purified by reversed phase silica gel prep. TLC with 10% $CHCl_3$ in MeCN to provide F1, F2 and F5 and with 10% H_2O in MeCN to provide F3, F4 and F6) and lutein [1, 9] (0.047 g purified by prep. TLC with 20% EtOAc in hexane), followed by fucoxanthin [10] (0.328 g, purified by prep. TLC on silica gel with 40% EtOAc in hexane).

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STRUCTURE OF VESUVIANIC ACID FROM *STEREOCAULON* SPECIES

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Key Word Index—*Stereocaulon alpinum*; lichen; depsidone; vesuvianic acid.

Abstract—Vesuvianic acid, which was previously isolated from *Stereocaulon vesuvianum* var. *pulvinatum* without a full elucidation of structure, has now been isolated from *S. alpinum*. More detailed spectral investigation indicates that vesuvianic acid, which was previously believed to be a new depsidone, is an ethyl ether of stictic acid which itself may be an artefact produced during extraction.

INTRODUCTION

Depsides and depsidones constitute the largest class of secondary metabolites derived from lichens [1]. Both types of compounds are diphenyl esters, but depsidones also contain a diphenyl ether linkage. Through our present work, which has involved analysis of the chemistry and biological activity of lichens indigenous to Iceland, it has come to our attention that the stability of depsides during extraction with chloroform can be threatened by traces of alcohol present as stabilizer. These findings have been discussed in a previous publication [2].

The isolation of vesuvianic acid from *Stereocaulon vesuvianum* var. *pulvinatum* was previously reported in 1977 [3]. Owing to lack of material, NMR data was not obtained and thus a complete structural elucidation could not be accomplished. However, on the basis of mass spectral, IR and UV data it was considered to be a novel depsidone. During work on *S. alpinum* we have now isolated a compound apparently identical with vesuvianic acid. The structure elucidation of this compound forms the basis of the present communication.

RESULTS AND DISCUSSION

Extraction of dried *S. alpinum* with petrol followed by chloroform allowed the isolation of a compound having identical mass spectral properties with those of vesuvianic acid [3]. IR and UV data were also comparable and were characteristic of a γ-lactonic depsidone. ^{13}C NMR, ^{1}H NMR and mass spectral data all implied the presence

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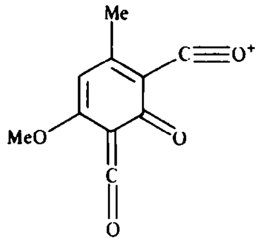

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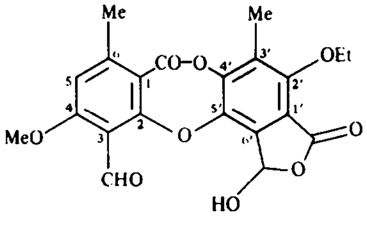

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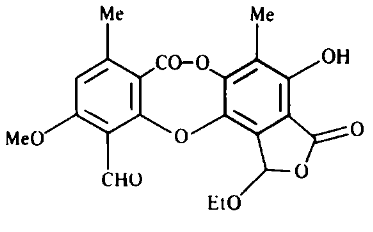

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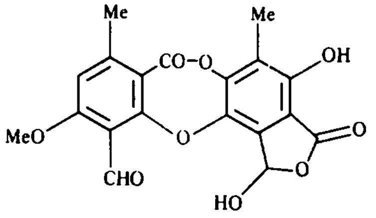

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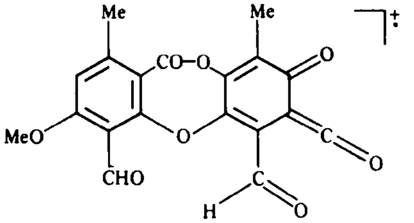

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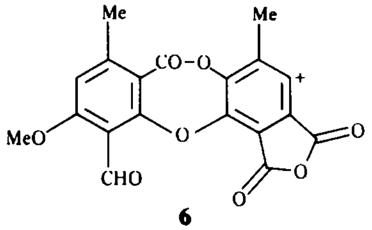
of a moiety (**1**) identical with ring A of stictic acid. Clear signals were observed for the presence of an ethyl ether and, on the basis of the combined spectral evidence, such an ethyl ether could only be accounted for by one of two structures, **2** or **3**. Structure **2** appeared more likely from IR and ^{1}H NMR data. In the IR spectrum of stictic acid (**4**), two bands due to OH stretching were observed, one at 3420 cm^{-1} due to the acetal hydroxyl, another at 3250 cm^{-1} due to the strongly H-bonded phenolic hydroxyl in position 2'. The presence of only one OH stretching band at 3400 cm^{-1} in the IR spectrum of the compound under investigation gave support to **2**, which is without such a strongly bonded hydroxyl. The absence of an intramolecularly bonded hydroxyl group signal in the ^{1}H NMR spectrum (cf. δ 10.29 for stictic acid) was also in agreement with **2**. The signals at δ 7.92 and 6.75 would



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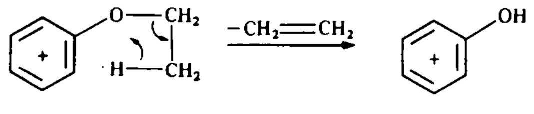

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thus be assignable to an acetal hydroxyl and an acetal H, respectively.

The complete assignment of the ^{13}C NMR spectrum (Table 1) could in theory yield support to either of the two possible structures, **2** or **3**. Readily assignable signals included three carbonyl carbons (δ 89.1, 186.6, 169.7), ethoxyl (δ 66.7, 14.9) and methoxyl (δ 56.6) carbons, two aromatic methyls (δ 22.3, 9.1) and two carbons bearing one H each (δ 111.9, 102.1). Signals for the quaternary carbons 1, 3, 1′ and 6′, although clearly discernible, were of low intensity. The structure of ring A, **1**, suggested by the mass spectrum, was confirmed by the signals at δ 186.6, 111.9 and 56.6 which were attributed to the aldehydic, aromatic and methoxyl carbons (C-3, C-5 and C-4, respectively). Furthermore, the signal at δ 22.3 which would be expected to arise from a methyl group positioned *ortho* to an ester linkage, was assigned to position 6. The ethoxyl carbons were readily assigned as was the methyl signal at δ 9.1. The signal at δ 102.1 was assigned to the acetal carbon and that at δ 189.1 to the 5-membered lactone carbonyl carbon. Other signals were assigned according to additivity calculations [4].

The base mass spectral fragment ion at m/z 368 could be related to either structure **2** or **3**. With regard to **3** it would be associated with the loss of an ethoxyl group and a H from the $[M]^+$ at m/z 414, giving rise to a fragment identical with **5**, which was quite prominent in the mass spectrum of stictic acid (**4**). Prominent peaks at m/z 340 and 312 observed in the mass spectrum could subsequently arise from this fragment. On the other hand, on loss of an ethoxyl fragment and an H, **2** could give rise to **6**. However, the next major fragmentations, involving successive losses of CO would be less likely to arise from **6**. An alternative fragmentation pattern of **2** involving the simultaneous loss of an ethylene fragment and 18 mu to give m/z 368 therefore seems more plausible. Being an aromatic ethyl ether, **2** could lose an ethylene fragment in a manner similar to that of phenetole (**7**), the principal mass spectral fragment of which is **8** [5]. Hence, **2**, on losing both an ethylene fragment and 18 mu would give rise to **5** of m/z 368 which could readily expel successive CO fragments. As mentioned, **5** could also be readily obtained from **3**.

It is clear that mass spectral and ^{13}C NMR data cannot be used to distinguish between the two possible structures, **2** or **3**, but the strong evidence of IR and ^{1}H NMR data in favour of the former, has led to the compound being assigned structure **2**. As mentioned earlier, NMR data of vesuvianic acid isolated originally was not available for comparison with that of the present material, but IR and mass spectral data were identical. UV data were similar although not identical.

It thus seems probable that the two compounds are identical although isolated from different species of *Stereocaulon* and that 'vesuvianic acid', instead of being a new depsidone, is an ethyl ether of stictic acid [2]. It may be that **2** is formed from stictic acid through contact with ethanol during the course of extraction. Ethanol present as the stabilizer in chloroform is extremely difficult to remove and can lead to the formation of artefacts during extraction [2]. Although acetone was used by Solberg in the extraction of *S. vesuvianum*, it is possible that a trace of ethanol could have been present thereby causing ether formation of stictic acid, which was isolated in substantial quantities together with atranorin from the lichen. In addition, the presence of atranol, which could also be produced by reaction with ethanol, would suggest that breakdown of atranorin had occurred but more work is needed to confirm this.

Table 1. ^{13}C NMR data for **2** (vesuvianic acid)

Carbon	Chemical shift (δ, $CDCl_3$)
1	114.8
2	160.6
3	107.9
4	163.6
5	111.9
6	151.4
1 $-CO_2-$	169.7
3 $-CHO$	186.6
4 $-OMe$	56.6
6 $-Me$	22.3
1′	114.2
2′	152.4
3′	120.9
4′	147.3
5′	132.2
6′	123.1
1′ $-CO-$	189.1
2′ $-O\underline{C}H_2Me$	66.7
2′ $-OCH_2\underline{Me}$	14.9
3′ $-Me$	9.1
6′ $-\overset{\vert}{C}HOH$	102.1

EXPERIMENTAL

^{13}C NMR was recorded at 100.6 MHz, ^{1}H NMR spectra at 400 MHz (**2**) or at 200 MHz (stictic acid). TMS was used as int. standard for NMR. MS were recorded at 70 eV. Mps are uncorr.

Extraction. Stereocaulon alpinum Laur. was collected from the moraines of Solheima glacier, V. Skaftafellssysla, Iceland in July and August 1980. The material was identified by Prof. H. Kristinsson, Institute of Biology, University of Iceland. A herbarium specimen of the lichen is deposited in the museum at Chelsea College, University of London. The plant material was air-dried and thoroughly cleansed of all extraneous material prior to being ground into a fine powder. The powdered lichen thallus (800 g) was extracted successively with petrol bp 40–60°, $CHCl_3$, Me_2CO, EtOH, MeOH and H_2O. Several compounds were isolated from the $CHCl_3$ extract.

Isolation of stictic acid (**4**). The acid was isolated in an amorphous form from the cooled $CHCl_3$ extract, mp 259–262°; IR ν_{max}^{Nujol} cm^{-1}: 3420, 3250, 1745, 1730, 1680, 1600, 1550, 1080; UV λ_{max}^{EtOH} (poorly soluble): 228, 265 (sh), 307 nm; ^{1}H NMR (DMSO-d_6): δ 10.47 (1H, *s*, CHO), 10.29 (1H, *s*, Ar–OH, C-2′, exch. D_2O), 8.29 (1H, *s*, acetal OH, $-\overset{\vert}{C}HO\underline{H}$, exchangeable with D_2O), 7.10 (1H *s*, acetal-H, $-\overset{\vert}{C}\underline{H}OH$), 6.63 (1H, *s*, Ar–H, H-5), 3.92 (3H, *s*, OMe), 2.50 (3H, *s*, Ar–Me), 2.20 (3H, *s*, Ar–Me); MS m/z (%, rel. int.): 387 $[M+1]^+$ (5), 386 $[M]^+$ (23), 368 (15), 340 (11), 314 (18), 286 (11), 255 (14), 227 (27), 209 (18), 191 (23), 164 (15), 163 (34), 138 (25), 137 (19), 135 (22), 122 (15), 121 (28), 107 (18), 105 (13), 97 (59), 87 (50), 83 (61), 72 (100).

Isolation of **2**. The residue from evapn of the filtered $CHCl_3$ extract was chromatographed through a column of silica gel eluting with hexane containing increasing concns of $CHCl_3$ in hexane followed by $CHCl_3$ containing increasing concns of MeOH. The hexane–$CHCl_3$ (2:3, 1:4) and $CHCl_3$ eluate fractions were rechromatographed through a silica gel column eluting with petrol containing increasing concns of $CHCl_3$ in petrol followed by $CHCl_3$ containing increasing concns of MeOH in $CHCl_3$. Compound **2** (2 mg) was obtained from the petrol–$CHCl_3$ (3:2) eluate after recrystallization from EtOAc; mp 245°; IR ν_{max}^{KBr} cm^{-1}: 3400, 1745, 1730, 1700, 1604, 1300, 1220, 1130, 1080; UV λ_{max}^{EtOH} nm (poorly soluble): 237, 260 (sh), 297; ^1H NMR ($CDCl_3$): δ10.54 (1H, *s*, CHO), 7.92 (1H, *s*, exch. D_2O), 6.75 (1H, *s*), 6.49 (1H, *s*, Ar–H), 4.01 (2H, *q*, $J = 7.0$ Hz), 3.98 (3H, *s*, OMe), 2.57 (3H, *s*, Ar–Me), 2.30 (3H, *s*, Ar–Me), 1.29 (3H, *t*, $J = 7.0$ Hz); ^{13}C NMR ($CDCl_3$): see Table 1. MS: *m/z* (%, rel. int.): 416 $[M+2]^+$ (3), 415 $[M+1]^+$ (25), 414 $[M]^+$ (95), 370 (11), 369 (42), 368 (100), 341 (15), 340 (30), 312 (16), 287 (16), 285 (11), 210 (19), 191 (11), 58 (33), 43 (56). Accurate mass measurement: found: 414.0952; $C_{21}H_{18}O_9$ requires 414.0949.

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PHENYL INDANE FROM *ACORUS CALAMUS*

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Key Word Index—*Acorus calamus*; Araceae; rhizomes; *Z*-3-(2,4,5-trimethoxy phenyl)-2-propenal; 2,3-dihydro-4,5,7-trimethoxy-1-ethyl-2-methyl-3-(2,4,5-trimethoxyphenyl)indene.

Abstract—Besides three known compounds, two new compounds, namely *Z*-3-(2,4,5-trimethoxy phenyl)-2-propenal and a new phenyl indane have been isolated from the rhizomes of *Acorus calamus*. These compounds have been characterized from their spectral data and by synthesis.

INTRODUCTION

Asarone, the principal constituent of the essential oil of *Acorus calamus* is well known as an insect growth regulator [1]. Since asarone is not entirely responsible for the physiological activity shown by *A. calamus* oil, it was thought worthwhile to reexamine the oil.

RESULTS AND DISCUSSION

The rhizomes of *A. calamus* were dried and then exhaustively extracted with ethanol. The solvent was evaporated and the resulting extract was subjected to rigorous CC over silica gel to give five compounds. Three of these compounds were identified as *Z*-3-(2,4,5-trimethoxy phenyl)-1-propene (asarone, a liquid), 2,4,5-trimethoxybenzaldehyde, mp 115° (lit. [2], mp 115°) and acoradin (a dimer, mp 99°, lit. [2] mp 101°) from their spectral data and comparison with the literature [2]. Besides the known compounds two more compounds were isolated, **4** and **5**.

Compound **4**, $C_{12}H_{14}O_4$, $[M]^+$ 222, mp 85°, was obtained as a colourless crystalline compound from acetone in the form of needles. It gave a positive test with 2,4-DNP indicating the presence of a carbonyl function. The IR spectrum showed strong absorption at 1650 cm^{-1}

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*Contribution No. 273.