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TWO TRITERPENOID CARBOXYLIC ACIDS FROM *ACANTHOPANAX TRIFOLIATUS**

PH. D. TY,† M. LISCHEWSKI,‡ H. V. PHIET,† A. PREISS,‡ T. V. SUNG,? J. SCHMIDT‡ and G. ADAM‡

†Institute of Chemistry, National Research Centre of the S.R.V., Hanoi, Vietnam; *Institute for Plant Biochemistry, Academy of Sciences of the G.D.R., Halle/Saale, G.D.R.

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Key Word Index-Acanthoponax *trifoliatus*; Araliaceae; triterpenes; lupenes3α,1 la-dihydroxylup-20(29)-en-28-oic acid and 3α,11α,23-trihydroxylup-20(29)-en-28-oic acid.

Abstract-Two new triterpenoid carboxylic acids have been isolated from leaves of ***Acanthopanax trifoliatus*** and their structures elucidated as 3α,1 la-dihydroxylup-20(29)-en-28-oic acid and 3α,11α,23-trihydroxylup-20(29)-en-28-oic acid by physical data and chemical transformations.

INTRODUCTION

As a continuation of our phytochemical investigations on Vietnamese plants of medical and biological interest [1] we have examined the species ***Acanthopanax trifoliatus*** (L.) Merr., used in the folk medicine of Southeast Asia [2, 3] as a drug with ginseng-like activity. In this paper we report the isolation and structural elucidation of two new triterpenoid carboxylic acids from the leaves of this plant. On the basis of spectroscopic data and chemical transformations, the constituents are shown to be 3α,11α-dihydroxylup-20(29)-en-28-oic acid **(1)** and 3α,11α,23-trihydroxylup-20(29)-en-28-oic acid (2).

RESULTS AND DISCUSSION

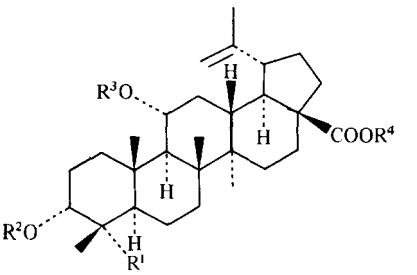
Extraction of dried leaves with methanol yielded after silica gel column chromatography 0.6 % of the less polar compound 1 ($C_{30}H_{48}O_4[M]^+$ at m/z 472.3454) and 1.2% of the main constituent 2 ($C_{30}H_{48}O_5[M]^+$ at m/z 488.3502). Their IR spectra showed absorptions assignable to hydroxyl, carboxyl and $>C=CH_2$ functions.

The acids **1** and 2 on treatment with diazomethane gave the corresponding methyl esters 3 and 4. Acetylation of 3 and 4 with acetic anhydride-pyridine for 16 hr at 20" afforded the diacetate 5 and the triacetate 6, respectively.

The mass spectra of **l-8** showed typical fragment ions **(a, b, c, d)** which had arisen from ring C cleavage [1, 4]. The appearance of the key ion **a** is evidence for a C-l 1 substitution [1, 5]. This is supported by ion **d** formed by cleavage of the C-9, C-l 1 bond. Furthermore, the e-type ion derived from **1, 2, 3** and 4 locates the carboxylic group of **1** and 2 at C-17 (C-28) [4]. The presence of a primary hydroxyl group on ring A of compound 2 is indicated by the loss of 30 mu (CH_2O) during the substituent elimination in 2, 4 and 8 (see Experimental).

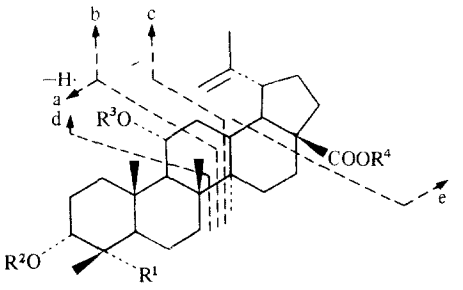
The ^1H NMR spectrum of **1** showed signals for six tertiary methyl groups (one of them shifted downfield to δ1.65), two secondary alcohol functions (3.22, 3β-H, *t*, $|J_{AX}+J_{BX}| = 5.4$ Hz; **3.81,** 11β-H, six-line pattern J = 10.5 Hz, J' = 5.0 Hz) and two olefinic protons (4.51 and 4.66, 2 x m).

The ^1H NMR spectrum of 2 displayed five tertiary methyl groups (one of them also shifted downfield to δ1.65), two olefinic protons (4.52 and 4.67, 2 x m), two secondary alcohol functions (3.50, 3β-H, t, $|J_{AX}+J_{BX}|$ = 5.4 Hz; 3.82, 11β-H, six-line pattern J = 10.5 Hz, J = 10.5 Hz, J' = 5.0 Hz) and two protons for a primary alcohol group (δ_A 3.20, J = 11.0 Hz and δ_B 3.42, J = 11.0 Hz). The ^{13}C NMR shift values of compounds **1**



	R¹	R²	R³	R⁴
1	Me	H	H	H
2	CH_2OH	H	H	H
3	Me	H	H	Me
4	CH_2OH	H	H	Me
5	Me	Ac	Ac	Me
6	CH_2OAc	Ac	Ac	Me
7	COOH	H	H	H
8	CH_2OH	H	AC	Me
9	CH_2OH	Ac	AC	Me
10	CH_2OAc	H	Ac	Me
11	COOH	Ac	AC	Me
12	COOMe	Ac	Ac	Me

*Part 12 in the series "Natural Products from Vietnamese Plants". For Part 11 see Lischewski, M., Viet-Nam, V., Phiet, H. V., Schmidt, J. and G. Adam (1984) ***Pharmazie 39, 276.***



Scheme 1. Main skeletal fragmentation of lupane derivatives **1–8**.

and **2** are shown in Table 1. The assignments of all 30 carbon atoms were made on the basis of the observed multiplicities as well as the reduced coupling constants in the SFORD spectra and comparison with reported ^{13}C NMR data of related triterpenes [1, 6, 7]. Thus the shift values of most carbon atoms with the exceptions of C-3 to C-6 and C-23 and for **1** also C-24 corresponded with those of **7** [1]. The remaining signals were identified by inspection of the ^{13}C NMR spectral data of triterpenes, which have the same A-ring partial structures as those expected for the compounds in the present study. Thus, a good agreement of the shift values for **1** was found with betulafolianetriol [6] and for **2** with 3α,23-dihydroxylup-20(29)-ene [7].

Table 1. ^{13}C NMR spectral data of **1** and **2** (50.3 MHz)

	1	**2**
C-1	36.3	36.0 *t*
C-2	27.0	27.2 *t*
C-3	75.3	76.0 *d*
C-4	38.5	41.3 *s*
C-5	49.6	44.0 *d*
C-6	18.6	18.4 *t*
C-7	36.0	35.7 *t*
C-8	42.8	42.9 *s*
C-9	56.3	56.3 *d*
C-10	40.0	39.7 *s*
C-11	69.9	70.0 *d*
C-12	38.5	38.5 *t*
C-13	37.7	37.7 *d*
C-14	43.0	42.9 *s*
C-15	30.2	30.2 *t*
C-16	32.9	32.9 *t*
C-17	56.6	56.6 *s*
C-18	49.5	49.5 *d*
C-19	47.6	47.6 *d*
C-20	150.9	150.9 *s*
C-21	31.3	31.3 *t*
C-22	37.5	37.4 *t*
C-23	29.8	72.0 *t*
C-24	22.9	18.4† *q*
C-25	18.6*	17.8† *q*
C-26	17.8*	17.1 *q*
C-27	14.8	14.9 *q*
C-28	178.8	178.8 *s*
C-29	110.1	110.1 *t*
C-30	19.6	19.6

*,† These assignments may be interchanged.

Spectra were recorded in pyridine-d_5 (TMS).

From the above-mentioned data, the new triterpenoic acids are regarded as 3α,11α-dihydroxylup-20(29)-en-28-oic acid (**1**) and 3α,11α,23-trihydroxylup-20(29)-en-28-oic acid (**2**), respectively.

Chemical transformation of **2** to the known [1] lupene derivative **12** was achieved in the following manner. Selective deacetylation of the triacetate **6** gave the 11-monoacetyl derivative **8**. Short-time reacetylation of **8** afforded a mixture of diacetates **9** and **10**, which were separated by silica gel column chromatography from unchanged triacetate **6**. Oxidation of **9** and **10** with pyridinium dichromate in dimethyl formamide gave the desired 23-oic acid **11** and the 3-oxo compound of **10**. The acid **11** was methylated with diazomethane to form the known dimethyl ester diacetate **12**, identical in all respects to authentic material [1].

EXPERIMENTAL

An EA-mass spectrograph of the research institute "Manfred von Ardenne", Dresden, was used to record the positive-ion mass spectra (10–16 eV, duoplasmatron ion source, plasma gas Ar, direct inlet system). Exact mass measurements were obtained from a JEOL JMS D-100 instrument operating at 75 eV. *Acanthopanax trifoliatus* (L.) Merr. was identified by Dr. Ph. V. Nguyen, Institute of Biology, National Research Centre of the S.R.V., Hanoi; a voucher specimen has been deposited there.

Isolation of compounds **1** *and* **2**. Dried and powdered leaves (200 g), collected near Hanoi in April 1981, were extracted with boiling MeOH for 6 hr. The solvent was removed *in vacuo* and the residue (32 g) chromatographed on a silica gel column using increasing concns of $CHCl_3$ in petrol followed by EtOAc in $CHCl_3$ as the eluant. Elution with petrol–$CHCl_3$ (1:1) gave 1.2 g (0.6%) **1**: mp 212–215° (Me_2CO–petrol); $[\alpha]_D^{25}$ 0 (*c* 0.44 in EtOH); IR ν_{max}^{nujol} cm^{-1}: 1640 (>C=CH_2), 1695 (COOH), 3070 (>C=CH_2), 3460 (br OH); MS *m/z* (rel. int.): 472.3454 ($C_{30}H_{48}O_4$ calc. 472.3552) $[M]^+$ (4), 454.3460 $C_{30}H_{46}O_3$ (calc. 454.3447) $[M-H_2O]^+$ (21), 436.3346 ($C_{30}H_{47}O_2$ calc. 436.3341) $[M-2H_2O]^+$ (19), 421 (11), 408 (6), 393 (5), 327 (7), 300 (18), 285 (19), 264 **[d]**$^+$ (13), 259 (19), 255 (16), 246 **[d** $-H_2O]^+$ (33), 237 **[a]**$^+$ (37), 234 **b** (98), 219 **[c** $-H]^+$ (46), 201 (57), 189 **[b** $-CO_2H]^+$ (80), 175 (86), 161 (49), 152 **[e]**$^+$ (83), 147 (71), 135 (100), 121 (94), 107 (82). ^1H NMR (Me_2CO-d_6–HMDS): δ0.78, 0.86, 0.90, 0.99, 1.00 (5 × *s*, 23-H_3, 24-H_3, 25-H_3, 26-H_3, 27-H_3), 1.65 (*s*, 30-H_3), 3.22 (*t*, $|J_{AX}+J_{BX}|$ = 5.4 Hz, 3β-H), 3.81 (six-line pattern, *J* = 10.5 Hz, *J'* = 5.0 Hz, 11β-H), 4.51 and 4.66 (2 × *m*, 29-H_2).

Further elution with $CHCl_3$–EtOAc (4:1) afforded 2.4 g (1.2%) **2**: mp 192–196° (Me_2CO–petrol); $[\alpha]_D^{25}$ +3.3° (*c* 0.30 in EtOH); IR ν_{max}^{nujol} cm^{-1}: 1645 (>C=CH_2), 1690 (COOH), 3075 (>C=CH_2), 3375 (br OH); MS *m/z* (rel. int.): 488.3502 ($C_{30}H_{48}O_5$ calc. 488.3502) $[M]^+$ (4), 470 (18), 452 (35), 440 $[M-H_2O-CH_2O]^+$ (28), 437 (20), 422 (32), 407 (15), 393 (8), 371 (27), 353 (16), 325 (19), 300 (22), 287 (22), 273 (17), 259 (25), 255 (28), 253 **[a]**$^+$ (24), 246 **[d** $-H_2O]^+$ (26), 235 **[a** $-H_2O]^+$ (69), 234 **[b]**$^+$ (100), 217 (59), 205 (53), 201 (58), 189 (76), 175 (81), 161 (56), 152 **[e]**$^+$ (49), 151 **[e** $-H]^+$ (53), 147 (71), 133 (71), 121 (94), 107 (82). ^1H NMR (Me_2CO-d_6–HMDS): δ0.63, 0.93, 1.00, 1.03 (4 × *s*, 24-H_3, 25-H_3, 26-H_3, 27-H_3), 1.65 (*s*, 30-H_3), 3.20 and 3.42 (two *d*, *J* = 11.0 Hz, 23-H_2), 3.50 (*t*, $|J_{AX}+J_{BX}|$ = 5.4 Hz, 3β-H), 3.82 (six-

line pattern, J = 10.5 Hz, J' = 5.0 Hz, 11β-H), 4.52 and 4.67 (2 × *m*, 29-H_2).

Methyl esters **3** *and* **4**. To **1** or **2** in MeOH was added excess CH_2N_2. After 5 min the solvent was removed *in vacuo* and the residue chromatographed on silica gel. Compound **3**: mp 179–200° (Me_2CO–petrol); $[\alpha]_D^{25}$ 0° (*c* 0.37 in EtOH); IR ν_{max}^{nujol} cm^{-1}: 1645 (>C=CH_2), 1720 (COOMe), 3075 (>C=CH_2), 3550 (br OH); MS *m/z* (rel. int.): 486 $[M]^+$ (21), 468 (28), 453 (17), 450 $[M-2H_2O]^+$ (22), 435 (11), 426 $[M-HCO_2Me]^+$ (17), 408 (12), 395 (8), 341 (8), 332 (9), 315 (23), 299 (12), 288 (10), 278 $[\mathbf{d}]^+$ (31), 273 (26), 260 $[\mathbf{d}-H_2O]^+$ (32), 255 (45), 250 (74), 248 $[\mathbf{b}]^+$ (94), 237 $[\mathbf{a}]^+$ (41), 234 $[\mathbf{c}]^+$ (45), 219 (52), 205 (48), 201 (63), 189 (100), 175 (85), 168 $[\mathbf{e}+2H]^+$ (54), 161 (44), 147 (50), 135 (88), 123 (66), 107 (51). ^{1}H NMR ($CDCl_3$–HMDS): δ0.80, 0.91, 0.91, 0.96, 1.00 (5 × *s*, 23-H_3, 24-H_3, 25-H_3, 26-H_3, 27-H_3), 1.65 (*s*, 30-H_3), 3.30 (*t*, $|J_{AX}+J_{BX}|$ = 5.4 Hz, 3β-H), 3.60 (*s*, COOMe), 3.89 (six-line pattern, J = 10.5 Hz, J' = 5.0 Hz, 11β-H), 4.55 and 4.69 (2 × *m*, 29-H_2).

Compound **4**: mp 230–234° (Me_2CO–petrol); $[\alpha]_D^{25}$ 0° (*c* 0.38 in EtOH); IR ν_{max}^{nujol} cm^{-1}: 1645 (>C=CH_2), 1730 (CO_2Me), 3075 (>C=CH_2), 3300 (br OH); MS *m/z* (rel. int.): 502 $[M]^+$ (30), 484 (35), 466 (45), 454 $[M-H_2O-CH_2O]^+$ (30), 451 (28), 442 $[M-HCO_2Me]^+$ (30), 436 $[M-2H_2O-CH_2O]^+$ (39), 425 (24), 407 (19), 393 (21), 385 (28), 367 (13), 325 (19), 314 (24), 301 (19), 287 (11), 278 $[\mathbf{d}]^+$ (32), 273 (33), 255 (49), 253 $[\mathbf{a}]^+$ (38), 250 (74), 248 $[\mathbf{b}]^+$ (98), 235 (61), 234 $[\mathbf{c}]^+$ (54), 217 (66), 205 (62), 201 (66), 189 (100), 175 (99), 168 $[\mathbf{e}+2H]^+$ (54), 161 (55), 147 (64), 133 (64), 121 (84), 109 (73). ^{1}H NMR ($CDCl_3$–HMDS): δ0.65, 0.89, 0.97, 1.03 (4 × *s*, 24-H_3, 25-H_3, 26-H_3, 27-H_3), 1.65 (*s*, 30-H_3), 3.27 and 3.47 (two *d*, J = 11.0 Hz, 23-H_2), 3.52 (*t*, $|J_{AX}+J_{BX}|$ = 5.4 Hz, 3β-H), 3.60 (*s*, COOMe), 3.88 (six-line pattern, J = 10.5 Hz, J' = 5.0 Hz, 11β-H), 4.55 and 4.70 (2 × *m*, 29-H_2).

Acetates **5** *and* **6**. Acetylation of **3** or **4** with Ac_2O–pyridine (12 hr at 20°) gave after silica gel purification the acetates **5** and **6**, respectively. Compound **5**: amorphous; $[\alpha]_D^{25}$ −21.2° (*c* 0.32 in EtOH); IR ν_{max}^{nujol} cm^{-1}: 1645 (>C=CH_2), 1735 (ester), 3075 (>C=CH_2); MS *m/z* (rel. int.): 570 $[M]^+$ (25), 510 $[M-HOAc]^+$ (92), 467 (22), 451 (69), 450 $[M-2HOAc]^+$ (76), 428 (32), 407 (24), 391 $[M-2HOAc-CO_2Me]^+$ (30), 375 (22), 367 (14), 321 **a** (68), 320 **d** (45), 314 (46), 299 (34), 279 (21), 267 (30), 260 $[\mathbf{d}-HOAc]^+$ (62), 255 (75), 247 $[\mathbf{b}-H]^+$ (83), 241 (47), 234 $[\mathbf{c}]^+$ (54), 219 (88), 201 (87), 187 (99), 175 (96), 161 (54), 147 (59), 134 (100), 121 (85), 107 (65). ^{1}H NMR ($CDCl_3$–HMDS): δ0.80, 0.83, 0.93, 0.93, 1.01 (5 × *s*, 23-H_3, 24-H_3, 25-H_3, 26-H_3, 27-H_3), 1.62 (*s*, 30-H_3), 1.89 and 2.02 (2 × *s*, acetates), 3.62 (*s*, COOMe), 4.52 (*t*, $|J_{AX}+J_{BX}|$ = 5.4 Hz, 3β-H), 4.52 and 4.67 (2 × *m*, 29-H_2), 5.15 (six-line pattern, J = 10.5 Hz, J' = 5.0 Hz, 11β-H).

Compound **6**: amorphous:; $[\alpha]_D^{25}$ −12.4° (*c* 0.30 in EtOH); IR ν_{max}^{nujol} cm^{-1}: 1645 (>C=CH_2), 1735 (ester), 3075 (>C=CH_2); MS *m/z* (rel. int.): 628 $[M]^+$ (9), 568 $[M-HOAc]^+$ (74), 525 (13), 509 (53), 508 $[M-2HOAc]^+$ (56), 493 (16), 486 (24), 449 (24), 448 $[M-3HOAc]^+$ (30), 433 (18), 389 $[M-3HOAc-CO_2Me]^+$ (19), 379 **a** (51), 337 (11), 320 **d** (19), 314 (28), 299 (22), 277 (60), 260 $[\mathbf{d}-HOAc]^+$ (53), 255 (55), 247 $[\mathbf{b}-H]^+$ (80), 241 (32), 234 **c** (42), 217 (68), 201 (81), 187 (100), 175 (81), 159 (46), 147 (58), 133 (66), 119 (62), 107 (56). ^{1}H NMR ($CDCl_3$–HMDS): δ0.94, 0.94, 0.96, 1.00 (4 × *s*, 24-H_3, 25-H_3, 26-H_3, 27-H_3), 1.62 (*s*, 30-H_3), 1.89, 1.94 and 1.98 (3 × *s*, acetates), 3.61 (COOMe), 3.73 and 3.99 (two *d*, J = 11.0 Hz, 23-H_2), 4.52 and 4.68 (2 × *m*, 29-H_2), 4.72 (*t*, $|J_{AX}+J_{BX}|$ = 5.4 Hz, 3β-H), 5.15 (six-line pattern, J = 10.5 Hz, J' = 5.0 Hz, 11β-H).

11-*Monoacetate of compound* **4** (**8**). Compound **6** (259 mg) was deacetylated using 0.2 N NaOMe in MeOH (4.1 ml) for 1 hr at 20°. Acidification with HOAc and evapn and extraction of the residue with EtOAc gave, after CC, 114 mg **8**: amorphous; $[\alpha]_D^{20}$ −2.8° (*c* = 0.42 in EtOH); IR ν_{max}^{nujol} cm^{-1} 1640 (>C=CH_2), 1730–1740 (COOMe and OAc), 3380 (br OH); MS *m/z* (rel. int.): 544 $[M]^+$ (11), 484 $[M-HOAc]^+$ (68), 466 (19), 451 (11), 436 $[M-HOAc-H_2O-CH_2O]^+$ (19), 425 (36), 424 $[M-HOAc-HCO_2Me]^+$ (42), 407 (14), 402 (11), 393 (17), 377 (8), 320 $[\mathbf{d}]^+$ (13), 314 (27), 295 $[\mathbf{a}]^+$ (67), 260 $[\mathbf{d}-HOAc]^+$ (46), 255 (49), 247 $[\mathbf{b}-H]^+$ (66), 241 (38), 235 $[\mathbf{a}-HOAc]^+$ (63), 234 $[\mathbf{c}]^+$ (58), 217 (56), 205 (56), 201 (71), 187 (88), 175 (100), 161 (52), 147 (59), 133 (59), 121 (74), 107 (58). ^{1}H NMR ($CDCl_3$–HMDS): δ0.63, 0.93, 0.94, 0.99 (4 × *s*, 24-H_3, 25-H_3, 26-H_3, 27-H_3), 1.62 (*s*, 30-H_3), 1.90 (*s*, OAc), 3.29 and 3.48 (2 × *d*, J = 11.0 Hz, 23-H_2) 3.56 (*t*, $|J_{AX}+J_{BX}|$ = 5.4 Hz, 3β-H), 3.59 (*s*, COOMe), 4.51 and 4.67 (2 × *m*, 29-H_2), 5.13 (six-line pattern, J = 11.0 Hz, J' = 5.0 Hz, 11β-H).

Transformation of **8** *to* **11**. Acetylation of **8** (106 mg, 1 ml Ac_2O, 1 ml pyridine, 30 min, 20°) yielded, after CC, a mixture of **9** and **10** (60 mg), besides **6** (35 mg). The mixture of **9** and **10** (60 mg) was oxidized with PDC (151 mg) in DMF (1 ml) for 6 hr at 20°. Addition of saturated $NaHCO_3$ soln (10 ml) and extraction with EtOAc afforded the 3-oxo compound of **10** (21 mg) in the organic layer. Acidification of the aq. layer with dilute HCl and extraction with EtOAc gave the acid **11**. Evapn of the organic layer gave a residue which was dissolved in MeOH and methylated with an excess of ethereal CH_2N_2. The dimethyl ester diacetate (**12**) formed was chromatographed over silica gel (13 mg) and shown to be identical in all respects ($[\alpha]_D$, IR, MS, ^{1}H NMR) to authentic material [1].

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