PERGAMON

Phytochemistry 51 (1999) 119–123

PHYTOCHEMISTRY

Prenylated flavonoids and a dihydro-4-phenylcoumarin from *Dorstenia poinsettifolia*

Bonaventure T. Ngadjui[a,*], Gilbert W.F. Kapche[a], Helene Tamboue[b], Berhanu M. Abegaz[c], Joseph D. Connolly[d]

[a]*Department of Organic Chemistry, University of Yaoundé 1, BP 812, Yaoundé, Cameroon*
[b]*Departement de Chimie, Ecole Normale Supérieure de Yaoundé, BP 47, Yaoundé, Cameroon*
[c]*Department of Chemistry, University of Botswana, P/Bag 0022, Gaborone, Botswana*
[d]*Department of Chemistry, University of Glasgow, Glasgow G12 8QQ, UK*

Received 11 June 1998

Abstract

The twigs of *Dorstenia poinsettifolia* yielded poinsettifolactone, a new dihydro-4-phenylcoumarin, and dorspoinsettifolin, a new 7,8-(2,2-dimethylpyrano)-4′-methoxyflavanone. Three known prenylated flavonoids, 4-hydroxylonchocarpin, 4-methoxylonchocarpin and 4′-hydroxyisolonchocarpin, were also isolated and identified. Structures were established by spectroscopic analysis and chemical correlations.

Keywords: Dorstenia poinsettifolia; Moraceae; Twigs; Prenylated flavonoids; Dihydro-4-phenylcoumarin; Dorspoinsettifolin; Poinsettifolactone

1. Introduction

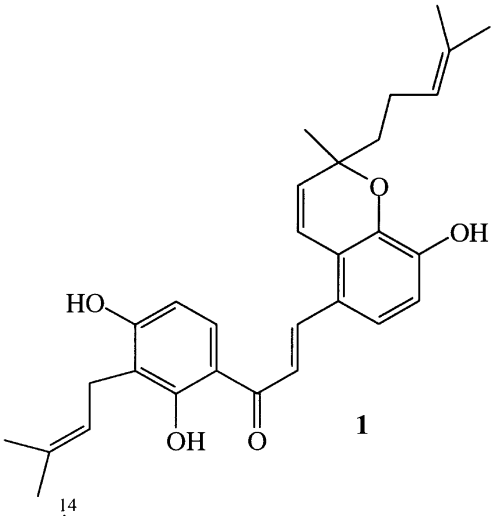
Dorstenia poinsettifolia Eng. is an undergrowth small herb indigenous to the rainforest zone in West Africa (Satabie, 1985). Its leaves are used for the treatment of yaws and infected wounds (Thomas, Thomas, Bromley, & Mbenkum, 1989). Previous phytochemical studies on this plant have led to the isolation and characterization of two new geranylated flavonoids poinsettifolins A (**1**) and B (**2**) (Ayafor, Sterner, Tsopmo, Kapnaing, & Ngnokam, 1997). As part of our program to study the chemical constituents of Cameroonian *Dorstenia* species (Abegaz, Ngadjui, Dongo, & Tamboue, 1998; Ngadjui, Abegaz, Dongo, Tamboue, & Kouam, 1998; Ngadjui, Dongo, Happi, Bezabih, & Abegaz, 1998; Ngadjui et al., submitted for publication), we have investigated the CH_2Cl_2/MeOH extract of the twigs of *D. poinsettifolia*. In the present paper, we describe the isolation and structural elucidation of a new prenylated flavonoid, dorspoinsettifolin (**3**), and a new dihydro-4-phenylcoumarin, poinsettifolactone (**4**), as well as the known 3′,4′-(2,2-dimethylpyrano)-2′,4-dihydroxychalcone (**5**) (Dagne, Bekele, & Waterman, 1989), 3′,4′-(2,2-dimethylpyrano)-2′-hydroxy-4-methoxychalcone (**6**) (Singhal, Barua, Sharma, & Baruah, 1983) and 7,8-(2,2-dimethylpyrano)-4′-hydroxyflavanone (**7**) (Talapatra, Mallik, & Talapatra, 1980; Dagne et al., 1989). Known compounds were identified from spectroscopic and physical data and comparison with authentic samples.

2. Results and discussion

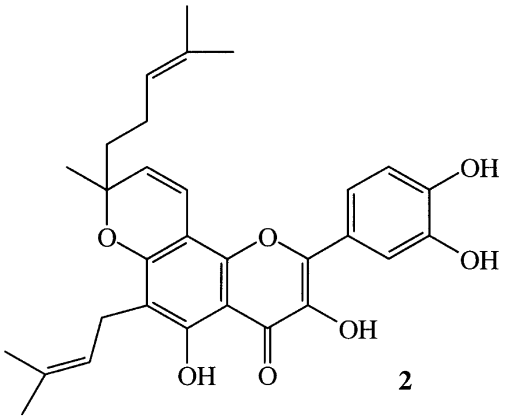
Dorspoinsettifolin (**3**) was obtained as yellow needles (m.p. 150–151°C). Its molecular formula was determined as $C_{21}H_{20}O_4$ from NMR and mass spectral data. It was clear from the NMR spectra that dorspoinsettifolin was a flavanone. Thus an oxymethine, a carbonyl group and a methylene at δ_c 79.5 (d), 190.9 (s) and 44.1 (t), respectively, and an ABX system (δ_H 2.81 (dd, $J=16.8$, 2.5 Hz), 3.02 (dd, $J=16.8$, 13.2 Hz) and 5.43 (dd, $J=13.2$, 2.5 Hz), typically assignable to 2H-3 and H-2 of a flavanone) could all be observed. The aromatic region of the ^{1}H NMR spectrum displayed eight proton resonance signals, two of which were assigned to the pyran group (see below). Four of the protons form an AA′BB′ system at δ 6.95 (2H, d, $J=8.5$ Hz) and 7.40 (2H, d, $J=8.5$ Hz) located in ring B. The remaining two proton signals form an AX system at δ 6.49 and 7.74 (both d, $J=8.7$ Hz) and were assigned to H-6 and H-5, respectively, because of the downfield chemical shift of the latter. The signals that could be assigned to the 2,2-dimethylpyran group were as follows: δ 1.44 and 1.46 (both 3H s, 2 × Me), δ 5.56 (d,

* Corresponding author.

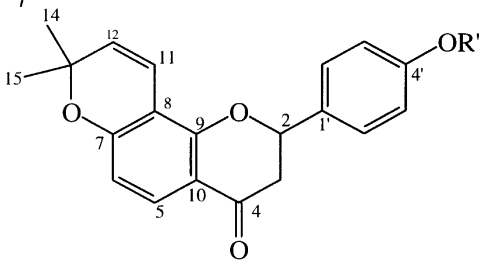
0031-9422/99/$ - see front matter
PII: S0031-9422(98)00621-9



1

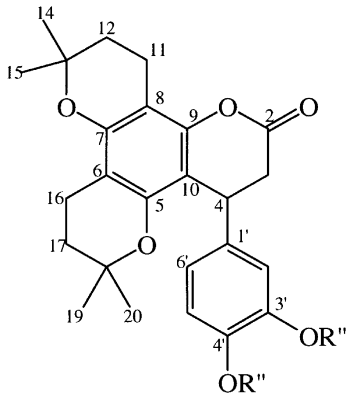


2



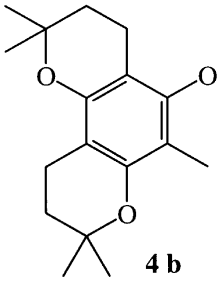
3 R' = Me

7 R' = H

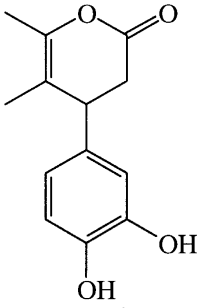


4 R" = H

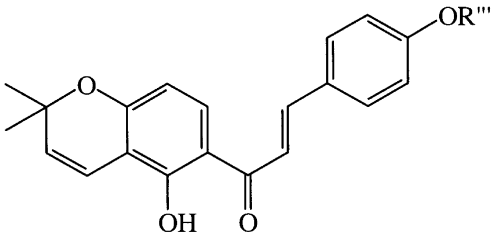
8 R" = OAc



4 b



4 a



5 R"' = H

6 R"' = Me

Structure 1.

Table 1
^{13}C NMR spectral data of **3** ($CDCl_3$) and **7** (CD_3COCD_3) at 90 MHz

Carbon	3	7
2	79.5 (d)	80.8 (d)
3	44.1 (t)	44.5 (t)
4	190.9 (s)	190.6 (s)
5	128.8 (d)	130.0 (d)
6	111.1 (d)	111.3 (d)
7	159.8 (s)[a]	159.9 (s)[c]
8	109.4 (s)	110.2 (s)
9	157.7 (s)[a]	158.5 (s)[c]
10	114.7 (s)	115.8 (s)
1′	131.0 (s)	131.1 (s)
2′	127.6 (d)	129.0 (d)
3′	114.1 (d)	116.0 (d)
4′	159.6 (s)[a]	158.6 (s)[c]
5′	114.1 (d)	116.0 (d)
6′	127.6 (d)	129.0 (d)
11	115.9 (d)	116.4 (d)
12	127.9 (d)	128.2 (d)
13	77.5 (s)	78.1 (s)
14	28.5 (q)[b]	28.4 (q)[b]
15	28.1 (q)[b]	28.1 (q)[b]
OMe	55.3 (q)	–

Signals with the same superscripts in the same column may be interchanged. Multiplicities were determined from DEPT spectra.

$J=10.0$ Hz) and δ 6.62 (d, $J=10.0$ Hz). The proton spectrum of **3** also showed a methoxyl group at δ 3.84. The foregoing data are consistent with structure **3**, for which the name dorspoinsettifolin is proposed. This structure was confirmed by the ^{13}C NMR data (Table 1) and chemical correlations. Thus cylisation of **6** in formic acid and methylation of **7** with diazomethane both readily gave dorspoinsettifolin (**3**). An isomeric compound, 6-methoxyisolonchocarpin, has been reported from *Lonchocarpus subglauscescens* (Leguminosae) (Magalhaes, Tozzi, Saes, & Magalhaes, 1996).

Poinsettifolactone (**4**) was isolated as an amorphous yellow powder (m.p. 211–213°C). Its molecular formula was determined as $C_{25}H_{28}O_6$ from NMR and EI mass spectral data. Its IR spectrum showed a strong carbonyl absorption at ν_{max} 1750 cm^{-1}, characteristic of a δ-lactone. The UV spectrum displayed absorption at λ_{max} 287 nm. $AlCl_3$/HCl-induced shifts in the UV spectrum indicated that compound **4** contains two *ortho* dihydroxyl groups (Mabry, Markham, & Thomas, 1970). This was confirmed in the ^{13}C NMR (δ 143.5 (s); 142.3 (s)) and in the ^1H NMR (δ 5.63 and 5.47 (both, brs) which disappeared on the acetylation of the compound) spectra. The aromatic region of the ^1H NMR spectrum of poinsettifolactone displayed three proton resonance signals which form an ABX system (a doublet at δ 6.76 ($J=8.2$ Hz), an *ortho*/*meta* coupled double doublet at δ 6.57 ($J=8.2$, 2.1 Hz) and a meta coupled signal at δ 6.68 ($J=2.1$ Hz)) located in a trisubstituted benzene ring. This ^1H NMR spectrum also showed a set of three-proton double doublets at δ 2.99 ($J=16.0$, 2.7 Hz), 2.95 ($J=16.0$, 6.3 Hz) and 4.42 ($J=6.3$, 2.7 Hz). Correlations in the HMBC spectrum Table 3 suggested the presence of partial structure **4a**. Furthermore, the ^1H NMR spectrum also showed proton resonance signals for two 2,2-dimethyldihydropyran groups (four benzylic proton signals at δ 2.66 (2H, m), 2.56 (2H, m) coupled to four methylene proton signals at δ 1.73 (t, $J=6.8$ Hz), 1.71 (t, $J=6.7$ Hz) and four methyl proton signals at δ 1.34, 1.30, 1.29 and 1.12 ppm). The ^{13}C NMR spectrum displayed five oxygenated aromatic carbon signals, two of which were assigned to the *ortho* dihydroxyl carbons. The remaining three are *meta* oriented in a fully substituted benzene ring because of their chemical shifts (δ 151.5, 149.0 and 147.7). The foregoing analysis is consistent with a second partial structure **4b** for poinsettifolactone. Analysis of the HMBC and HMQC spectra of this compound led to structure **4** for poinsettifolactone. The carbon resonances of **4** were assigned using DEPT, HMBC and HMQC. ^{13}C- and ^1H-NMR assignments are given in Table 2 and HMBC correlations are shown in Table 3.

Poinsettifolactone (**4**) is a member of a small group of dihydrocoumarins with a C-4 phenyl group. Other

Table 2
^{13}C NMR spectral data of **4** and **8** in $CDCl_3$ at 90 MHz

Carbon	4	8
2	169.8 (s)	168.2 (s)
3	36.7 (t)	35.7 (t)
4	33.5 (d)	33.9 (d)
5	147.8 (s)	147.7 (s)
6	105.8 (s)	105.8 (s)
7	149.0 (s)	149.0 (s)
8	100.7 (s)	100.8 (s)
9	151.5 (s)	151.8 (s)
10	104.7 (s)	103.9 (s)
1′	135.1 (s)	140.7 (s)
2′	113.9 (d)	122.1 (d)
3′	142.5 (s)	141.3 (s)
4′	143.5 (s)	141.8 (s)
5′	115.2 (d)	123.3 (d)
6′	119.2 (d)	124.9 (d)
11	16.5 (t)	16.8 (t)
12	31.9 (t)	32.0 (t)
13	74.4 (s)	74.6 (s)[b]
14	27.3 (q)[a]	27.3 (q)[c]
15	27.1 (q)[a]	27.0 (q)[c]
16	16.9 (t)	16.6 (t)
17	32.0 (t)	32.1 (t)
18	74.4 (s)	74.5 (s)[b]
19	26.6 (q)[a]	26.7 (q)[c]
20	25.9 (q)[a]	25.9 (q)[c]
CH_3CO		20.6 (q), 20.6 (q)
CH_3CO		168.2 (s) 168.0 (s)

Signals with the same superscripts in the same column may be interchanged. Multiplicities were determined from DEPT spectra.

Table 3
^1J (from HMQC) and ^2J, ^3J-gradient HMBC correlations for **4**

Proton	Position	^1J-correlated carbon	^3J, ^2J-correlated carbons
6.76	5′	115.2	119.2 (C-6′), 135.1 (C-1′), 143.5 (C-4′)
6.68	2′	113.9	119.2 (C-6′), 135.1 (C-1′), 142.3 (C-3′)
6.57	6′	119.2	135.1 (C-1′), 115.2 (C-5′), 143.5 (C-4′), 33.5 (C-4), 113.9 (C-2′)
4.42	4	33.5	119.2 (C-6′), 113.9 (C-2′)
2.99	3	36.7	104.7 (C-10), 135.1 (C-1′), 169.8 (C-2)
2.95	3	36.7	33.5 (C-4), 135.1 (C-1′), 169.8 (C-2)
2.66	11	16.5	31.9 (C-12), 149.0 (C-7), 100.6 (C-8), 74.4 (C-13, C-18)
2.56	16	16.9	32.0 (C-17), 149.0 (C-7), 147.8 (C-5), 105.8 (C-6), 74.4 (C-13, 18)
1.73	12	31.9	16.5 (C-11), 100.6 (C-8), 74.4 (C-13, C-18)
1.71	17	32.0	16.9 (C-16), 105.8 (C-6), 74.4 (C-13, C-18)

members of the series are calomelanols A-J, thwaitesic lactone and isothwaitesic lactone which were isolated, respectively, from *Pityrogramma calomelanos* (Polypodiaceae) (Asai, Iinuma, Tanaka, & Mizuno, 1991, 1992; Asai, Iinuma, Tanaka, Takenaka, & Mizuno, 1992) and *Calophyllum thwaitesii* (Guttiferae) (Samaraweera, Sotheeswaran, & Sultanbawa, 1983; Dharmaratne, Sotheeswaran, & Balasubramanian, 1984).

3. Experimental

3.1. General

M.p.'s uncorr.; UV-visible: MeOH solution; IR: KBr disk or $CHCl_3$ solution; EIMS: direct inlet, 70 eV; ^1H and ^{13}C-NMR ($CDCl_3$, CD_3COCD_3) 360 and 90 MHz, respectively, residual solvent peaks as internal references.

3.2. Plant material

Twigs of *D. poinsettifolia* were collected at Nkoljobe mountain, Yaounde, in the Central Province of Cameroon and a voucher specimen (No. 2138) is deposited at the National Herbarium.

3.3. Extraction, isolation and characterization

The air-dried and powdered plant material (1.5 kg) was extracted exhaustively with cold mixture of CH_2Cl_2–MeOH (1:1), MeOH and water. Removal of the solvent from the combined organic extracts under reduced pressure gave 100 g of residue. This residue was subjected to partition extraction with chloroform and ethyl acetate successively. The chloroform and ethyl acetate soluble fractions, after concentration in vacuo, yielded two dark green residues (50 and 15 g, respectively). The rest of the MeOH extract (30 g) was found to contain mostly tannins and was not investigated further. The $CHCl_3$ and EtOAc extracts were monitored by TLC plates and combined. Part of this combined extract (50 g) was subjected to repeated chromatographic separations on Silica gel and Sephadex LH20 columns and preparative TLC, eluting with CH_2Cl_2, CH_2Cl_2–MeOH mixtures and MeOH. Frs eluted with CH_2Cl_2 yielded 4-methoxylonchocarpin (30 mg) and 4-hydroxylonchocarpin (60 mg). 4′-Hydroxyisoloncocarpin (15 mg) and dorspoinsettifolin (10 mg) were obtained from fractions eluted with CH_2Cl_2–MeOH, 19:1. Poinsettifolactone (30 mg) was isolated from frs eluted with CH_2Cl_2–MeOH, 17:3. Known compounds were identified by comparison (m.p., ^1H, ^{13}C-NMR) with authentic specimens.

3.3.1. 7,8-(2,2-Dimethylpyrano)-4′-methoxyflavanone (dorspoinsettifolin) (**3**)

Yellow needles, m.p. 150–151°C, $[\alpha]_D$ −15 (MeOH, *c* 0.05); UV λ_{max}^{MeOH} nm log(ε): 231 (3.84), 253 (3.91), 265 (3.94) and 309 (3.34). $\lambda_{max}^{MeOH+AlCl_3}$ nm log(ε): no change. IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3010, 1675 (C═O), 1595, 1576, 1515, 1440, 1380, 1335, 1180, 1115, 1070. EIMS *m/z* (ref. int.): 336 ($[M]^+$, 25), 321 ($[M–Me]^+$, 48), 187 ($[M–Me–C_9H_{10}O]^+$, 100), 159 (5), 134 (13), 119 (10), 91 (10). ^1H-NMR (360 MHz, $CDCl_3$) δ: 1.44, 1.46 (3H, each s, Me), 2.81 (1H, dd, J=16.8, 2.5 Hz, H-3a), 3.02 (1H, dd, J=16.8, 13.2 Hz, H-3b), 3.84 (3H, s, OMe), 5.43 (1H, dd, J=13.2, 2.5 Hz, H-2), 5.56 (1H, d, J=10 Hz, H-12), 6.49 (1H, d, J=8.7 Hz, H-6), 6.62 (1H, d, J=10 Hz), H-11), 6.95 (2H, d, J=8.5 Hz, H-3′,5′), 7.40 (2H, d, J=8.5 Hz, H-2′,6′) and 7.74 (1H, d, J=8.7 Hz, H-5). ^{13}C-NMR (90 MHz, $CDCl_3$): Table 1.

3.3.2. Cyclisation of 4-methoxylonchocarpin (**6**)

4-Methoxylonchocarpin, **6** (20 mg), in formic acid (5 ml) was heated under reflux for 24 h. After cooling the reaction mixture was extracted with 3 × 20 ml of $CHCl_3$. The $CHCl_3$ extract, after concentation in vacuo, was purified by prep. TLC to give dorspoinsettifolin (**3**) (15 mg, 75%) identical (m.p., NMR, IR) with the natural compound.

3.3.3. Methylation of 4′-hydroxyisolonchocarpin (7)

CH_2N_2 was added to **7** (5 mg) dissolved in EE. After 30 min the reaction mixture was evaporated to give **3** (m.p. TLC).

3.3.4. Poinsettifolactone (4)

Amorphous yellow powder m.p. 211–213°; $[\alpha]_D^{20}$ +33 ($CHCl_3$, *c* 0.04) UV λ_{max}^{MeOH} nm log(ε): 287 (4.35) $\lambda_{max}^{MeOH+AlCl_3}$ nm log(ε): 295 (4.46), $\lambda_{max}^{MeOH+AlCl+HCl}$ nm log(ε): 287 (4.28). IR ν_{max}^{KBr} cm^{-1}: 3410–3440 (OH), 2970, 2930, 1750 (δ-lactone), 1620, 1600, 1520, 1440, 1160, 1120, 960. EIMS *m/z* (ref. int.): 424 ($[M]^+$, 92), 409 ($[M–Me]^+$, 10), 382 (10), 370 ($[M-55+H]^+$, 40), 369 ($[M-55]^+$, 100), 368 (30), 340 (20), 313 (35), 282 (10), 271 (20), 203 (25), 136 (15) and 55 (10). ^1H NMR (360 MHz, $CDCl_3$) δ: 1.12, 1.29, 1.30, 1.34 (3H each, s, Me), 1.71 (2H, t, *J*=6.7 Hz, 2H-17), 1.73 (2H, t, *J*=6.8 Hz, 2H-12), 2.56 (2H, m, 2H-16), 2.66 (2H, m, 2H-11), 2.95 (1H, dd, *J*=16.0, 6.3 Hz, H-3a), 2.99 (1H, dd, *J*=16.0, 2.7 Hz, H-3b), 4.42 (1H, dd, *J*=6.3, 2.7 Hz, H-4), 5.47 (1H, brs, OH), 5.63 (1H, brs, OH), 6.57 (1H, dd, *J*=8.2, 2.1 Hz, H-6′), 6.68 (1H, d, *J*=2.1 Hz, H-2′) and 6.76 (1H, d, *J*=8.2 Hz, H-5′). ^{13}C NMR (90 MHz, $CDCl_3$): Table 2.

3.3.5. Acetylation of poinsettifolactone (4)

4 (15 mg) in CH_2Cl_2 (2 ml) was treated with acetic anhydride (2 ml) in the presence of a catalytic amount of DMAP for 1 h. The reaction was monitored by TLC. The reaction mixture was mixed with celite (4 g) and evaporated into dryness in vacuo and the powder obtained was introduced onto a silica gel column and eluted with hexane/EtOAc (7:3) to give the diacetate **8** (13 mg, 72%): white powder in MeOH m.p. 159–160°C. UV λ_{max}^{MeOH} nm log(ε): 286 (4.25). $\lambda_{max}^{MeOH+AlCl_3}$ nm log(ε): no change. IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 1765 (C=O), 1625, 1600, 1510, 1450, 1370, 1320, 1260, 1160, 1120, 960. EIMS *m/z* (ref. int.): 508 ($[M]^+$, 100), 466 (10), 453 ($[M-55]^+$, 95), 452 (15), 411 (35), 398 (8), 355 (25), 311 (20), 284 (15), 271 (10), 203 (15). ^1H NMR (360 MHz, $CDCl_3$) δ: 1.14, 1.30, 1.31, 1.32 (3H each, s, Me), 1.71 (2H, brt, *J*=6.9 Hz, 2H-17), 1.75 (2H, brdt, *J*=7.0, 2.0, 2H-12), 2.54 (2H, m, 2H-16), 2.71 (2H, m, 2H-11), 2.98 (1H, dd, *J*=16.2, 7.1 Hz, H-3a), 3.05 (1H, dd, *J*=16.2, 2.2 Hz, H-3b), 4.51 (1H, dd, *J*=7.1, 2.2 Hz, H-4), 7.00 (1H, dd, *J*=8.5, 2.0 Hz, H-6′), 7.06 (1H, d, *J*=2.0 Hz, H-2′) and 7.08 (1H, d, *J*=8.5 Hz, H-5′): ^{13}C NMR (90 MHz, $CDCl_3$): Table 2.

Acknowledgements

B.T.N. acknowledges the Royal Society of London for a three-month travel grant to the Department of Chemistry of the University of Glasgow and the IPICS (Uppsala) under the auspices of NABSA. We are grateful to Dr. E. Dongo and Dr. D.S. Rycroft for helpful discussions.

References

Abegaz, B. M., Ngadjui, B. T., Dongo, E., & Tamboue, H. (1998). *Phytochemistry*, in press.

Asai, F., Iinuma, M., Tanaka, T., & Mizuno, M. (1991). *Phytochemistry*, *30*, 3091.

Asai, F., Iinuma, M., Tanaka, T., & Mizuno, M. (1992). *Heterocycles*, *33*, 229.

Asai, F., Iinuma, M., Tanaka, T., Takenaka, M., & Mizuno, M. (1992). *Phytochemistry*, *31*, 2487.

Ayafor, J. F., Sterner, O., Tsopmo, A., Kapnaing, P., & Ngnokam, D. (1997). *7th NAPRECA Symposium on Natural Products*, Tanzania (p. 52).

Dagne, E., Bekele, A., & Waterman, P. G. (1989). *Phytochemistry*, *28*, 1897.

Dharmaratne, H. R. W., Sotheeswaran, S., & Balasubramanian, S. (1984). *Phytochemistry*, *23*, 2601.

Mabry, T. J., Markham, K. R., & Thomas, M. B. (1970). *The systematic identification of flavonoids* (p. 165). New York: Springer Verlag, 1970.

Magalhaes, A. F., Tozzi, A. M. G. A., Saes, B. H. L. N., & Magalhaes, E. G. (1996). *Phytochemistry*, *42*, 1459.

Ngadjui, B. T., Abegaz, B. M., Dongo, E., Tamboue, H., & Kouam, F. (1998). *Phytochemistry*, *48*, 349.

Ngadjui, B. T., Abegaz, B. M., Tabopda, T. K., Dongo, E., Kapche, G. W. F., & Sandor, P. *Phytochemistry*, submitted for publication.

Ngadjui, B. T., Dongo, E., Happi, E. N., Bezabih, M.-T., & Abegaz, B. M. (1998). *Phytochemistry*, *48*, 733.

Satabie, B. (1985). In Mesres (Ed.), *Flore du Cameroun Moraceae-Cecropiaceae* (p. 74). ISSN.

Samaraweera, U., Sotheeswaran, S., & Sultanbawa, M. U. S. (1983). *J. Chem. Perkin Trans.*, *1*, 703.

Singhal, A. K., Barua, N. C., Sharma, R. P., & Baruah, J. N. (1983). *Phytochemistry*, *22*, 1005.

Talapatra, S. K., Mallik, A. K., & Talapatra, B. (1980). *Phytochemistry*, *19*, 1199.

Thomas, D. W., Thomas, J. M., Bromley, W. A., & Mbenkum, F. T. (1989). *Korup Ethnobotany Survey, Final Report*. Surrey: W.W.F.