Pergamon

PII: S0031-9422(96)00464-5

Phytochemistry, Vol. 44, No. 3, pp. 437–440, 1997

0031-9422/97 $17.00 + 0.00

CRYPTOCARYA LIEBERTIANA AND *OCOTEA BULLATA*—THEIR PHYTOCHEMICAL RELATIONSHIP

SIEGFRIED E. DREWES,* MARION H. HORN and STEPHEN MAVI†

Department of Chemistry and Chemical Technology, University of Natal, Pietermaritzburg, 3200, South Africa; †National Herbarium and National Botanical Garden, P.O. Box 8100, Causeway, Zimbabwe

(*Received* 26 *April* 1996)

Key Word Index—*Ocotea bullata*; *Cryptocarya liebertiana*; Lauraceae; ocobullenone; ocobullenone precursor; cryptofolione; phytochemical relationship.

Abstract—Isolation of the major components from the bark of mature *Cryptocarya liebertiana* shows that ocobullenone and its precursor (both previously isolated from *Ocotea bullata*) are present in detectable concentrations in this source. These particular compounds are absent in other South African *Cryptocarya* species. The result is significant since *Cryptocarya* bark is currently being utilized by traditional healers as an alternative to *O. bullata* bark. Copyright © 1997 Elsevier Science Ltd

INTRODUCTION

Interest in the Lauraceae, a pantropical family with about 50 genera and 2500–3500 [1] species, is of considerable interest to both taxonomists and natural product chemists. In his monograph on dicotyledonous flowering plants, Rohwer [1] makes the following statement about *Cryptocarya* (a prominent genus of the Lauraceae): "in *Cryptocarya* there are various patterns of exine sculpture, from almost smooth to distinctly regulose [2, 3] or densely covered with minute spinules. This indicates that the genus *Cryptocarya* may not be a natural group". In South Africa the family Lauraceae is represented by the following species: *Ocotea bullata, O. kenyensis, Cryptocarya latifolia, C. woodii, C. myrtifolia, C. wyliei* and *C. libertiana*. The chemical constituents of all of these species (other than *C. libertiana*) have been examined previously by us [4–7]. It is hoped to elaborate on the phytochemical relationships between these plants in a future joint publication with authoritative taxonomists in the area.

The present investigation relates specifically to our interest in establishing a chemical link between *O. bullata* and representatives of the *Cryptocarya* genus in view of the fact that traditional healers have recently turned to the latter genus as an alternative (and more accessible) source for their "muthi" (medicinal) requirements [8]. To date eight new α-pyrones [4–7] have been isolated from *C. latifolia, C. woodii, C. myrtifolia and C. wyliei*. Some of these α-pyrones are shown in (**1**), (**2**) and (**3**). They are clearly different from the neolignan ocobullenone (**4**) and its open chain precursor (**5**) from *O. bullata*.

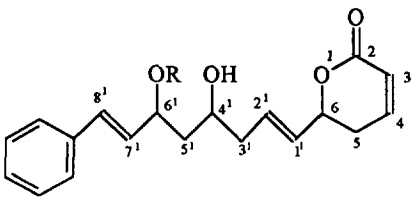
Since the four *Cryptocarya* species mentioned exhibited no traces of ocobullenone or related compounds we turned our attention to the relatively rare (in South Africa) *Cryptocarya libertiana*. This tree has a limited distribution in the province of Kwazulu-Natal (Ngoye forest) and also occurs in the Northern Province in the Louis Trichardt district. It grows reasonably plentifully in Zimbabwe. Authenticated bark specimens from these areas were obtained and examined.

RESULTS AND DISCUSSION

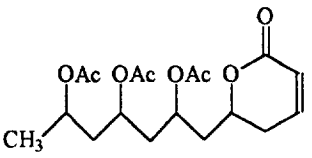
The Ngoye specimen afforded the two known α-pyrones, cryptocaryalactone and deacetylcryptocaryalactone, previously obtained from *C. bourdilloni* and *C. moschata* [9, 10]. The bark from Louis Trichardt gave a new α-pyrone (**6**), clearly a dehydration product of cryptofolione (**1a**). The NMR spectral evidence supports fully the proposed structure. Strong confirmation for this conclusion is provided by the mass spectrum which exhibits the base peak at m/z 159. This represents the stable fragment (**7**) and confirms the existence of a single hydroxy group.

The sample from Zimbabwe consisted of bark from an immature tree as well as that of a mature specimen. The major compound in the former was again cryptofolione (**1a**). It was accompanied by the monoacetate (**1b**). The ^{1}H NMR spectrum was very similar to that of cryptofolione but the proton on the C-6′ (site of the acetate) shifts from δ 4.63 to 5.69, as can be expected. Examination of the mature bark gave an unexpected result. There appeared to be no α-pyrones

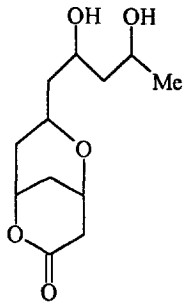
*Author to whom correspondence should be addressed.



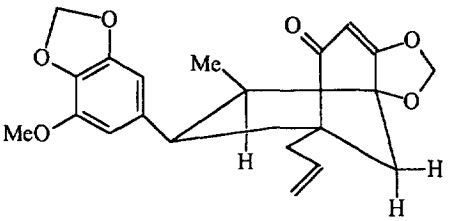
1a R = H Cryptofolione
1b R = Ac



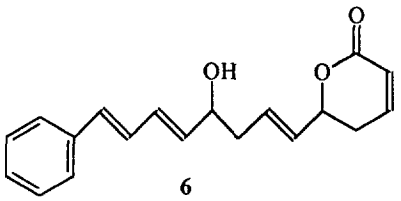
2 "Triacetate"



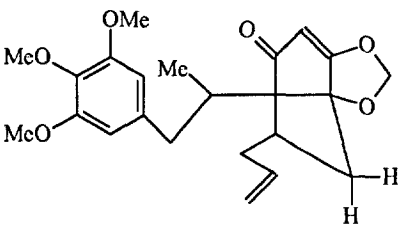
3 Bicyclic derivative



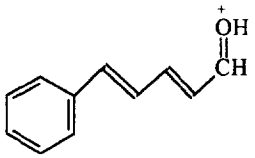
4 Ocobullenone



6



5



7 m/z 159

present, instead, ocobullenone (**4**) and its open chain precursor (**5**) were isolated in 0.01 and 0.007% (based on dry bark) yield, respectively. From the examination of the extract, these were undoubtedly the major constituents present. Comparison of the two compounds with authentic specimens from *O. bullata* (^{1}H NMR, ^{13}C NMR, mass fragmentation, GC retention times) confirmed their identity. The 'new' (**5**) was racemic and its mp was 128° as opposed to 105° reported for the original (**5**) [11]. The 'new' ocobullenone had a rotation of +158° (as opposed to +204°) and its mp was 136° (compared with 151°) [4]. We have no doubt that these two compounds are truly identical and that the slightly depressed constants are due to the presence of traces of the diastereomeric iso-ocobullenone [11] which proved impossible to remove (Fig. 1).

These results, apart from illustrating, by chemical means, that *Ocotea bullata* has two major compounds in common with a specific *Cryptocarya* species, and thereby providing some scientific foundation for the observed use of both genera for common medicinal purposes, also raises some interesting questions. For example, what happens when cryptofolione (**1**) (which is universally present in all the *Cryptocarya* examined) once *C. liebertiana* reaches maturity? Do the other South African *Cryptocaryas* possibly contain mere

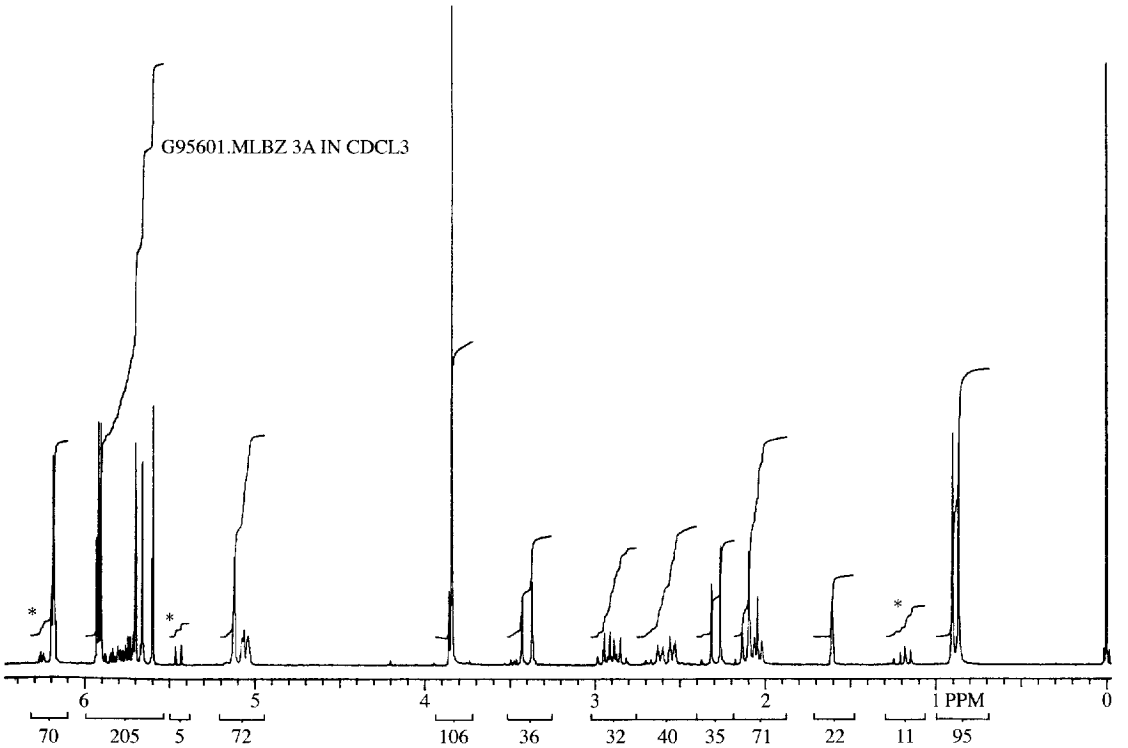


Fig. 1. 1H NMR spectra of ocobullenone (**4**) containing traces of iso-ocobullenone.*

trace amounts of the ocobullenone-type of compound? Will *C. liebertiana* which also exists further north in Africa (e.g. in Zambia and Kenya), have similar constituents to the plant from Zimbabwe? In order to find answers to these questions we are investigating the family Lauraceae on a much broader front and are including the genus *Dahlgrenodendron* which has been placed with the Lauraceae although this is a contentious point [1].

EXPERIMENTAL

General. NMR: ^{1}H (200 MHz) and ^{13}C (50 MHz); EI-MS: 70 eV; CC: silica gel 60 (Macherey Nagel); Chromatotron: silica gel 60F_{254}.

Plant Material. Cryptocarya liebertiana Engl. was collected in the Ngoye forest in Northern Kwazulu-Natal and a voucher specimen deposited in the Herbarium of the University of Zululand. The same plant was also collected in the Hangklip Indigenous Forest Reserve near Louis Trichardt and a voucher specimen (no. 07934) deposited in the H.G.W.J. Schweickerdt Herbarium in Pretoria. Voucher specimens collected from two *C. liebertiana* trees from Zimbabwe (near Harare) were deposited in the National Herbarium in Harare (no. 1906).

Isolation. Milled bark of *C. liebertiana* from the Ngoye forest (340 g) was extracted at RT with $CHCl_3$ to afford crude extract (3.9 g). Using the usual sepn procedures described previously [7], three compounds were isolated and characterized. These were known α-pyrones, cryptocaryalactone (42 mg) [8, 9], deacetylcryptocaryalactone (720 mg), [8, 9] and cryptofolione (300 mg) (**1a**) [5]. It was found that the latter compound previously described as an oil by us, crystallizes very slowly at 0° to afford a semi-crystalline solid, mp 40–50°, $[\alpha]_D^{25}$ +64° ($CHCl_3$, *c* 0.31).

The milled bark (600 g) from the Louis Trichardt source was successively extracted with $CHCl_3$ and Me_2CO to yield, respectively, 4 g and 5.2 g extract. Further purification (CC with $CHCl_3$–MeOH, 96:4 and Chromatotron hexane–EtOAc, 1:1 of both frs gave cryptofolione (**1a**) in good yield (410 mg) and the new compound (69 mg) (**6**).

(+)-6-(4′-*Hydroxy*-8′-*phenyloct*-1′,5′,7-*trienyl*)-5,6-*dihydro*-2*H*-*pyran*-2-*one* (**6**). Unstable oil $[\alpha]_D^{25}$ +56° ($CHCl_3$, *c* 0.50). ^{1}H NMR ($CDCl_3$, 200 MHz): δ 2.30-2.46 (4H, *m*, H-3′, H-5), 4.30 (1H, *q*, *J* = 6.3 Hz, H-4′), 4.90 (1H, *q*, *J* = 6.2 Hz, H-6), 5.66 (1H, *dt*, *J* = 6.2 Hz, H-1′), 5.80 (1H, *m*, *J* = 6.3, 15.1 Hz, H-5′) 5.92 (1H, *m*, *J* = 6.1 Hz, H-2′) 6.03 (1H, *dt*, *J* = 1.9, 9,9 Hz, H-3), 6.35 (1H, *ddd*, *J* = 1.08, 15.2 Hz, H-6′) 6.55 (1H, *d*, *J* = 15.5 Hz, H-8′) 6.76 (1H, *dd*, *J* = 15.6 Hz, H-7′) 6.86 (1H, *m*, *J* = 9.8 Hz, H-4), 7.30 (5H, *m*, Ar-H). ^{13}C NMR ($CDCl_3$, 50 MHz): δ 29.7 (C-5), 40.1 (C-3′), 71.5 (C-4′), 7.78 (C-6), 121.5 (C-3), 126.4 (Ar-C), 127.7 (Ar-C), 128.8 (C-7′), 128.6 (Ar-C), 130.1 (C-6′), 130.6 (C-1′), 131.0 (C-2′), 133.0 (C-8′), 135.4 (C-5′), 137.0 (Ar-C), 144.8 (C-4), 164.0 (C-2). MS *m/z* (rel. int): 296 $[M]^+$(4), 207 (5), 159 (100), 129 (24), 91 (90), 81 (16).

From the milled bark of *C. liebertiana* (immature specimen) (220 g), extract 3.4 was obtained after $CHCl_3$ extraction. Sepn by the procedures described

above gave cryptofolione (**1a**) (520 mg) and the new compound (**1b**) (42 mg), the acetyl derivative of cryptofolione.

(+)-6-(6′-*Acetoxy*-4′-*hydroxy*-8′-*phenyloct*-1′,7′-*dienyl*)-5,6-*dihydro*-2*H*-*pyran*-2-*one* (**1b**). It is an unstable oil $[\alpha]_D^{25}$ +86.1° ($CHCl_3$, *c* 0.42). ^1H NMR ($CDCl_3$, 200 MHz): δ 1.75 (2H, *m* 5′H), 2.13 (3H, *s*, OAc), 2.27 (2H, *m*, *J* = 6.5, 12.9 Hz, H-3′), 2.48 (2H, *m*, H-5), 2.92 (1H, *bs*, OH), 3.68 (1H, *m*, H-4′), 4.90 (1H, *m*, H-6), 5.69 (2H, *m*, H-1′, H-6′), 5.89 (1H, *m*, H-2′) 6.03 (1H, *dt*, *J* = 1.9, 9.9 Hz, H-3), 6.18 (1H, *dd*, *J* = 6.9, 15.9 Hz, H-7′), 6.62 (1H, *dd*, *J* = 15.9 Hz, H-8′), 6.87 (1H, *m*, H-4), 7.19-7.41 (Ar-H). ^{13}C NMR ($CDCl_3$, 50 MHz): δ 21.3 ($OCOCH_3$), 29.7 (C-5), 39.9 (C-3′), 42.5 (C-5′), 66.6 (C-4′), 71.9 (C-6′), 77.9 (C-6), 121.4 (C-3), 126.6 (Ar-C), 127.2 (C-7′), 128.1 (Ar-C), 129.6 (C-1′), 131.3 (C-2′), 132.2 (C-8′), 136.0 (Ar-C), 144.8 (C-4), 164.1(C-2), 171.5 ($OCOCH_3$).

Acknowledgements—The authors thank the University Research Committee and the Foundation for Research Development (FRD) for financial support. Mrs Carla Willis (Department of Forestry, Louis Trichardt) and Professor A. E. van Wyk (Pretoria University) kindly collected and identified the *C. liebertiana* sample from Louis Trichardt, and Mrs Anne Hutchings (Botany Department, University of Zululand) provided us with an authenticated sample of *C. liebertiana* from Ngoye Forest.

REFERENCES

1. Rohwer, J. G. (1993) in *The Families and Genera of Vascular Plants* (Kubizki, K., ed.), pp. 366–391. Springer, Berlin.
2. Van der Merwe, J. J. M., Van Wyk, A. E. and Kok, P. D. F. (1988) *S. Afr. J. Botany* **54**, 80.
3. Van der Merwe, J. J. M., Van Wyk, A. E. and Kok, P. D. F. (1990) *Grana* **29**, 185.
4. Drewes, S. E., Sehlapelo, B. M. and Sandor, P. (1993) *Phytochemistry* **32**, 1352.
5. Drewes, S. E., Sehlapelo, B. M. and Scott-Shaw, R. (1994) *Phytochemistry* **37**, 847.
6. Drewes, S. E., Sehlapelo, B. M., Horn M. M., Scott-Shaw, R. and Sandor, P. (1995) *Phytochemistry* **38**, 1427.
7. Drewes, S. E., Horn M. M. and Scott-Shaw, R. (1995) *Phytochemistry* **40**, 321.
8. Cunningham, A. B. (1988) in *An Investigation of the Herbal Medicine Trade in Natal/Kwazulu*, p. 70, Institute of Natural Resources No. 29, University of Natal, Pietermaritzburg.
9. Govindachari, T. R. and Partasarathy, P. C. (1971) *Tetrahedron Letters* **37**, 3401.
10. Spencer, G. F., England, R. E. and Wolf, R. B. (1984) *Phytochemistry* **23**, 2499.
11. Drewes, S. E., Horn, M. M. Sehlapelo, B. M., Ramesar, N., Field, J. S., Scott-Shaw, R. and Sandor, P. (1995) *Phytochemistry* **38**, 1505.