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Limonoids from *Astrotrichilia voamatata*

Dulcie A. Mulholland[a,*], Sianne L. Schwikkard[a], Milijaona Randrianarivelojosia[b]

[a]*Natural Products Research Group, Department of Chemistry, University of Natal, Durban 4041, South Africa*
[b]*Laboratory of Pharmacology, EES Sciences, University of Antananarivo, BP 906, Antananarivo 101, Madagascar*

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Abstract

The stem bark of *Astrotrichilia voamatata* (Meliaceae) yielded the novel limonoids voamatin A and voamatin B. The stem bark of *Cipadessa boivinina* (Meliaceae) yielded sitosterol and stigmast-4-en-3-one, but no limonoids.

Keywords: Meliaceae; *Astrotrichilia voamatata*; *Cipadessa boivinina*; Limonoids; Voamatin A; Voamatin B

1. Introduction

In continuation of our investigations into the chemistry of the Meliaceae of Madagascar, the stem bark of two species, *Astrotrichilia voamatata* Leroy, collected from Feverive East, Madagascar (008-MJ-M.Dul) and *Cipadessa boivinina* Baillon (003-MJ-M.Dul) collected from Ankarafantsika in the northwest of Madagascar was investigated. Both were collected by M.R. and identified by comparison against authentic specimens at the Parc de Botanique et de Zoologie de Tsimbazaza. Voucher specimens are deposited at the University of Antananarivo.

No limonoids have been reported previously from the *Cipadessa* genus. *Cipadessa fruticosa* has been reported to contain clerodane and labdane diterpenoids (Rojatkar, Chiplunkar & Nagasampagi, 1994; Rojatkar & Nagasampagi, 1994) and *Cipadessa cinerascens* has yielded flavonoid glycosides (Liang, Zhong & Xiao, 1991, 1994). *Astrotrichilia asterotricha*, also from Madagascar, has yielded dammaranes (Mulholland, Nair & Taylor, 1994) and the complex limonoid, astrotrichilin (Mulholland, Nair & Taylor, 1996). In this investigation, the stem bark of *C. boivinina* yielded the common phytosterols, stigmasterol and stigmast-4-en-3-one, but, in agreement with previous investigations of *Cipadessa* species, no limonoids. *A. voamatata* yielded the limonoids, vomatins A and B which have not been reported previously. These limonoids are related to those isolated previously from *Ekebergia pterophylla* (Kehrli, Taylor & Niven, 1990), *Ekebergia capensis* (Mulholland & Iourine, 1998) and *Heynea trijuga* (Purushothaman, Venkatanarasimhan, Sarada, Connolly & Rycroft, 1987).

2. Results and discussion

The NMR spectra of voamatins A and B were very similar and, on acetylation (Ac_2O/py), they yielded the same compound, voamatin C. The HRMS indicated identical molecular formulae of $C_{36}H_{46}O_{11}$ for voamatins A and B. Peaks at m/z 517 $[M\text{-}131]^+$ and m/z 500 $[M\text{-}148]^+$ indicated the presence of a cinnamate ester in both compounds.

The ^1H NMR spectrum of voamatin A indicated that it was a limonoid of the ekebergolactone type. Resonances ascribable to protons in the β-substituted furanyl ring occurred at δ 7.58(H-23), δ 7.46 (H-21) and δ 6.40 (H-22). Ring D was oxidised to a C-16 lactone, with H-17 occurring as a sharp singlet at δ 6.35 and 2H-15 occurring as a pair of doublets at δ 2.69

* Corresponding author. Tel.: +27-31-260-3091; fax: +27-31-260-3091.

E-mail address: mulholld@scifs1.und.ac.za (D.A. Mulholland)

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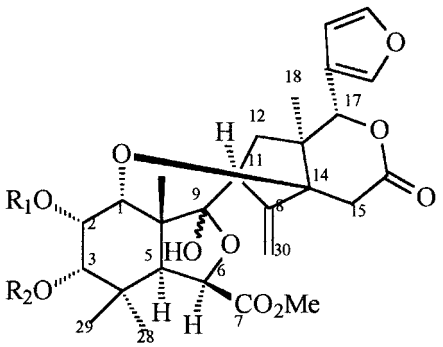
and δ 2.85 (J 17.5Hz) which were not further coupled. The COSY spectrum showed coupled resonances at δ 3.70, δ 4.19 and δ 5.09 ascribable to H-1, H-2 and H-3 respectively. On acetylation, these resonances were shifted to δ 3.80. δ 5.35 and δ 5.09 respectively. Thus a hydroxy group was placed at C-2 and an ester at C-3 in voamatin A. The ^{1}H NMR spectrum confirmed that the ester present was a *trans*-cinnamate. Ring B was opened to give a carbomethoxy group at C-7 (δ 175.5) and a 8,30-double bond, C-8 and C-30 occurring at δ 145.0 and δ 114.0 respectively in the ^{13}C NMR spectrum. The nonequivalent H-30 protons occurred as singlets at δ 5.40 and δ 4.90 in the ^{1}H NMR spectrum. Doublets ascribable to H-5 and H-6 occurred at δ 2.95 and δ 4.65 and C-5 and C-6 as methine resonances at δ 50.72 and δ 76.09 respectively, indicating substitution at C-6. A double-doublet at δ 3.29 was assigned to H-11 and was coupled to two H-12 protons and allylically coupled to one of the H-30 protons. The absence of coupling to H-9 suggested a contracted ring C. A fully substituted carbon resonance at δ 109.5 was assigned to C-9 and indicated the presence of a hemiketal carbon atom.

The molecular formula indicated that two more rings were necessary so a 1,14-oxide bridge and a 6,9-oxide to form a hemiketal ring was proposed. This gave a structure with the correct molecular formula. The stereochemistry at C-1, 2, 3 and 6 was established by NOE difference experiments. Irradiation of H-5α gave a positive NOE for H-6, indicating it was also α and also the 3H-28 resonance. Irradiation of the H-3 resonance gave a positive NOE for the 3H-29 resonance and the H-2 resonance. As the methyl group at C-29 is β, H-3 and H-2 were also assigned the β configuration. Irradiation of H-2β gave positive NOEs for H-1 and H-3. This confirmed that the substitution in ring A was 1α, 2α, 3α agreeing with X-ray results found previously for *Ekebergia* compounds (Kehrli et al., 1990). Irradiation of the 3H-18 signal caused enhancement of the H-12α signal at δ 1.69. Irradiation of this signal gave a positive enhancement for H-11, establishing H-11 as α. Thus structure **1** was assigned to voamatin A.

The ^{1}H NMR data for voamatin B were very similar to those of voamatin A and the ^{13}C NMR spectra were almost identical. On acetylation (Ac_2O/py) the identical product was produced as on acetylation of voamatin A. Under the acetylating conditions equilibration of the hemiketal occurred leading to the more stable β epimer. A Dreiding model shows that both hemiketals are indeed possible. A previous report of a related compound, trijugin A from *Heynea trijuga,* only reports the presence of one isomer (Purushothaman et al., 1987). A NOE experiment in which the irradiation of the 3H-19 resonance caused an enhancement of the C-9 hydroxy group signal enabled these researchers to establish that the hydroxy group was β. Unfortunately, in the case of the voamatins, the hydroxy group was superimposed on other signals and attempts at irradiating the signal were unsuccessful. The most notable difference in the ^{1}H NMR spectra of voamatins A and B were the H-30 resonances. These occurred at δ 5.40 and 4.90 in voamatin A and at δ 5.50 and 5.10 in voamatin B. These resonances occurred at δ 5.50 and 5.13 in trijugin A, suggesting that voamatin B and trijugin A both have a β-hydroxy group at C-9. It is likely that the stereochemistry of the common acetate is the same as that of trijugin A, being the most stable form, and this is supported by the chemical shifts of the two H-30 protons which occur at δ 5.47 and 5.05.



		R_1	R_2
1	9α-OH	H	Cinnamate
2	9β-OH	H	Cinnamate
3	9β-OH	Ac	Cinnamate

3. Experimental

Dried milled bark of *Cipadessa boivinina* (1 kg) was extracted successively with hexane, methylene chloride and methanol in a soxhlet apparatus. The hexane extract (10.73 g) yielded, after column chromatography using silica gel (Merck 9385), the known compounds stigmast-4-en-3-one and stigmasterol. NMR spectra of the crude methylene chloride and methanol extracts showed no limonoids were present so these extracts were not investigated further.

Bark (1 kg) of *Astrotrichilia voamatata* was extracted in the same way as described above. A white crystalline mixture of voamatins A and B precipitated out of the methylene chloride extract and the compounds could be separated by their slightly different solubilities.

Table 1
^{1}H NMR Data for **1**, **2** and **3** (300 MHz, $CDCl_3$, *J* in brackets)

Proton	**1**	**2**	**3**
1β	3.70 *d* (3.67)	3.80 *bs*	3.80 *bs*
2β	4.19 *m*	4.12 *bs*	5.35 *m*
3β	5.09 *d* (3.66)	5.10 *d* (3.66)	5.09 *bs*
5α	2.95 *d* (7.88)	2.95 *d* (7.88)	3.09 *d* (7.88)
6α	4.65 *d* (7.88)	4.65 *d* (7.88)	4.79 *d* (7.88)
11α	3.29 *dd* (5.17,9.69)	3.35 *dd* (5.35,10.0)	3.29 *dd*
12α	1.69 *dd* (9.69,13.36)	1.72 *dd* (13.45,10.00)	1.71 *m*
12β	2.15 *dd* (13.36,5.17)	2.15 *dd* (13.45,5.35)	2.18 *m*
15α	2.85 *d* (17.5)	2.89 *d* (17.5)	2.63 *s*
15β	2.69 *d* (17.5)	2.75 *d* (17.5)	2.63 *s*
17β	6.35 *s*	6.35 *s*	6.34 *s*
18	0.70 *s* (3H)	0.78 *s* (3H)	0.73 *s* (3H)
19	1.10 *s* (3H)	1.12 *s* (3H)	1.16 *s* (3H)
21	7.46 *s*	7.50 *s*	7.49 *s*
22	6.40 *s*	6.40 *s*	6.39 *s*
23	7.58 *s*	7.53 *s*	7.49 *s*
28	1.13 *s* (3H)	1.15 *s* (3H)	1.16 *s* (3H)
29	1.00 *s* (3H)	1.02 *s* (3H)	1.06 *s* (3H)
30_A	5.40 *s*	5.50 *s*	5.47 *s*
30_B	4.90 *s*	5.10 *s*	5.05 *s*
OCH_3	3.76 *s* (3H)	3.80 *s* (3H)	3.80 (3H)
H-2′	7.63 *d* (15.85)	7.68 *d* (15.85)	7.64 *d* (15.85)
H-3′	6.28 *d* (15.85)	6.28 *d* (15.85)	6.28 *d* (15.85)
Phenyl	7.05–7.30 *m* (5H)	7.10–7.30 *m* (5H)	7.10–7.30 *m* (5H)

NMR spectra were recorded in $CDCl_3$ on a Varian 300 MHz spectrometer. ^{1}H and ^{1}C NMR data of voamatins A, B and their acetate are given in Table 1 and 2. HRMS and EIMS were recorded at the Cape Technikon on Kratos HRMS 9/50 and Finnigan 1020 GC MS instruments. IR spectra were recorded on a Nicolet Impact 400D instrument.

Voamatin A, 12-deacetoxy-3-deoxy-3α-cinnamoyloxy-2α,9α-dihydroxytrijugin (**1**). (100 mg), colourless crystals, mp 261°C, HRMS M^+ at *m*/*z* 648.2551 ($C_{36}H_{40}O_{11}$ requires 648.2566), EIMS *m*/*z* 648 [M^+], 517, 500, 257, 231, 161, 131.

IR υ_{max} (NaCl)(cm^{-1}):3540, 2950, 1745, 1700, 1625, 1460, 1435, 1300, 1150, 1075. $[\alpha]_D = -12.30$(*c* = 0.610 g/100 cm^{-3}, $CHCl_3$).

Voamatin B, 12-deacetoxy-3-deoxy-3α-cinnamoyloxy-2α, 9β-dihydroxytrijugin (**2**). (80 mg) colourless crystals, mp 239, HRMS M^+ at *m*/*z* 648.2557 ($C_{36}H_{40}O_{11}$ requires 648.2566), EIMS *m*/*z* 648, 517, 500, 257, 230, 161.

Table 2
^{13}C NMR Data for Voamatins A, **1**, and B, **2** (75 MHz, $CDCl_3$)

Carbon	**1**	**2**	Carbon	**1**	**2**
1	76.92	76.90	18	16.98	17.05
2	66.13	66.13	19	17.81	17.82
3	78.85	78.84	20	122.31	122.26
4	36.61	36.62	21	143.80	143.83
5	50.72	50.73	22	108.15	108.15
6	76.09	76.11	23	140.08	140.02
7	175.47	175.47	28	26.81	26.83
8	145.09	145.06	29	21.26	21.27
9	109.52	109.54	30	113.97	114.06
10	53.57	53.57	1′	166.96	166.93
11	51.96	51.96	2′	146.92	147.24
12	36.61	36.62	3′	116.33	116.07
13	45.50	45.45	Phenyl	128.65(2C)	129.65(2C)
14	87.66	87.75		128.13(2C)	128.15(2C)
15	33.56	33.58		133.86	133.81
16	168.96	168.87		130.51	130.55
17	80.03	80.18	OMe	52.56	52.58

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