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TRITERPENES FROM *PALIURUS HEMSLEYANUS*

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Key Word Index—*Paliurus hemsleyanus*; Rhamnaceae; zizyberenalic acid; colubrinic acid; ceanothanolic acid; 2-*O*-(*p*-coumaroyl)-ceanothanolic acid; euscaphic acid methyl ester.

Abstract—Separation of the water-insoluble fraction of the ethanol extract from the root of *Paliurus hemsleyanus* led to the isolation of three new triterpenes. They are ceanothic acid derivatives named zizyberenalic acid, ceanothanolic acid and 2-*O*-(*p*-coumaroyl)-ceanothanolic acid. Additional NMR data for the known colubrinic acid and euscaphic acid, the latter being characterized as methyl ester, were also assigned. © 1997 Elsevier Science Ltd

INTRODUCTION

Paliurus hemsleyanus Rehd. is a deciduous tree, widely distributed in southern China [1]. The chemical constituents of the related species *P. ramosissimus* have been studied and several triterpenes of ceanothane and lupane types such as ceanothic acid and betulinic acid have been isolated [2, 3]. In a continuing study on the chemotaxonomy of the rhamnaceous plants, we investigated for the first time the chemical ingredients of *P. hemsleyanus*, the sole species of *Paliurus* plant growing in mainland China. This paper describes the isolation and the structural determination of three new triterpenes and NMR data of two known triterpenes from the water-insoluble fraction of the roots.

RESULTS AND DISCUSSION

The water-insoluble non-alkaloidal fraction of the ethanol extract of the roots was divided into hexane, chloroform and methanol soluble fractions. The chloroform fraction was chromatographed on silica gel and the compounds separated were recrystallized from methanol to give three new compounds **2**–**4** in addition to stigmasterol (containing a minor amount of sitosterol), the 3-*O*-*β*-glucosides of both sterols, betulin, betulinic acid, ceanothic acid, colubrinic acid (**1**) and euscaphic acid (**5**). Among these, compound **5** was separated as its *O*-methylated derivative **5a**, obtained by treating **5** with diazomethane.

Compounds **1**–**4** are ceanothic acid derivatives. All of them contain a 19-isopropylidene group (IR *ca* 1642, 881 cm^{-1}; ^1H NMR δ *ca* 4.71 *br s*; 4.89 *br s*; 1.75 *s*, Me), and a 17-carboxylic acid function (δ_C *ca* 179.0). The ^1H and ^{13}C NMR spectral data (Tables 1 and 2) revealed their structural differences in ring A. We describe below the details of their structural elucidation.

Compound **1** had a molecular formula $C_{30}H_{46}O_4$, deduced from the HR EI mass spectrum. It contained an aldehyde group (IR 2743 cm^{-1}; δ_H 10.11, *d*, J = 4.4 Hz; δ_C 206.1 *d*). The COSY spectrum revealed the coupling of the aldehydic proton to a methine proton at δ 2.58 (*dd*, J = 4.4 and 8.8 Hz) which also coupled to H-3 (δ 4.66, *d*, J = 8.8 Hz). These data together with comparison of its ^{13}C NMR data to those of ceanothic acid [3] suggested **1** to be colubrinic acid or its isomer. NOE studies displayed the enhancement of aldehydic proton upon irradiation at Me-10 (H-25) singlet, confirming 1*β*-CHO orientation and **1** to be colubrinic acid [4–6].

Compound **2**, mp 214–216°, had a molecular formula $C_{30}H_{44}O_3$, deduced from HR EI mass spectrum. It contained an *α*,*β*-unsaturated aldehyde function (UV λ_{max} 239; IR 2728, 1686 cm^{-1}; ^1H NMR δ 9.82 *s*, H-2; 6.49 *s*, H-3; ^{13}C NMR δ 191.6 *d*, C-2; 157.0 *s*, C-1; 163.5 *d*, C-3). These data suggested **2** to be a 1,3-didehydro derivative of ceanothic acid [9]. The NOE experiments which enhanced H-2 and H-24 (δ 0.86, *s*) upon irradiation of H-3, and enhanced H-5 (δ 1.50, *m*) and H-24 upon irradiation of H-23 (δ 1.06, *s*) also proved the partial structure in ring A. The rest of the structure is the same as that of ceanothic acid, according to a comparison of their ^1H NMR and ^{13}C NMR data. Accordingly, compound **2** is zizyberenalic acid [6]. This compound has been prepared from dehy-

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Table 1. Distinct ^{1}H NMR data (δ in ppm/J in Hz)* of compounds **1**–**4** in pyridine-d_5

Position	1	2	3	4†
1		2.53 *dd* (4.4, 8.8)	1.94 *dt* (4.6, 8.6)	2.00 *m*
2	9.86 *s*	10.11 *d* (4.4)	4.36 *dd* (4.6, 10.0)	4.78 *br s*
			4.06 *dd* (8.6, 10.0)	
3	6.56 *s*	4.66 *d* (8.8)	4.15 *d* (8.6)	4.06 *d* (8.7)
13	2.70 *dt* (3.5, 12.5)	2.64 *dt* (3.5, 12.7)	2.68 *dt* (3.5, 12.1)	2.67 *m*
16	2.61 *br d* (12.6) (β)	2.60 *m* (β)	2.60 *br d* (12.6) (β)	2.60 *br d* (12.0) (β)
18	1.71 *m*	1.71 *t* (12.0)	1.69 *t* (11.4)	1.69 *t* (11.4)
19	3.47 *dt* (3.9, 11.0)	3.43 *dt* (3.9, 11.0)	3.48 *dt* (3.9, 11.0)	3.48 *dt* (3.9, 11.0)
23	1.09 *s*	1.23 *s*	1.23 *s*	1.25 *s*
24	0.88 *s*	1.07 *s*	0.97 *s*	0.97 *s*
25	1.11 *s*	1.08 *s*	0.79 *s*	0.85 *s*
26	1.07 *s*	1.04 *s*	1.01 *s*	1.03 *s*
27	0.99 *s*	1.07 *s*	1.01 *s*	1.03 *s*
29	1.77 *s*	1.80 *s*	1.78 *s*	1.75 *s*
30	4.89 *br s* (*E*)	4.92 *br s* (*E*)	4.91 *br s* (*E*)	4.88 *br s* (*E*)
	4.74 *br s* (*Z*)	4.77 *br s* (*Z*)	4.76 *br s* (*Z*)	4.68 *br s* (*Z*)

* The rest of proton signals were picked up from HSQC, HMQC or COSY-45 spectra and their chemical shifts appeared roughly as follows: **1** H-5 1.50, H-6 1.40, H-7 1.37, H-9 1.97 *dd* (2.8, 12.7); **2**–**4** H-5 ~ H-7 1.40, H-9 1.80; **1**–**4** H-11 1.40 and 1.71, H-12 1.24 (α) and 1.92 (β), H-15 1.86 and 1.18, H-16 2.61 and 1.56, H-21 2.21 and 1.50, H-22 2.21 and 1.56.

† $\delta_{H\text{-}2',6'}$ 7.55 (AA′), $\delta_{H\text{-}3',5'}$ 7.22 (XX′) J_{AX} = 8.6 Hz; $\delta_{H\text{-}7'}$ 7.93 *d*, $\delta_{H\text{-}8'}$ 6.62 *d*, $J_{H\text{-}7',8'}$ = 15.9 Hz.

dration of colubrinic acid (zizyberenalic acid) upon thermolysis [6], but is isolated now for the first time from a natural source.

Compound **3**, mp 286°, had a molecular formula $C_{30}H_{48}O_4$, deduced from the HR EI mass spectrum, i.e. two hydrogens more than in **1**. It contained a primary alcohol function (^{1}H NMR δ_H 4.06 *dd*; 4.36 *dd*; ^{13}C NMR δ_C 64.6 *t*, C-2), identified by a COSY spectrum revealing the coupling of H-1 (δ 1.95, *m*) to the two methylene protons, and H-3 (δ 4.15, *d*, J = 8.6 Hz). These data suggested a 1-CH_2OH substitution and thereby **3** might be simply a 2-dihydro analogue of colubrinic acid. A NOE study causing the enhancement of H-3, H-24 (δ 0.97, *s*) and H-26 (δ 1.02, *s*) upon irradiation at H-25 (δ 0.79) assured H-3β. No NOE is observed between H-25 and H-1, and the coupling constant (8.6 Hz) of H-1 and H-3β is consistent with that of colubrinic acid, thus located H-1α. These results established the structure **3**. To our knowledge, compound **3** is a novel natural product and we name it ceanothanolic acid.

The ^{1}H NMR and ^{13}C NMR data of **1**–**3** were assigned by analysis of NOED, 2D NMR spectra (COSY-45, HMQC, HSQC and HMBC). Among these, the close carbon signals of C-8 (δ 42.4, *s*, **3**) and C-14 (δ 43.2, *s*, **3**), both of which are coupled to H-26 and H-27, were distinguished by the observation of the three-bond coupling of C-14 to H-16β (δ 2.60, *br d*) in the HMBC spectrum of **3**.

Compound **4**, mp > 295°, had a molecular formula $C_{39}H_{54}O_6$, deduced from the FAB mass spectrum and ^{13}C NMR data. It contained a *trans-p*-coumaroyl moiety, as exemplified by the ^{1}H NMR spectrum with an AA′XX′ system at δ 7.55 (2H) and 7.22 (2H), J_{AX} = 8.6 Hz, and an AX system at δ 7.93 and 6.62, J_{trans} = 15.9 Hz; IR absorption at 1680 and 829 cm^{-1} for conjugated ester and *para*-disubstituted phenyl group, respectively [7]; UV absorption maxima at 227, 312 nm and bathochromic shift under alkaline conditions. Except for signals attributed to this moiety, the ^{1}H NMR and ^{13}C NMR data of **4** were very similar to those of **3**. Hence, **4** is likely to be the *O-trans-p*-coumaroyl ester of **3**, with the additional mass unit $C_9H_6O_2$ compared to **3**. This ester was located at C-2 as evidenced by the downfield shift of H-2, δ 4.78 (2H, *br s*) relative to δ 4.06 (*dd*) and 4.36 (*dd*) in **3**. The COSY-45 spectrum revealed the following coupling relationship: H-3 (δ 4.06, *d*, J = 8.7 Hz) $\langle-\rangle$ H-1 (δ 2.00, *m*) $\langle-\rangle$ H-2 (δ 4.78, *br s*), supporting the partial structure for ring A. The (1*R*,3*R*)-configuration, the same as that in **3**, was assigned from the consistent coupling constant between H-1 and H-3 (8.7 Hz) and the similarity of chemical shifts for the adjacent carbons and protons (Tables 1 and 2). This established **4** to be the novel natural product 2-*O*-(*trans-p*-coumaroyl)-ceanothanolic acid. Its ^{13}C NMR data were assigned by correlation with those of **3** (Table 2).

The structure assigned for **4** was supported by the FAB and EI mass spectra, both of which showed the base peak at *m/z* 147 (**A**) for the *p*-coumaroyl moiety, and major fragment ions at *m/z* 454 (**B**) and 437 (**C**), obtained presumably from a McLafferty rearrangement and subsequent cleavage of the C-3 hydroxyl function.

Compound **5** was characterized as the methyl ester **5a**, which had a molecular formula $C_{31}H_{50}O_5$, deduced from HREI mass spectrum. The IR spectrum showed hydroxyl (3454 cm^{-1}), ester (1726 cm^{-1}), and trisubstituted olefinic functions (1645, 889 cm^{-1}). Besides the *O*-methyl singlet (δ 3.58), its ^{1}H NMR spectrum ($CDCl_3$) exhibited 8 methyl signals (7 singlets and one doublet), two coupled carbinoyl proton

Table 2. HMBC data of compounds **1** and **3**, and ^{13}C NMR data (δ in ppm, mult.*) of compounds **1**–**4**

Position	1	HMBC of **1** corr. H (#)	2	3	HMBC of **3** corr. H (#)	4†
1	157.9 *s*	2, 25	73.9 *d*	62.9 *d*	25	62.3 *d*
2	191.7 *d*	3	206.1 *d*	64.6 *t*	1, 3	65.7 *t*
3	164.2 *d*	23, 24	80.9 *d*	87.0 *d*	1, 2, 23, 24	84.2 *d*
4	44.0 *s*	3, 23, 24	41.2 *s*	39.7 *s*	3, 23, 24	39.7 *s*
5	63.7 *d*	3, 23, 24, 25	63.0 *d*	62.7 *d*	23, 24, 25	60.4 *d*
6	17.2 *t*		18.5 *t*	18.7 *t*	5	18.8 *t*
7	35.7 *t*	26	34.6 *t*	35.1 *t*	26	35.1 *t*
8	43.5 *s*	26, 27	42.3 *s*	42.4 *s*	7, 26, 27	42.5 *s*
9	48.2 *d*	25, 26	50.5 *d*	50.9 *d*	1, 7, 25, 26	50.8 *d*
10	52.7 *s*	2, 3, 25	48.2 *s*	44.5 *s*	1, 25	44.6 *s*
11	24.9 *t*		24.9 *t*	24.2 *t*		23.9 *t*
12	26.0 *t*		25.6 *t*	25.8 *t*		25.9 *t*
13	38.6 *d*	27	38.5 *d*	38.5 *d*	18, 27	38.7 *d*
14	43.1 *s*	26, 27	43.1 *s*	43.2 *s*	16, 26, 27	43.3 *s*
15	30.6 *t*	27	30.4 *t*	30.5 *t*	27	30.6 *t*
16	33.2 *t*		32.9 *t*	33.1 *t*	18	33.1 *t*
17	56.7 *s*		56.5 *s*	56.6 *s*	18	56.6 *s*
18	50.0 *d*		49.7 *d*	50.0 *d*	13, 19	50.8 *d*
19	47.9 *d*	29, 30	47.8 *d*	47.9 *d*	18, 29, 30	47.9 *d*
20	151.3 *s*	29	151.2 *s*	151.3 *s*	29	151.3 *s*
21	31.4 *t*		31.2 *t*	31.4 *t*	19, 22	31.3 *t*
22	37.8 *t*		37.6 *t*	37.7 *t*		37.7 *t*
23	28.3 *q*	24	26.3 *q*	26.2 *q*	24	26.5 *q*
24	20.6 *q*	23	25.6 *q*	25.8 *q*	23	25.8 *q*
25	19.5 *q*		14.8 *q*	14.6 *q*		14.7 *q*
26	18.2 *q*		17.0 *q*	17.3 *q*		17.3 *q*
27	15.1 *q*		15.0 *q*	15.0 *q*		15.0 *q*
28	179.1 *s*	18	178.8 *s*	178.8 *s*	18	178.8 *s*
29	19.7 *q*	30	19.4 *q*	19.6 *q*	30	19.6 *q*
30	110.2 *t*	29	110.0 *t*	110.0 *t*	29	110.1 *t*

* Multiplicities were obtained from DEPT experiments.

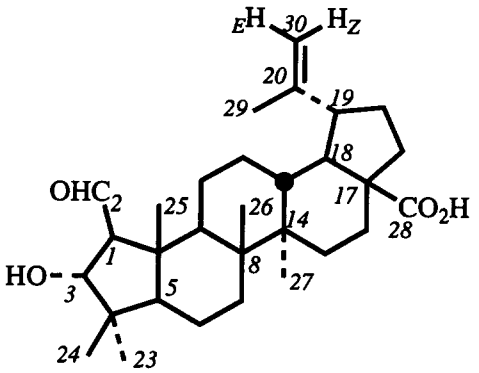
† Carbon signals of *p*-coumaroyl moiety in **4** appeared at δ 126.4 (*s*, C-1′), 130.8 (*d*, C-2′ and C-6′), 116 (*d*, C-3′ and C-5′), 161.4 (*s*, C-4′), 145.1 (*d*, C-7′), 115.8 (*d*, C-8′) and 167.8 (*s*, C-9′).

signals at δ 3.39 (H-3, *d*, $J = 3.0$ Hz) and 3.95 (H-2, *dt*, $J = 11.5$ and 3.0 Hz), and an olefinic proton signal at δ 5.34 (H-12, *t*, $J = 3.6$ Hz). These data and characteristic mass fragment ions at *m/z* 278 (**D**) and 219 (**F**), obtained from a retro-Diels–Alder fragmentation process and subsequent cleavage of the carboxylic ester, respectively, suggested **5a** to be a Δ^{12}-ursen-28-oic acid triterpene. Further NOED studies (data, see Experimental) supported the identification of **5a** as euscaphic acid methyl ester. Ring D and ring E are *cis*-fused as evidenced by the weak NOE between H-18 and 28-OMe. The NOE of H-18 to H-12, H-20 and H-29 also established the 18*S*,19*R*,20*R*-configuration, which was confirmed recently by X-ray crystallography [8]. Although the ^1H NMR and ^{13}C NMR data of the parent compound in deuteriated pyridine have been assigned [9, 10], the data of this methyl ester measured in $CDCl_3$ were unambiguously assigned and listed in the experimental section for reference by 2D NMR techniques (COSY-45, HSQC and HMBC) and NOED. The large effect on proton chemical shifts measured in these two solvents was observed.

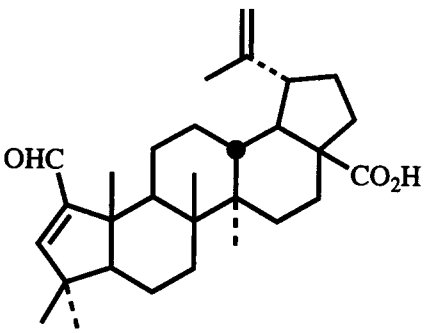
Compounds **1**, **3** and **4** have a ceanothane skeleton but different orientations of substituents at C-1 and C-3 in comparison with those of ceanothic acid. Compound **4** represents the second natural occurrence of an aromatic carboxylic acid ester of ceanothane type, the first one having been reported recently from *Zizyphus jujuba* [11]. These compounds could be biogenetically derived from betulinic acid, which after dehydration and oxidative ring A opening might yield the seco-betulinic acid dialdehyde (**6**). Subsequent aldol condensation of the dialdehyde would give products with different configurations at C-1 and C-3 (Scheme 1). Among the four isomers, the one having 1*S*,3*R*-configuration (**1**) yielded **2** after dehydration, and yielded **3** and **4**, respectively, after reduction and subsequent esterification. On the other hand, the compound having 1*R*,3*S*-configuration gave ceanothic acid after C-2 oxidation.

EXPERIMENTAL

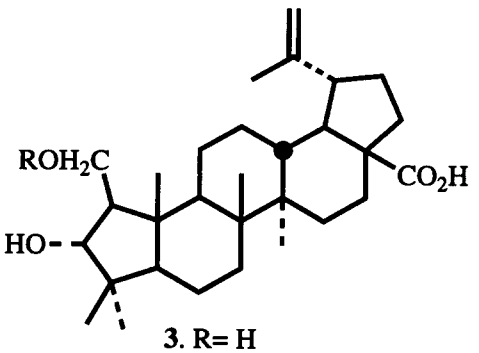
General. Mps: uncorr; ^1H NMR (400.13 MHz) and ^{13}C NMR (100.61 MHz): pyridine-d_5 or $CDCl_3$ using



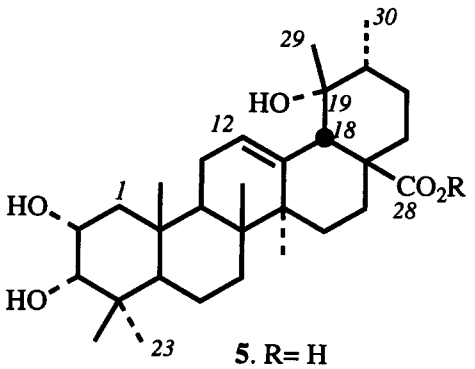
1



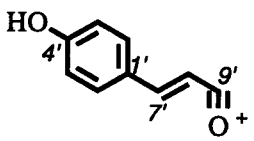
2



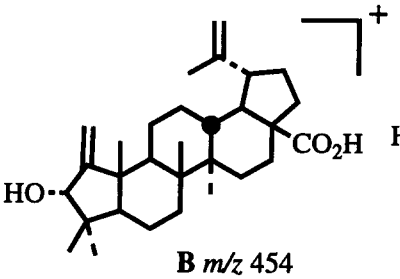
3. R= H
4. R= trans-*p*-coumaroyl



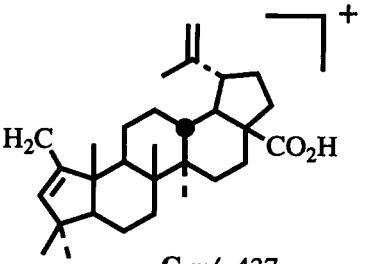
5. R= H
5a. R= Me



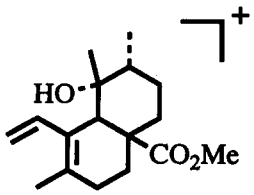
A *m/z* 147



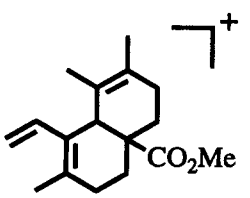
B *m/z* 454



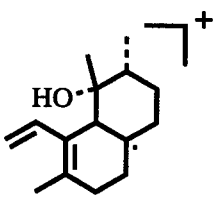
C *m/z* 437



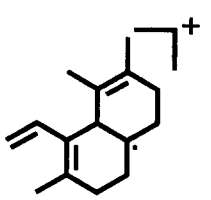
D *m/z* 278



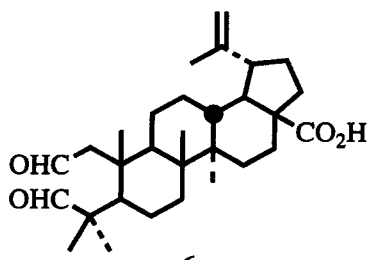
E *m/z* 260



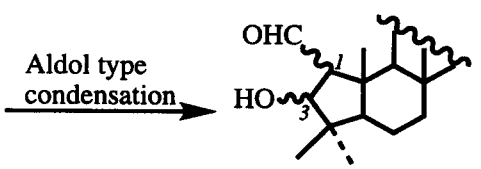
F *m/z* 219



G *m/z* 201



6



1. 1-*S*, 3-*R*

Scheme 1.

the solvent peak as int. standard; MS: direct inlet system; UV: MeOH; IR: KBr disc.

Plant material. The roots of *P. hemsleyanus* Rehd. were collected in October, 1991 in Fangchang County, Anhwei province, China. A specimen was authenticated by Dr Liu Yong-Long and colleagues at the Institute of Medicinal Plant Development, Chinese Academy of Medical Sciences, Beijing, People's Republic of China.

Extraction and isolation. Dried ground powders of the roots (14.0 kg) were macerated with 95% EtOH (100 l × 3) at 50°. The EtOH soln was condensed under red. pres. to give 0.8 kg of EtOH extract. The extract (686 g) was then triturated with H_2O and 5% HOAc to give H_2O-soluble, alkaloidal and H_2O-insoluble frs. The H_2O-insoluble fr. (204 g) was divided into frs. soluble in *n*-C_6H_{14} (7 g), $CHCl_3$ (60 g) and MeOH (53 g) by trituration with the corresponding solvent (*ca* 0.5 l × 3). Part of $CHCl_3$ fr. (16 g) were then sepd on a silica gel column (0.6 kg, finer than 230 mesh), eluted with 0 to 20% MeOH in $CHCl_3$ to give 19 subfrs. Another silica gel column (10 g) for subfr. 1 (307 mg) eluted with 0 to 20% Me_2CO in hexane gave four frs, of which fr. 2 (84 mg) and fr. 4 (51 mg) yielded lupeol (30 mg) and betulinaldehyde (6 mg), respectively, upon recrystallization from MeOH. Recrystallization of subfrs 2 (122 mg), 4 (42 mg), 6 (137 mg), 8 (1.13 g), 10 (594 mg), 12 (210 mg), and 19 (1.46 g) from MeOH, respectively, yielded sitosterol and stigmasterol mixt. (28 mg, the latter as major) (fr. 2), betulin (18 mg), **2** (75 mg), betulinic acid (300 mg), **1** (130 mg), **3** (57 mg), and mixture of 3-*O*-*β*-glucosyl sitosterol-*β*-D-glucoside and 3-*O*-*β*-glucosyl stigmasterol [12]. Another silica gel column (7 g, 230–400 mesh) for subfr. 14 (171 mg) eluted with 0 to 40% Me_2CO in toluene gave a fr. (26 mg) which yielded **4** (6 mg) after recrystallization from MeOH. Subfr. 15 (221 mg) was *O*-methylated with freshly prepd ethereal CH_2N_2 and the crude products were sepd on a silica gel column (0 to 20% Me_2CO in $CHCl_3$) to give a fr. (13 mg) which yielded **5a** (3 mg) after recrystallization from MeOH. Recrystallization of subfr. 17 (266 mg) from $CHCl_3$ yielded ceanothic acid (70 mg).

Zizyberenalic acid (**2**). R_f 0.24 [MeOH–$CHCl_3$–HOAc (10:190:1)]; mp 214–216°; $[\alpha]_D^{18}$ +24° (MeOH; *c* 0.50). IR ν_{max} cm^{-1}: 2500–3100 (COOH), 2728 (CHO), 1714 (sh) and 1686 (>C=O), 1642 and 881 (>C=CH_2), 1452, 1378. UV λ_{max} nm (log ε): 239 (3.48), 266 (sh, 3.22). ^{1}H NMR and ^{13}C NMR: Tables 1 and 2; NOED data (%): H-2 to H-3 (12.3), H-23 to H-24 (4.8), H-24 to H-3 (5.7) and H-23 (6.3), H-25 to H-26 (5.0). HR EI MS $[M]^+$ *m/z*: 452.3272 ($C_{30}H_{44}O_3$ requires 452.3290). EIMS *m/z* (rel. int.): 452 $[M]^+$ (12), 315 (2), 248 (3), 229 (2), 216 (3), 211 (3), 205 (2), 203(2), 189 (13), 187 (2), 138 (100).

Ceanothanolic acid (**3**). R_f 0.40 [MeOH–$CHCl_3$–HOAc (20:180:1)]; mp 286°; $[\alpha]_D^{18}$ −15° (pyridine; *c* 0.50). IR ν_{max} cm^{-1}: 2500–3500 (COOH, OH), 1682 (>C=O), 1641 and 881 (>C=CH_2), 1452, 1377, 1105. ^{1}H NMR and ^{13}C NMR: Tables 1 and 2; NOED data (%): H-23 to H-24 (7.7), H-24 to H-3 (8.0) and H-23 (5.6) and H-25 (7.7), H-25 to H-3 (9.0), H-24 (4.3) and H-26 (11.3), H-26 to H-25 (6.0) and H-13 (4.5), H-27 to H-18 (13.4). HR EI MS $[M]^+$ *m/z*: 472.3511 ($C_{30}H_{48}O_4$ requires 472.3553). EIMS *m/z* (rel. int.): 472 $[M]^+$ (7), 454 (18), 426 (10), 259 (55), 248 (65), 223 (60), 216 (10), 206 (65), 203 (40), 189 (90), 177 (35), 177 (30), 148 (90).

2-O-(trans-p-*Coumaroyl*)-*ceanothanolic acid* (**4**). R_f 0.34 [Me_2CO–$CHCl_3$ (1:9), × 2]; mp > 295°. IR ν_{max} cm^{-1}: 2500–3500 (COOH, OH), 1706 and 1680 (>C=O), 1607 and 1515 (HC=CH), 1637 and 887 (>C=CH_2), 1378, 829 (aryl CH). UV λ_{max} nm (log ε): 227 (3.48), 296 (sh, 3.22), 312 (4.42); MeOH + KOH: 357 (4.56). ^{1}H NMR and ^{13}C NMR: Tables 1 and 2; FABMS *m/z* (rel. int.): 619 $[M+H]^+$ (2), 437 (15), 147 (100), 91 (55); EIMS *m/z* (rel. int.): 471 (50), 454 (55, **B**), 437 (60, **C**), 203 (80), 189 (90), 147 (100, **A**).

Known compounds with additional data: Colubrinic acid (**1**): R_f 0.54 [MeOH–$CHCl_3$–HOAc (20:180:1)]; mp 262–264°; $[\alpha]_D^{18}$ −17° (pyridine: *c* 0.50). ^{1}H NMR and ^{13}C NMR: Tables 1 and 2; NOED data (%): H-23 to H-24 (15.9), H-24 to H-3 (12.5) and H-23 (8.6), H-25 to H-2 (12.0), H-3 (10.8) and H-26 (3.2), H-26 to H-25 (7.1) and H-13 (16). HR EI MS $[M]^+$ *m/z*: 470.3414 ($C_{30}H_{46}O_4$ requires 470.3396). Euscaphic acid methyl ester (**5a**): R_f 0.34 [Me_2CO–$CHCl_3$ (1:9), × 2]; mp 220–223°; $[\alpha]_D^{18}$ +31° (pyridine; *c* 0.65). IR ν_{max} cm^{-1}: 3454 (OH), 1726 (>C=O), 1645 and 889 (>C=CHR), 1189 (C—O). ^{1}H NMR ($CDCl_3$): δ 5.34 (1H, *br t*, *J* = 3.4 Hz, H-12), 3.98 (1H, *ddd*, *J* = 11.5, 7.0, 2.5 Hz, H-2), 3.58 (3H, *s*, 28-OMe), 3.41 (1H, *d*, *J* = 2.5 Hz, H-3), 2.58 (1H, *s*, H-18), 2.50 (1H, *dt*, *J* = 5.6, 13.0 Hz, H-16α), 1.62 (1H, *dd*, *J* = 12.4, 4.7 Hz, H-1β), 1.60 (1H, *m*, H-15), 1.38 (1H, *m*, H-20), 1.24 (3H, *s*, H-27), 1.21 (1H, *m*, H-1α), 1.19 (3H, *s*, H-29), 1.00 (3H, *s*, H-23), 0.93 (3H, *s*, H-25), 0.92 (3H, *d*, *J* = 6.4 Hz, H-30), 0.84 (3H, *s*, H-24), 0.65 (3H, *s*, H-26); NOED data ($CDCl_3$) (%): H-18 to H-12 (13.6), H-20 (6.9), 28-OMe (1.0) and H-29 (3.3), H-23 to H-3 (4.4), H-24 (3.3), H-24 to H-2 (4.6), H-3 (5.2), H-24 (3.1) and H-25 (2.7), H-25 to H-2 (6.8), H-24 (2.5) and H-26 (6.6), H-26 to H-25 (9.4), H-29 to H-12 (8.1), H-18 (5.7) and H-30 (7.9), H-30 to H-20 (6.3) and H-29 (9.5). ^{13}C NMR ($CDCl_3$): δ 41.7 (*t*, C-1), 66.4 (*d*, C-2), 78.9 (*d*, C-3), 38.3 (*s*, C-4), 48.1 (*d*, C-5), 18.1 (*t*, C-6), 32.6 (*t*, C-7), 40.2 (*s*, C-8), 46.9 (*d*, C-9), 38.2 (*s*, C-10), 23.7 (*t*, C-11), 129.1 (*d*, C-12), 138.2 (*s*, C-13), 41.3 (*s*, C-14), 28.2 (*t*, C-15), 25.5 (*t*, C-16), 47.9 (*s*, C-17), 53.2 (*d*, C-18), 73.1 (*s*, C-19), 41.1 (*d*, C-20), 26.0 (*t*, C-21), 37.4 (*t*, C-22), 28.5 (*q*, C-23), 21.8 (*q*, C-24), 16.21 (*q*, C-25), 16.7 (*q*, C-26), 24.7 (*q*, C-27), 178.4 (*s*, C-28), 27.4 (*q*, C-29), 16.1 (*q*, C-30) and 51.5 (*q*, 28-OMe); Major HMBC (*J* = 8 Hz): H-18 to C-12, C-13, C-17, C-19 and C-28, H-23 to C-3, C-4, C-5 and C-24, H-24 to C-3, C-4, C-5 and C-23, H-25 to C-1, C-5, C-9 and C-10, H-26 to C-7, C-8, C-9 and C-14, H-27 to C-8, C-13, C-14 and C-15, H-29 to C-18, C-19 and C-20, H-30 to C-19, C-20 and C-21, 28-OMe to C-28. HR EI MS $[M]^+$ *m/z*:

502.3659 ($C_{31}H_{50}O_4$ requires 502.3658); EIMS *m/z* (rel. int.): 502 $[M]^+$ (33), 484 (16), 443 (37), 442 (100), 425 (13), 278 (16, **D**), 260 (36, **E**), 223 (43), 219 (60, **F**), 218 (54), 201 (80, **G**).

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