SOME HISTORICAL CHEMISTRY: THE STRUCTURES OF DIEUCARVELONES A, B AND C AND THE STRUCTURE OF α-AMYRILENE

Siegfried HUNECK[a], Joseph D. CONNOLLY[b1,*] and Louis FARRUGIA[b2]

[a] *Fliederweg 34A, D 06179 Langenbogen/Saalkreis, Germany; e-mail: shuneck@gmx.de*
[b] *School of Chemistry, University of Glasgow, Glasgow G12 8QQ, Scotland, UK; e-mail: [1] joec@chem.gla.ac.uk, [2] louis.farrugia@glasgow.ac.uk*

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This paper is dedicated to Professor Pavel Kočovský on the occasion of his sixtieth birthday and to our many friends and colleagues from the Czech-Polish Isoprenoid Conferences.

Spectroscopic examination of some historical samples from the terpene collection of Prof. H. Wienhaus resulted in the structure elucidation of dieucarvelones A, B and C, products of treatment of eucarvone with zinc in alkali and in confirmation of the structure of α-amyrilene, a rearrangement product of α-amyrin. The relative stereochemistry of dieucarvelones A and C was established by X-ray crystallographic analyses.
Keywords: X-ray diffraction; Terpenoids; Structure elucidation; Dieucarvelones; α-Amyrilene.

A. v. Baeyer (1835–1917, Munich, Nobel Prize 1905) and O. Wallach (1847–1951, Göttingen, Nobel Prize 1910) were important pioneers of terpene chemistry. In 1894 Baeyer prepared eucarvone by treatment of carvone hydrobromide with alkali[1]. Its structure **1**, with its unusual seven-membered ring, was eventually established by Wallach[1]. At that time, H. Wienhaus was a co-worker of Wallach and took two samples labelled "Eucarvon" when he moved to the Institute of Plant Chemistry and Wood Research at Tharandt in Saxony. One of us (S.H.) was assistant professor in this institute from 1961–1969 and rescued these samples from the terpenoid collection of Wienhaus. The two samples are crystalline and therefore cannot be eucarvone (a liquid). Spectroscopic investigation revealed that they are crystalline mixtures of dieucarvelones, products of the treatment of eucarvone with zinc in alkali (*vide infra*). A third sample rescued from the Wienhaus collection was labelled α-amyrilene and consisted of huge crystals prepared by the Swedish chemists Vesterberg and

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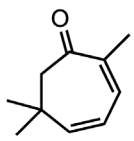
Westerlind in 1922. This paper describes our investigations of these samples using modern spectroscopic methods.

THE DIEUCARVELONES

In 1899 [2] and again in 1914 [3], Wallach reported that the treatment of eucarvone **1** with zinc in alkaline solution afforded a series of isomeric dimeric compounds that he named the dieucarvelones. Based on melting points, he claimed to have separated four isomers, the α-, β-, γ- and δ-forms, by fractional crystallisation from acetic acid/water. In 1957, Büchi and Saari reinvestigated[4] the problem of the structures of the dieucarvelones. They succeeded in preparing the α- and β-isomers but, despite repeated crystallisations, could never isolate the other two isomers. However, they did manage to isolate two new isomers, the ε- and ζ-forms. Chromatography of the β-isomer over alumina afforded the α- and ε-isomers and hence the β-isomer was clearly a mixture. All the dieucarvelones had characteristic double bond peaks in their IR spectra at 3010, 1640 and 760 cm^{-1} and so Büchi concluded that they were unsaturated despite their failure to decolourise bromine in acetic acid or potassium permanganate. He eventually decided that structure **2** was the correct gross structure for these compounds and proposed a reductive Michael addition mechanism involving zinc as in **3**, for their formation. There are ten possible stereoisomers of structure **2**, two *meso* forms and four pairs of enantiomers. Büchi concluded his paper by writing "We have not yet had the courage to embark on a determination of the configurations of the dieucarvelones".



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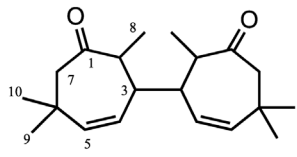

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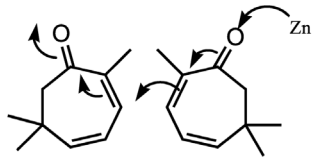

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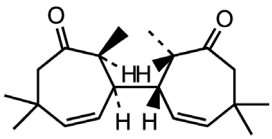
DISCUSSION

The ^{13}C NMR spectra of the two crude dieucarvelone samples 1 and 2 showed that three compounds were present in each but in different proportions. TLC analysis was complicated by the fact that the compounds did not absorb in the UV and showed a reluctance to react with permanganate or with iodine. In time spots did appear with both reagents. Sample 1 contained mainly two compounds, dieucarvelones A (most polar) and B (least polar) in a ratio of about 4:1 with a tiny amount of a third compound, dieucarvelone C, of intermediate polarity. Sample 2 contained C, A and B in a ratio of roughly 3:2:1. These approximate ratios were estimated from the ^{13}C NMR spectra of the mixtures and the analytical TLC plates were in broad agreement.

Preparative TLC separation of sample 1 (approximately 30 mg) on silica gel plates proved to be remarkably successful, affording essentially pure crystalline samples of dieucarvelones A and B. The plates were eluted three times using 10% ethyl acetate in petrol as eluent. The bands were detected by observing their shadows on an overhead projector.

The separation was not as straightforward for sample 2. The least polar band, dieucarvelone B, was readily detected but separation between the next two bands was never observed. Fortunately, removal of the front part of the band gave pure crystalline dieucarvelone C, while the rest of the band gave a mixture of C and A.

The three crystalline dieucarvelones all gave similar ^{1}H and ^{13}C NMR spectra consistent with Büchi's proposed dimeric structure **2**. In all cases the two halves give identical spectra and hence the molecules must be centrosymmetric or *meso*. It was clearly impossible to determine the relative configurations of the whole molecules by NMR and so we decided to use X-ray crystal structure analysis. Satisfactory crystals were grown for dieucarvelones A and C and X-ray analyses revealed the structures and relative stereochemistry as in **4** and **5**, respectively.



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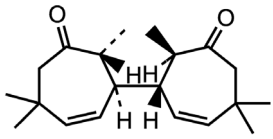

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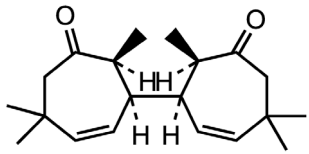
Dieucarvelone A **4** was crystallised from ethanol and had m.p. 122–123 °C. Its EIMS showed a parent ion at *m/z* 302, consistent with the expected molecular formula $C_{20}H_{30}O_2$. A reasonably intense peak at *m/z* 151 was also observed. In the CIMS only the $(M + H)^+$ peak at *m/z* 303 was observed. The ^1H and ^{13}C NMR spectra (see Tables I and II) revealed the presence of two tertiary methyl groups, a secondary methyl group, an isolated methylene group [δ_H 2.11 and 3.14 (AB system, J = 11.3 Hz, 2H-7)], two methines [δ_H 3.05 (bs, H-3), 2.37 (q, J = 6.7 Hz, H-2)], a *cis*-disubstituted double bond [δ_H 5.15 (bd, J = 11.4 Hz, H-4), 5.30 (d, J = 11.4 Hz, H-5); δ_C 127.1 (C-4), 139.1 (C-5)], a quaternary carbon [δ_C 36.2 (C-6)] and a saturated ketone [δ_C 211.5 (C-1)]. The COSY spectrum showed the coupling of H-2 to the secondary methyl group and to H-3. The small coupling between H-2 and H-3 suggested that they are probably *cis* (though we do not know the conformation of the seven-membered ring!). Support for this suggestion was found in a ROESY correlation between the two protons. In turn, H-3 coupled with the double bond proton H-4 and even had a small unresolved allylic coupling to H-5. One of the C-7 methylene protons had a ^4J W coupling with H-5. Further confirmation of the structure came from the HMBC spectrum, especially the three-bond correlations of the methyl groups. It is not possible to deduce the relative configurations of the two rings. The X-ray analysis shows that the molecule is *2S*, *3R*, *3'R*, *2'S* as drawn (Fig. 1). The crystal is chiral as a result of spontaneous resolution of the racemic product on crystallisation.

Dieucarvelone C **5** was also crystallised from ethanol and had m.p. 107–108 °C. Its ^1H and ^{13}C NMR spectra (see Tables I and II) revealed the presence of two tertiary methyl groups, a secondary methyl group, an isolated methylene group [δ_H 2.04 (dd, J = 11.1, 1.5 Hz, H-7) and 3.06 (d, J = 11.1 Hz, H-7)], two methines [δ_H 2.68 (ddd, J = 10.9, 4.2, 1.6 Hz, H-3), 2.44 (dq, J = 10.9, 7.0 Hz, H-2)], a *cis*-disubstituted double bond [δ_H 5.24 (dd, J = 11.8, 4.2 Hz, H-4), 5.35 (dt, J = 11.4, 1.6 Hz, H-5); δ_C 126.0 (C-4), 141.5 (C-5)], a quaternary carbon [δ_C 37.0 (C-6)] and a saturated ketone [δ_C 213.0 (C-1)]. The COSY spectrum showed the coupling of H-2 to the secondary methyl group and to H-3. The coupling between H-2 and H-3 is large suggesting that these protons are probably *trans* (though we do not know the conformation of the seven-membered ring!). In turn, H-3 coupled with the double bond protons H-4 and H-5. In this case the allylic coupling to H-5 was well-resolved. One of the C-7 methylene protons had a ^4J W coupling with H-5. Further confirmation of the structure came from the HMBC spectrum, especially the three-bond correlations of the methyl groups. To determine the relative configurations of the two rings we again resorted to X-ray

crystal structure analysis which showed that the molecule is 2*R*, 3*R*, 3′*R*, 2′*R* as drawn and that H-2 and H-3 are *trans* as predicted (Fig. 2). The crystal is chiral as a result of spontaneous resolution of the racemic product on crystallisation.

Dieucarvelone B was also crystallised from ethanol but no suitable crystal for X-ray analysis could be found. It had m.p. 145–146 °C. Its ^{13}C and ^{1}H NMR spectra (see Tables I and II) revealed the presence of two tertiary methyl groups, a secondary methyl group, an isolated methylene group [δ_H 2.11 (dd, J = 11.3, 1.1 Hz, H-7), 3.24 (d, J = 11.1 Hz, H-7)], two methines [δ_H 3.11 (bs, H-3), 2.58 (q, J = 6.8 Hz, H-2)], a *cis*-disubstituted double bond [δ_H 5.21 (bd, J = 11.4 Hz, H-4), 5.30 (d, J = 11.4 Hz, H-5); δ_C 126.5 (C-4), 140.6 (C-5)], a quaternary carbon [δ_C 37.2 (C-6)] and a saturated ketone [δ_C 212.2 (C-1)]. As for the previous compounds, the COSY spectrum showed the coupling of H-2 to the secondary methyl group and to H-3. The coupling between H-2 and H-3 is small as in dieucarvelone A **4** suggesting a *cis* relationship. In turn, H-3 coupled with the double bond protons H-4 and H-5. In this case, the allylic coupling to H-5 is again small. One of the C-7 methylene protons had a ^{4}J W coupling with H-5. The gross structure was again confirmed by the three-bond correlations of the methyl groups in the HMBC spectrum. Since H-2 and H-3 are *cis* dieucarvelone B must be the *meso* isomer **6** with the configurations 2*S*, 3*R*, 3′*S*, 2′*R* as drawn. We already know that eucarvelone A has the centrosymmetric arrangement 2*S*, 3*R*, 3′*R*, 2′*S*.



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At present, it is difficult to relate dieucarvelones A, B and C directly to the isomers published by Wallach and by Büchi since melting point comparisons are inconclusive. It is likely that the previous isomers were mixtures.

CRYSTALLOGRAPHIC DETAILS

Diffraction data for compounds **4** and **5** were measured at 100 K on a Nonius KappaCCD diffractometer, using MoKα X-radiation. Data reduction using Denzo[5] and merging using SORTAV [6] afforded a unique and complete

data set to θ_{max} = 30°. Initial refinements using a data set merged with *222* symmetry (i.e. containing an almost complete set of Friedel pairs) indicated that the absolute configuration could not be determined with any certainty. A data set merged with *mmm* symmetry, with all Friedel pairs merged, was used for the final refinements. The absolute structures of the deposited models are therefore arbitary. Full crystallographic details, in-

TABLE I
^{13}C NMR chemical shifts of the dieucarvelones ($CDCl_3$, 100 MHz)

C	A **4**	B **6**	C **5**
1	211.5	212.2	213.0
2	47.5	50.6	50.5
3	37.7	37.8	38.2
4	127.1	126.5	126.0
5	139.1	140.6	141.5
6	36.2	37.2	37.0
7	50.9	51.7	52.0
8	8.9	9.1	15.7
9	29.2	30.8	30.4
10	29.9	30.9	30.9

TABLE II
^{1}H NMR chemical shifts of the dieucarvelones ($CDCl_3$, 400 MHz; *J* in Hz)

H	A **4**	B **6**	C **5**
2	2.37 (q, 6.7)	2.58 (q, 6.8)	2.44 (dq, 10.9, 7.0)
3	3.05 (bs)	3.11 (bs)	2.68 (ddd, 10.9, 4.2, 1.6)
4	5.15 (bd, 11.4)	5.21 (bd, 11.4)	5.24 (dd, 11.8, 4.2)
5	5.30 (d, 11.4)	5.30 (d, 11.4)	5.35 (dt, 11.8, 1.6)
7	2.11 (d, 11.3) 2.11 (dd, 11.3, 1.1)	2.04 (dd, 11.1, 1.5) 3.24 (d, 11.3)	3.24 (d, 11.3) 3.06 (d, 11.1)
8	1.01 (d, 6.6)	0.99 (d, 6.7)	1.04 (d, 7.0)
9	0.97 (s)	1.00 (s)	1.02 (s)
10	1.09 (s)	1.10 (s)	1.07 (s)

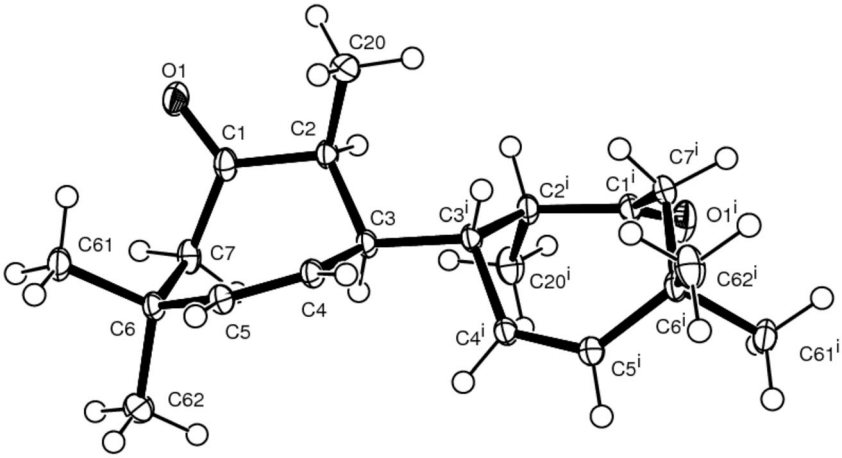


FIG. 1
ORTEP view of compound **4** (dieucarvalone A). Thermal ellipsoids are drawn at the 50% probablity level. Primed atoms are related to unprimed atoms by the crystallographic two-fold axis passing through the midpoint of C3–C3^{i}, symmetry code: x, $1 - y$, $2 - z$

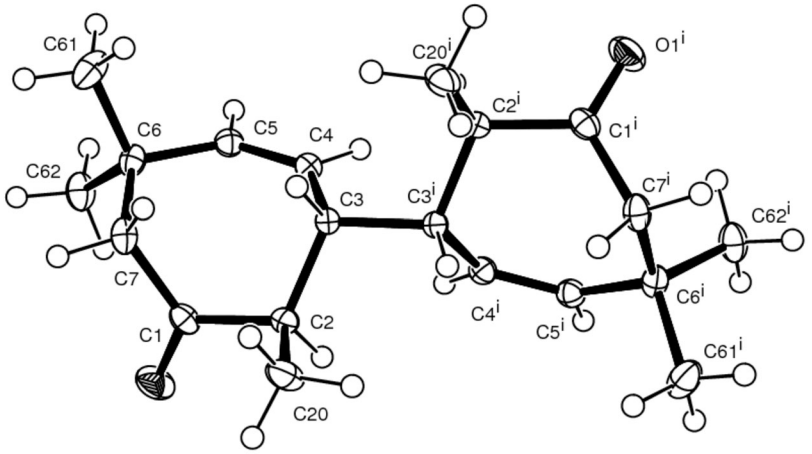
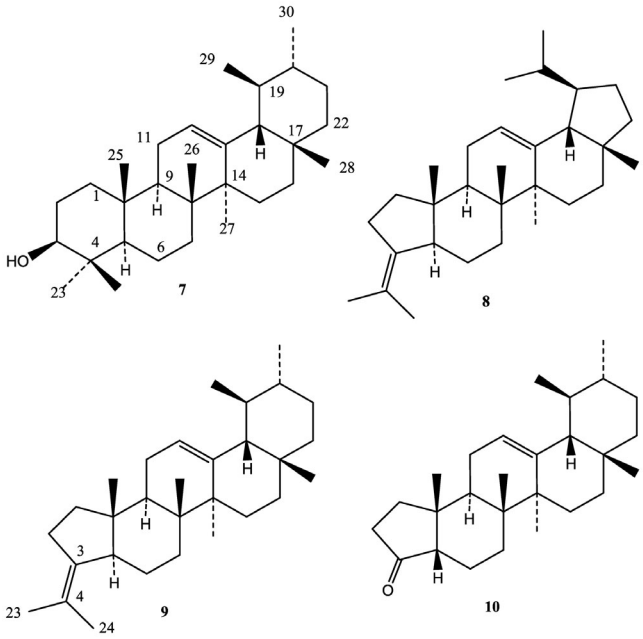


FIG. 2
ORTEP view of compound **5** (dieucarvalone C). Thermal ellipsoids are drawn at the 50% probablity level. Primed atoms are related to unprimed atoms by the crystallographic two-fold axis passing through the midpoint of C3–C3^{i}, symmetry code: x, $1/2 - y$, $1/2 - z$

cluding final refined coordinates have been deposited in CIF format with the Cambridge Crystallographic Data Centre, deposition codes CCDC 861354 and CCDC 861355.

α-AMYRILENE

α-Amyrin **7** is a pentacyclic triterpenoid which is widely distributed in Nature. In 1922 Vesterberg and Westerlind treated α-amyrin with PCl_5 and converted it into a new compound m.p. 134 °C, which they named α-amyrilene[7]. At that time neither the structure of α-amyrin nor



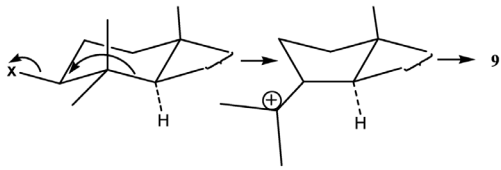
α-amyrilene was known. Spring and his colleagues reinvestigated[8,9] this problem in 1955 and proposed structure **8** for α-amyrilene. Unfortunately, they used the wrong structure for α-amyrin and the correct structure of α-amyrilene should be **9**. However they correctly deduced that the reaction with PCl_5 involved a contraction of ring A with formation of an isopropylidene group. Ozonolysis of the isopropylidene group afforded a cyclopentanone **10** (ν_{max} = 1740 cm^{-1}) which was stable to base and hence the AB ring junction is *cis*. The formation of α-amyrilene can be readily rationalised in modern mechanistic terms as shown in Scheme 1 which

TABLE III
^{13}C NMR chemical shifts of α-anyrilene **9** and α-amyrin **7**[a]

Carbon	**9**	**7**
1	39.5	38.7
2	28.2	27.2
3	135.5	78.3
4	120.9	38.7
5	55.8	55.2
6	25.5	18.3
7	32.9	32.9
8	39.9	40.0
9	45.3	47.7
10	44.0	36.9
11	21.3	23.3
12	124.7	124.3
13	140.1	139.3
14	42.2	42.0
15	26.8	26.6
16	28.7	28.7
17	33.9	33.7
18	59.4	58.9
19	39.7	39.6
20	39.7	39.8
21	31.3	31.2
22	41.6	41.5
23	19.5[b] (1.78)	28.1
24	22.9[b] (1.62)	15.6
25	15.0 (0.76)	15.6
26	17.0 (1.08)	16.8
27	23.3 (1.12)	23.3
28	28.8 (0.84)	28.8
29	17.5 (0.83)	17.4
30	21.4 (0.95)	21.3

[a] The ^{1}H NMR shifts of the methyl groups are shown in parentheses. [b] May be interchanged.

suggests that the AB *trans* ring junction should be retained in the product. Evidence for this will be provided during the discussion of the NMR spectra which are entirely consistent with structure **9** for α-amyrilene.



SCHEME 1

The ^{13}C NMR spectrum (Table III) of α-amyrilene **9** is in agreement with the molecular formula $C_{30}H_{48}$. It showed the presence of trisubstituted [δ_C 124.7 (CH-12), 140.1 (C-13); δ_H 5.18 (bt, H-12)] and tetrasubstituted [δ_C 135.5 and 120.9 (C-3 and C-4)] double bonds and eight methyl groups. The ^1H NMR spectrum revealed that two of the methyls are vinylic and two secondary. Comparison of its ^1H and ^{13}C NMR spectroscopic properties with those of α-amyrin **7** indicated that the two compounds were very similar, as expected, and that the differences were associated with ring A. The ^{13}C NMR shifts of the two compounds are listed in Table III. The HMBC correlations of the methyl groups of α-amyrilene and comparison with the chemical shifts of α-amyrin enabled the structure to be determined and all the resonances to be assigned. The presence of an isopropylidene moiety followed from the mutual HMBC correlations of Me-23 and Me-24 and their correlations with the olefinic carbons C-3 and C-4. The HMBC correlations of the remaining tertiary/secondary methyls (Table IV) confirmed the α-amyrin nature of rings B, C, D and F and enabled the carbon resonances to be assigned. The residual question concerning the configuration of the proton at C-5 was resolved by the NOESY spectrum which showed a clear correlation between H-5 and H-9α. Hence H-5 is also α and the AB ring junction is *trans*. During the ozonolysis experiment carried out by Spring and his colleagues (*vide supra*) epimerisation occurred at C-5 to give a *cis* AB ring junction.

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