization from acetonitrile gave material suitable for analysis, m.p. 199·5–201°; NMR (CH_2Cl_2) τ −2·64 (s, 1), 2·23 (q, 1), 2·72 (m, 2), 3·61 (s, 1), 5·98 (s, 3), 6·06 (s, 3), 6·09 (s, 3); UV max: 246 (29,800), 256 (27,000), 318 (13,300), 371 (4430) mμ. (Found: C, 63·73; H, 4·85. $C_{16}H_{14}O_6$ requires: C, 63·57; H, 4·67%).

2,2',3,4,5',6-*Hexamethoxybenzophenone* (**21**). 1,2,3,5-Tetramethoxybenzene[19] (750 mg, 0·0038 moles) was allowed to react with 1·0 g (0·0055 moles) of 2,5-dimethoxybenzoic acid and 5 ml of trifluoroacetic acid for one week. The soln was poured into dil NaOH and then extracted with CH_2Cl_2 which was evaporated to give 1·65 g of crude benzophenone. Crystallization attempts failed; NMR (CH_2Cl_2) τ 2·80 (m, 1), 3·10 (m, 2), 3·40 (s, 1), 6·18–6·72 (6 singlets, each 3H).

2-*Hydroxy*-2',3,4,5',6-*pentamethoxybenzophenone* (**22**). The above benzophenone was dissolved in 25 ml of dry ether containing a slight excess of $AlCl_3$. After standing overnight the soln was hydrolyzed with dil acid, and extracted with ether. Crystallization of the residue from the extract yielded a first crop of 482 mg, yellow needles, m.p. 127–130°. Recrystallization (CH_2Cl_2–hexane) gave material suitable for analysis, m.p. 128–130°; NMR (CH_2Cl_2) τ −2·85 (s, 1), 3·13 (m, 3), 3·25 (s, 1), 6·10 (s, 3), 6·12 (s, 3), 6·25 (s, 3), 6·34 (s, 3), 6·59 (2, 3); UV max: 302, 338 sh mμ. (Found: C, 62·31; H, 5·75. $C_{18}H_{20}O_7$ requires: C, 62·06; H, 5·79%).

1,3,4,7-*Tetramethoxanthone* (**4**). The benzophenone **22** (145 mg) was refluxed in pyridine (10 ml) and tetramethylammonium hydroxide (10% in water, 10 ml) for 48 hr. The solution was acidified with dil HCl and extracted with CH_2Cl_2, which gave on evaporation 106 mg of a light yellow residue. Crystallization yielded a first crop of 87 mg, m.p. 175–180°, which was dissolved in CH_2Cl_2 and washed with Claisen's alkali to remove starting material. Crystallization from CH_2Cl_2–hexane gave white needles, first crop 53 mg, m.p. 186–188°; NMR (CH_2Cl_2) τ 2·35 (q, 1), 2·68 (m, 2), 3·57 (s, 1), 6·01 (s, 3), 6·03 (s, 3), 6·10 (s, 6); UV max: 367, 309, 259, 235 mμ.

1-*Hydroxy*-3,4,7-*trimethoxyxanthone* (**13**). Synthetic 1,3,4,7-tetramethoxyxanthone (52 mg) was dissolved in 15 ml ether and excess $AlCl_3$. After 2 days TLC indicated very little demethylation. $AlBr_3$ (50 mg) was added, the solvent evaporated, and the residue heated at 100° for 1 hr. TLC indicated that some demethylation had occurred, since a spot with properties similar to the xanthones of Fraction 3 had appeared. Dil HCl was added and the mixture was heated on the steam bath 10 min. After cooling, the soln was extracted with CH_2Cl_2, which was dried and filtered. Evaporation of solvent yielded 40 mg of a light yellow crystalline residue that contained both product and starting material by TLC, the latter predominating. Preparative TLC, sublimation, and crystallization from CH_2Cl_2–hexane gave yellow needles (5 mg), m.p. 182–184°; UV max: 234, 267, 312, 384 mμ. (Found: C, 63·61; H, 4·64. $C_{16}H_{14}O_6$ requires: C, 63·57; H, 4·67%).

2-*Hydroxy*-1,3,7-*trimethoxyxanthone* (**17**). 2,5-Dimethoxybenzoic acid (1·0 g, 0·0055 moles) was allowed to react with 1,4-dihydroxy-3,5-dimethoxybenzene[22] (1·0 g, 0·0058 moles) in 7 ml of trifluoroacetic anhydride for 4 weeks at room temp. The reaction was poured into cold water and extracted with CH_2Cl_2 to give 1·09 g of dark reddish brown oil. TLC showed a new spot but attempts to crystallize the product were unsuccessful.

The crude oil was refluxed under N_2 for 48 hr in 10 ml of pyridine and 10 ml of 10% tetramethylammonium hydroxide. After acidification with dil HCl, extraction with CH_2Cl_2 gave 990 mg of dark residue that was sublimed to give 440 mg of product. A sample of this material was purified by preparative TLC on silica gel and crystallization from CH_2Cl_2–hexane gave light yellow needles, m.p. 188–190°; UV max: 225 sh (24,200), 245 (36,400), 255 sh (28,700), 284 (12,500), 322 (12,700), 368 (7390) mμ. (Found: C, 63·60; H, 4·63. $C_{16}H_{14}O_6$ requires: C, 63·57; H, 4·67%).

Methylation with Me_2SO_4 and NaH in THF yielded 1,2,3,7-tetramethoxyxanthone,[2] m.p. 133–135° (lit.[2] 135–136°); NMR (CH_2Cl_2) τ 2·35 (q, 1), 2·68 (m, 2), 3·28 (s, 1), 6·02 (s, 6), 6·09 (s, 3), 6·13 (s, 3); UV max: 243, 258, 280, 312, 355 mμ.

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