Tetrahedron Vol. 47, No 38, pp 8195-8202, 1991
Printed in Great Britain

0040-4020/91 $3.00+.00
© 1991 Pergamon Press plc

Molvizarin and Motrilin: Two Novel Cytotoxic Bis-Tetrahydrofuranic γ-Lactone Acetogenins from *Annona Cherimolia* [1]

Diego CORTES*

Laboratoire de Pharmacognosie, Faculté de Médecine et Pharmacie, Université de Rouen, 76800 Saint Étienne du Rouvray, France.

Saw H. MYINT and Reynald HOCQUEMILLER

Laboratoire de Pharmacognosie, SDI 62330 CNRS, Faculté de Pharmacie, Université Paris-Sud, 92290 Châtenay-Malabry, France.

(*Received in Belgium* 8 *July* 1991)

Key Words: acetogenins; bis-tetrahydrofuranic γ-lactones; cytotoxicity; Annona cherimolia.

ABSTRACT: *Two new cytotoxic adjacent bis-tetrahydrofuranic acetogenins, molvizarin and motrilin, have been isolated from the cytotoxic methanolic extract of* Annona cherimolia *seeds. Their structures were established on the basis of 2D-NMR spectroscopic techniques. While molvizarin (1) belongs to the very rare type of C_{35} bis-tetrahydrofuranic acetogenins, motrilin (2) is the first example of a C-29 hydroxylated acetogenin.*

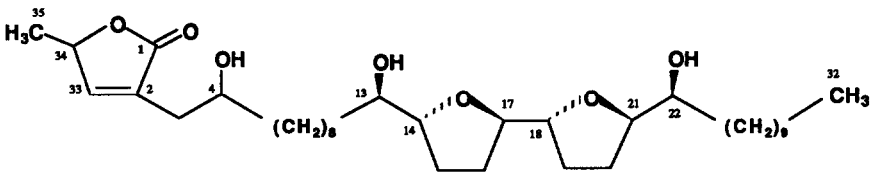
INTRODUCTION

Annona cherimolia (Annonaceae) is a tree native of tropical South America (Peru),[2] now cultivated for its edible fruits ("cherimoya") in a small pseudo-tropical area in the south of Spain: the "cherimoyo-vale"; this is a region with a moderated climate (10 to 28 °C), located between the Sierra Nevada and the Mediterranean coast in the province of Granada, inside the perimeter limit of the villages: Motril, Molvizar, Itrabo, Otívar, Jete, La Herradura and Almuñecar. *A. cherimolia* is used in traditional medicine as an insecticide and parasiticide.[3] Aporphinic alkaloids have been isolated from the seeds[4] and one of them, liriodenine, shows notable antimicrobial and cytotoxic activities;[5] in addition, five bis-tetrahydrofuranic (bis-THF) γ-lactone acetogenins have been obtained: cherimolin, dihydrocherimolin, laherradurin, asimicin and almunequin, which exhibit potent antimicrobial and antiparasitic activities.[4,6,7]

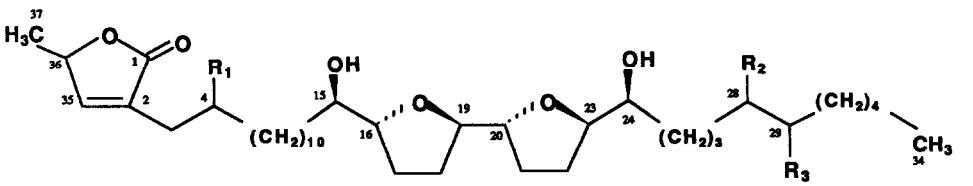
A new extraction of the *A. cherimolia* seeds led to the isolation of two novel bis-THF acetogenins exhibiting a cytotoxic activity: molvizarin (**1**) and motrilin (**2**).

RESULTS AND DISCUSSION

Screening of methanolic extract of the seeds of *A. cherimolia* showed cytotoxic and antiparasitic activities against *Entamoeba histolytica, Nippostrongylus brasiliensis, Molinema dessetae* and *Artemia salina*.[7] The cytotoxic methanolic extract was evaporated and the residue redissolved in ethyl acetate. The AcOEt-soluble portion was fractionated by flash chromatography. Four bioactive adjacent bis-THF γ-lactone acetogenins were isolated, two of which are new, molvizarin (**1**) and motrilin (**2**). The other two being the known compounds rolliniastatin-2 (**3**)[8] and squamocin (**4**).[9]



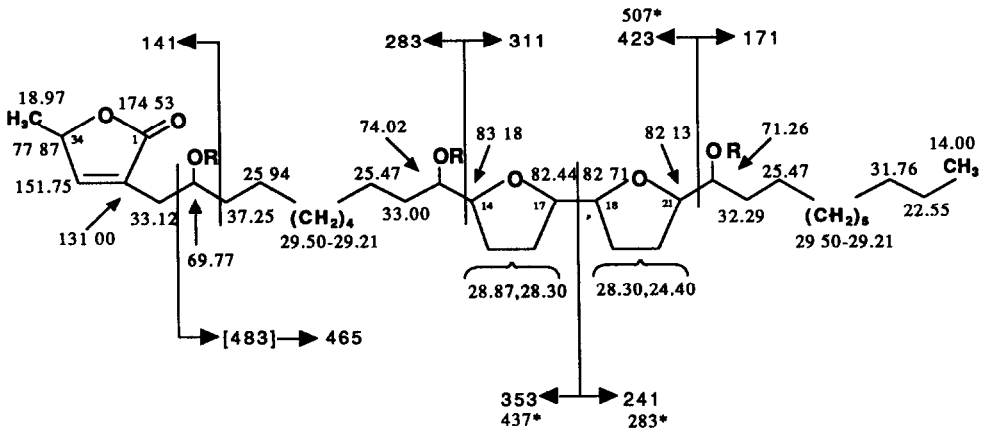
1: molvizarin



2: R_1= R_2= H; R_3= OH: motrilin
3: R_1= OH; R_2= R_3= H: rolliniastatin-2
4: R_1= R_3= H; R_2= OH: squamocin

Molvizarin (**1**) was obtained in an amorphous form. The molecular formula $C_{35}H_{62}O_7$ was determined by analysis of the CIMS (*m/z* 595, MH^+), and confirmed by the HRMS of its triacetate derivative (**1a**) (MH^+: *m/z* 721.4863, found; 721.4890, calcd. for $C_{41}H_{68}O_{10}$) indicating that **1** has three hydroxyl groups. The IR spectrum of **1** showed characteristic absorptions of α,β-unsaturated γ-lactone (1755 cm^{-1}, C=O) and hydroxyl groups (3420 cm^{-1}, OH). The protons of **1** were assigned by analysis of the 2D homodecoupling experiment (COSY 45, see Table 1). Examination of the ^1H NMR spectrum shows the typical aspect of the adjacent bis-THF γ-lactone acetogenins, isolated from Annonaceous species:[10,11] a) an α,β-unsaturated γ-methyl γ-lactone system (CH_3-35, H-34 and H-33); b) an ABX system corresponding to the 4-hydroxyl acetogenins class[12,13] (H-4 and two H-3); c) two multiplets belonging to six protons on oxygen-bearing carbons in an adjacent bis-THF moiety with the remaining two hydroxyl groups (H-13, H-14, H-17, H-18, H-21 and H-22); and d) multiplets of methylenic resonances and a methyl triplet, belonging to the alkyl chain, typical of the acetogenins.[11]

Figure 1. ^{13}C NMR data (50 MHz, $CDCl_3$, δ) of molvizarin (**1**); MS fragment ions of molvizarin, R=H (**1**) and molvizarin triacetate, R=Ac (**1a**) (*).



In the ^{13}C NMR spectrum of **1**, the multiplicities of the carbon atoms were determined by SPIN ECHO correlated spectroscopy.[14] The 2D ^{1}H-^{13}C heteronuclear correlation spectrum was carried out to obtain the relationship between the carbon atoms and their respective protons (see Table 1). The carbon resonances corresponding to 4-hydroxylated α,β-unsaturated γ-methyl γ-lactone moiety (Figure 1), are very similar to the "4,15,24-trihydroxylated bis-THF acetogenins type", for example, asimicin,[12] rolliniastatin-1[15] and rolliniastatin-2 (**3**).[8] However, the remaining methine signals associated with oxygen bearing carbon atoms (in the bis-THF α,α'-dihydroxylated system), coupled to typical proton multiplets at δ 3.84 and δ 3.37, are very similar to those of rolliniastatin-2 (**3**)[8], squamocin (**4**) (=annonin I)[9,11] and desacetyluvaricin[16] (relative configuration: *threo-trans-threo-trans-erythro* [17]); but are different to those of asimicin [12] (relative configuration: *threo-trans-threo-trans-threo* [17]) and rolliniastatin-1 (relative configuration: *threo-cis-threo-cis-threo* [15,17]). In addition, the typical downfield shift, in the ^{1}H NMR of molvizarin acetate (**1a**), of the geminal protons to the acetoxyl groups, as well as the resonances of two acetates adjacent to bis-THF system (δ 2.075 and δ 2.045), suggest[17] that the relative configuration for **1** at these six chiral centers is: *threo-trans-threo-trans-erythro*.

The MS fragmentations and the molecular formula of **1** (Figure 1) showed that molvizarin is an unusual C_{35} bis-THF acetogenin (bis-THF γ-lactone acetogenins contain usually C_{37} carbon atoms [11]); neoannonin is the only other example that belongs to this class.[18] The position of the adjacent bis-THF rings was confirmed by the cleavage at the α-positions of THF moieties: *m/z* 283 (base peak, C-13/C-14), *m/z* 353 (C-17/C-18) and *m/z* 423 (C-21/C-22) and corresponding losses of H_2O (Figure 1). Thus, molvizarin (**1**) is the first C_{35} bis-THF γ-lactone acetogenin with three hydroxyl groups in positions 4, 13 and 22.

Table 1. ^1H NMR, ^1H -^1H COSY 45 and ^1H -^{13}C heteronuclear correlation spectra of **1**.

H	δ (a)	Coupling in COSY-45 spectra	Coupling in ^1H-^{13}C spectra (multiplicity SPIN ECHO)
3a	2.50 *ddd*	H-3b (2.35), H-4 (3.84), H-33 (7.16)	33.12 (CH_2)
3b	2.35 *dd*	H-3a (2.50), H-4 (3.84)	33.12 (CH_2)
4	3.84 *m*	H-3a (2.50), H-3b (2.35), H-5 (1.46)	69.77 (CH)
5	1.46 *m*	H-4 (3.84)	37.25 (CH_2)
6	1.36 *m*		25.94 (CH_2)
7-10	1.25 *m*		29.50-29.21 (CH_2)
11	1.36 *m*		25.47 (CH_2)
12	1.36 *m*		33.00 (CH_2)
13	3.37 *m*	H-12 (1.36), H-14 (3.84)	74.02 (CH)
14	3.84 *m*	H-13 (3.37), H-15,16 (1.96, 1.62)	83.18 (CH)
15,16	1.96,1.62 *m*	H-14 (3.84), H-17 (3.84)	28.87, 28.30 (CH_2)
17	3.84 *m*	H-15,16 (1.96, 1.62)	82.44 (CH)
18	3.84 *m*	H-19,20 (1.96, 1.62)	82.71 (CH)
19,20	1.96,1.62 *m*	H-18 (3.84), H-21 (3.84)	28.30, 24.40 (CH_2)
21	3.84 *m*	H-19,20 (1.96, 1.62)	82.13 (CH)
22	3.84 *m*	H-23 (1.36)	71.26 (CH)
23	1.36 *m*	H-22 (3.84)	32.29(CH_2)
24	1.36 *m*		25.47 (CH_2)
25-29	1.25 *m*		29.50-29.21 (CH_2)
30	1.25 *m*		31.76 (CH_2)
31	1.25 *m*		22.55 (CH_2)
32	0.86 *t*	H-31 (1.25)	14.00 (CH_3)
33	7.16 *d*	H-34 (5.04), H-3a (2.50)	151.75 (CH)
34	5.04 *dq*	H-33 (7.16), CH_3-35 (1.42)	77.87 (CH)
35	1.42 *d*	H-34 (5.04)	18.97 (CH_3)

(a): $J_{3a\text{-}3b}$ = 16Hz; $J_{3a\text{-}4}$ = 3Hz; $J_{3b\text{-}4}$ = 8Hz; $J_{3a\text{-}33}$ < 1Hz; $J_{32\text{-}31}$= 7Hz; $J_{33\text{-}34}$ = 1.5Hz; $J_{34\text{-}35}$ = 7Hz

The molecular formula of motrilin (**2**), $C_{37}H_{66}O_7$, was deduced from analysis of the CIMS of **2** (*m/z* 623, MH^+), as well as that of motrilin acetate (**2a**) (*m/z* 749, MH^+) and confirmed by HRMS of its 2,35-dihydrogenated derivative (**2b**) (MH^+ *m/z* 625.5012, found; 625.5042, calcd. for $C_{37}H_{68}O_7$). The structure of the bis-THF acetogenin, **2**, was elucidated by two dimensional NMR spectroscopy: ^1H-^1H COSY 45 and ^1H-^{13}C (see Table 2 and Figure 2). The proton resonance at δ 2.26 (2H) attributed to H-3, coupled with the carbon signal at δ 25.05, has been attributed to acetogenins not hydroxylated in position 4.[9,12] The ^1H and ^{13}C NMR shifts of the bis-THF α,α'-dihydroxylated system for **2**, are very similar to those of molvizarin (**1**), rolliniastatin-2 (**3**) and squamocin (**4**); all these structures showed *threo-trans-threo-trans-erythro* relative configuration. **2** presents a multiplet (1H) at δ 3.55 coupled with a methine carbon signal at δ 71.65, characteristic of a hydroxyl group in the alkyl chain, while this is also observed for squamocin (=annonin I) (**4**) (HC-OH in position 28: δ 3.52 / δ 71.61);[9,11] this system is absent from molvizarin (**1**) and rolliniastatin-2 (**3**). The only difference in ^{13}C NMR between motrilin (**2**) and squamocin (**4**) concerns the chemical shifts of CH_2-26 (δ 25.53 for **2**, and δ 21.96 for **4**), and its neighbouring carbon atoms (see Figure 3); the upfield shift in the C-26 of **4** is due to the two β-effects of the hydroxyl-methine carbon atoms 24 and 28.

Figure 2. ^{13}C NMR data (50 MHz, $CDCl_3$, δ) of motrilin (2); MS fragment ions of motrilin, R=H (2), motrilin triacetate, R=Ac (2a) (*) and 2,35-dihydromotrilin, R=H (2b) (**).

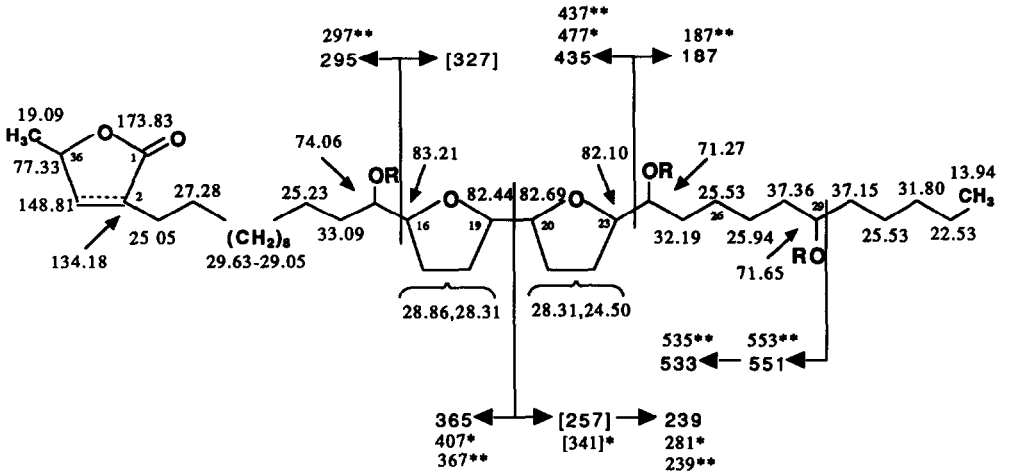
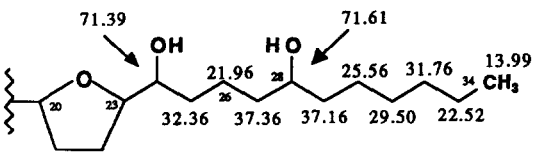


Figure 3. ^{13}C NMR (50 MHz, $CDCl_3$, δ) of the terminal dihydroxyalkyl chain of squamocin (4).



The chemical shifts obtained for **2** are consistent with the hydroxyl group placed in position 29 (Figure 2). This hypothesis is confirmed by the presence in CIMS of **2** and **2b**, of a cleavage between C-29 and C-30 (Figure 2). These results lead us to propose the structure **2** for motrilin, a bis-THF γ-lactone acetogenin with three hydroxyl groups in positions 15, 24 and 29, and a relative configuration *threo-trans-threo-trans-erythro* of the bis-THF α,α'-dihydroxylated system.

Up to now the bioactive bis-THF γ-lactone acetogenins have been isolated exclusively from Annonaceous species.[11] Molvizarin (**1**), motrilin (**2**), rolliniastatin-2 (**3**) and squamocin (**4**) isolated from *A. cherimolia* seeds, are trihydroxylated acetogenins with cytotoxic activities on KB and VERO cell culture systems (E.D. 50: 10^{-3} to 10^{-5} μg/ml in KB, human nasopharyngeal carcinoma cell, and E.D. 50: 10^{-2} to 10^{-3} μg/ml in VERO, monkey epithelialoid renal cell)[19,20] and to brine shrimp larvae (*Artemia salina*).[7]

Table 2. ^1H NMR, ^1H -^1H COSY 45 and ^1H -^{13}C heteronuclear correlation spectra of 2.

H	δ (a)	Coupling in COSY-45 spectra	Coupling in ^1H-^{13}C spectra (multiplicity SPIN ECHO)
3	2.26 *t*	H-4 (1.52), H-35 (6.97)	25.05 (CH_2)
4	1.52 *m*	H-3 (2.26), H-5 (1.26)	27.28 (CH_2)
5-12	1.26 *m*	H-4 (1.52)	29.63-29.05 (CH_2)
13	1.36 *m*		25.23 (CH_2)
14	1.38 *m*	H-15 (3.38)	33.09 (CH_2)
15	3.38 *m*	H-14 (1.38), H-16 (3.85)	74.06 (CH)
16	3.85 *m*	H-15 (3.38), H-17,18 (1.95,1.65)	83.21 (CH)
17,18	1.95,1.65 *m*	H-16 (3.85), H-19 (3.94)	28.86, 28.31 (CH_2)
19	3.94 *m*	H-17,18 (1.95,1.65)	82.44 (CH)
20	3.94 *m*	H-21,22 (1.95,1.65)	82.69 (CH)
21,22	1.95,1.65 *m*	H-20 (3.94), H-23 (3.85)	28.31, 24.50 (CH_2)
23	3.85 *m*	H-21,22 (1.95,1.65)	82.10 (CH)
24	3.88 *m*	H-25 (1.38)	71.27 (CH)
25	1.38 *m*	H-24 (3.88)	32.19(CH_2)
26	1.34 *m*		25.53 (CH_2)
27	1.34 *m*	H-28 (1.45)	25.94 (CH_2)
28	1.45 *m*	H-27 (1.34), H-29 (3.55)	37.36 (CH_2)
29	3.55 *m*	H-28 (1.45), H-30 (1.45)	71.65 (CH)
30	1.45 *m*	H-29 (3.55), H-31 (1.34)	37.15(CH_2)
31	1.34 *m*	H-30 (1.45)	25.53(CH_2)
32	1.26 *m*		31.80(CH_2)
33	1.32 *m*	CH_3-34 (0.89)	22.53 (CH_2)
34	0.89 *t*	H-33 (1.32)	13.94 (CH_3)
35	6.97 *d*	H-36 (4.98), H-3 (2.26)	148.81 (CH)
36	4.98 *dq*	H-35 (6.97), CH_3-37 (1.41)	77.33 (CH)
37	1.41 *d*	H-36 (4.98)	19.09 (CH_3)

(a): $J_{3\text{-}4}$ = 8Hz; $J_{3\text{-}35}$ < 1Hz; $J_{33\text{-}34}$= 7Hz; $J_{35\text{-}36}$ = 1.5Hz; $J_{36\text{-}37}$ = 7.5Hz

EXPERIMENTAL

General Methods. Optical rotations were measured with a Schmidt-Haensch Polartronic I. Uv spectra were obtained in EtOH on a Unicam 1800. Ir spectra were recorded in film on a Perkin-Elmer 257. The ^1H- and ^{13}C-NMR spectra (in deuteriochloroform solution) were obtained with a Bruker AC-200 at 200 and 50 MHz, respectively. EIMS and CIMS (CH_4 and NH_3) were performed on a Nermag-Sidar, and HRMS on a Kratos MS-80.

Plant material. The seeds of *A.cherimolia* were obtained from fruits collected in December 1989 in "cherimoyo-vale": Granada coast (Spain). A voucher specimen is deposited in the herbarium of the Department of Botany, University of Valencia (Spain), under the reference VF 10463.

Extraction and isolation. Extraction and fractionation were monitored by the brine shrimp test.[7] The dried and powdered seeds of *A. cherimolia* (1 kg) were macerated with methanol. The cytotoxic methanolic extract was evaporated, and the residue (50 g) was redissolved in ethyl acetate. The bioactive ethyl acetate-soluble extract (25 g), fractionated by flash chromatography (elution with CH_2Cl_2/AcOEt/MeOH 4:14:1), afforded four bioactive bis-THF γ-lactone acetogenins: molvizarin (100 mg, **1**), motrilin (35 mg, **2**), rolliniastatin-2 (300 mg, **3**) and squamocin (450 mg, **4**).

Molvizarin (**1**). Mp 36-38°C; $C_{35}H_{62}O_7$; $[\alpha]_D$ + 9.7° (MeOH; *c* 0.13); UV λ_{max} EtOH, nm (log ε) 215 (3.83); IR ν_{max} film cm^{-1} : 3420, 2920, 2845, 1755, 1460, 1315, 1195, 1055, 950, 750; CIMS (NH_3): *m/z*: 612 $(M + NH_4)^+$, 595 (MH^+); CIMS (CH_4), *m/z*: 595 (MH^+), 577 $(MH - H_2O)^+$, 559 $(MH - 2\ H_2O)^+$, 541 $(MH - 3\ H_2O)^+$, 465, 447, 423, 405, 387, 353, 335, 317, 311, 283 (100 %), 265, 247, 241, 223, 171, 141, see Figure 1; ^1H NMR: see Table 1; ^{13}C NMR: see Figure 1 and Table 1.

Molvizarin triacetate (**1a**). Treatment of **1** (10 mg) with acetic anhydride/pyridine (r.t., overnight) and subsequent work-up gave compound **1a** (quantitative yield) as an oil; IR ν_{max} film cm^{-1} : 1755, 1730; HRMS, *m/z* : 721.4863 $(MH^+$, found), 721.4890 (calcd. for $C_{41}H_{68}O_{10}$); CIMS (CH_4), *m/z*: 721 (MH^+), 661 (100 %, MH - AcOH)$^+$, 601 (MH - 2AcOH)$^+$, 541 (MH - 3AcOH)$^+$, 507, 465, 447, 437, 377, 317, 283, 223, see Figure 1; ^1H NMR δ: 0.88 (3H, *t*, *J*= 7 Hz, CH_3-32), 1.25-1.70 (34H, *m*, H-5 to -12 and H-23 to -31), 1.40 (3H, *d*, *J*= 7 Hz, CH_3-35), 1.55,1.95 (8H, 2*m*, H-15,16 and H-19,20), 2.025 (3H, *s*, $OCOCH_3$-4), 2.045 (3H, *s*, $OCOCH_3$-22), 2.075 (3H, *s*, $OCOCH_3$-13), 2.50 (2H, *m*, H-3), 3.86 and 3.97 (4H, 2*m*, H-17,18 and H-14,21), 4.84 (2H, *m*, H-13,22), 5.00 (1H, *dq*, *J*= 7 Hz, *J'*= 1.5 Hz, H-34), 5.09 (1H, *m*, H-4), 7.07 (1H, *d*, *J*= 1.5 Hz, H-33).

Motrilin (**2**). Mp 50-51°C; $C_{37}H_{66}O_7$; $[\alpha]_D$ + 10.8° (MeOH; *c* 0.13); UV λ_{max} EtOH, nm (log ε) 211 (4.07); IR ν_{max} film cm^{-1} : 3330, 2910, 2845, 1745, 1320, 1050, 945, 865, 750; CIMS (CH_4), *m/z*: 623 (MH^+), 605 $(MH - H_2O)^+$, 587 $(MH - 2\ H_2O)^+$, 569 $(MH - 3\ H_2O)^+$, 551, 533, 435, 417, 399, 365, 347, 309, 295 (100 %), 267, 239, 221, 187, 169, 151, 141, see Figure 2; ^1H NMR: see Table 2; ^{13}C NMR: see Figure 2 and Table 2.

Motrilin triacetate (**2a**). Treatment of **2** (5 mg) with acetic anhydride/pyridine (r.t., overnight) and subsequent work-up gave compound **2a** (quantitative yield) as an oil; IR ν_{max} film cm^{-1} : 1745, 1730; CIMS (CH_4), *m/z*: 749 (MH^+), 689 (100 %, MH - AcOH)$^+$, 629 (MH - 2AcOH)$^+$, 569 (MH - 3AcOH)$^+$; EIMS, *m/z*: 551, 533, 477, 417, 407, 347, 281, 221, see Figure 2; ^1H NMR δ: 0.86 (3H, *t*, *J*= 7 Hz, CH_3-34), 1.25-1.70 (38H, *m*, H-4 to -14; H-25 to -28; and H-30 to -33), 1.40 (3H, *d*, *J*= 6.5 Hz, CH_3-37), 1.52,1.95 (8H, 2*m*, H-17,18 and H-21,22), 2.025 (3H, *s*, $OCOCH_3$-29), 2.040 (3H, *s*, $OCOCH_3$-24), 2.070 (3H, *s*, $OCOCH_3$-15), 2.25 (2H, *t*, *J*= 7.5 Hz, H-3), 3.87 and 3.96 (4H, 2*m*, H-19,20 and H-16,23), 4.84 (3H, *m*, H-15,24,29), 4.97 (1H, *dq*, *J*= 6.5 Hz, *J'*= 1.2 Hz, H-36), 6.98 (1H, *d*, *J*= 1.2 Hz, H-35).

2,33-Dihydromotrilin (**2b**). Motrilin (**2**) (5 mg) was treated in methanolic solution with 10% Pd/C and H_2, at room temperature and the atmospheric pressure for 2 hours to afford **2b** (quantitative yield); HRMS (CI, CH_4), *m/z* : 625.5012$(MH^+$, found), 625.5042 (calcd. for $C_{37}H_{68}O_7$); CIMS (CH_4), *m/z*: 625 (MH^+), 607 $(MH - H_2O)^+$, 589 $(MH - 2H_2O)^+$, 571 $(MH - 3H_2O)^+$, 553, 535, 437, 419, 401, 367, 349, 297 (100 %), 239, 187, 169, 151, 141, see Figure 2; ^1H NMR δ: 0.88 (3H, *t*, *J*= 7 Hz, CH_3-34), 1.25-1.60 (40H, *m*, H-4 to -14; H-25 to -28; H-30 to -33; and H-36), 1.41 (3H, *d*, *J*= 6.5 Hz, CH_3-37), 1.59,1.95 (8H, 2*m*, H-17,18 and H-21,22), 2.40-2.63 (3H, *m*, H-2,3), 3.38 (1H, *m*, H-15), 3.56 (1H, *m*, H-29), 3.85 (5H, *m*, H-16,19,20,23, 24), 4.46 (1H, *m*, H-36).

ACKNOWLEDGEMENTS

This research was sponsored by the "Direction de la Recherche et des Etudes Doctorales (DRED)", through a biennial contract with the "Réseau de Recherche Pharmacochimie". One of us (S.H.M.) gratefully acknowledges the French government for a scholarship grant under the cultural exchange program between France and Myanmar (ex-Birmanie). We are grateful to J. Cotte and A.M. Quéro, Laboratoire de Virologie, for cytotoxicity results, and to A. Cavé and M. Leboeuf, Laboratoire de Pharmacognosie, for helpful discussions in this paper. We are especially grateful to Carmen Martínez, Nicolás Cortes, Herminia Moral, Felix Cortes, M. Carmen Cortes and José Antonio García, for the collection of "cherimoya" seeds in the tropical area of Granada (Spain), in autumn 1989.

REFERENCES

1. Part 12 in the series "Acétogénines des Annonacées". For part 11, see ref. 20.
2. Fries, R.E. "Annonaceae", in "Die Natürlichen Pflanzenfamilien", Engler, A. and Prantl, K. Eds., 2e ed., vol. 17, Dunker and Humbolt, Berlin, 1959.
3. Barriga, H.G. Flora Medicinal de Colombia, vol. 1, Botánica Médica, Bogotá, 1974, p. 340.
4. Rios, J.L.; Cortes, D.; Valverde, S. *Planta Med.* **1989**, *55*, 321-323.
5. Wu, Y.-C.; Chen, C.-H.; Yang, T.-H.; Lu, S.-T.; McPhail, D.R.; McPhail, A.T.; Lee, K.-H. *Phytochemistry* **1989**, *28*, 2191-2195.
6. Cortes, D.; Rios, J.L.; Villar, A.; Valverde, S. *Tetrahedron Lett.* **1984**, *25*, 3199-3202.
7. Bories, C.; Loiseau, P.; Cortes, D.; Myint, S. H.; Hocquemiller, R.; Gayral, P.; Cavé, A.; Laurens, A. *Planta Med.* **1991**, in press.
8. Pettit, G.R.; Riensen, R.; Leet, J.E.; Polonsky, J.; Smith, C.R.; Schmidt, J.M.; Dufresne, C.; Schaufelberger, D.; Moretti, C. *Heterocycles* **1989**, *28*, 213-217.
9. Fujimoto, Y.; Eguchi, T.; Kakinuma, K.; Ikekawa, N.; Sahai, M.; Gupta, Y.K. *Chem. Pharm Bull.* **1988**, *36*, 4802-4806.
10. Jolad, S.D.; Hoffmann, J.J.; Schram, K.H.; Cole, J.R.; Tempesta, M.S.; Kriek, G.R.; Bates, R.B. *J. Org. Chem.* **1982**, *47*, 3151-3153.
11. Rupprecht, J.K.; Hui, Y.-H.; McLaughlin, J.L. *J. Nat. Prod.* **1990**, *53*, 237-278.
12. Rupprecht, J.K.; Chang, C.-J.; Cassady, J.M.; McLaughlin, J.L.; Mikolajczak, K.L.; Weisleder, D. *Heterocycles* **1986**, *24*, 1197-1201.
13. Cortes, D.; Myint, S.H.; Laurens, A.; Hocquemiller, R.; Lebœuf, M.; Cavé, A. *Can. J. Chem.* **1991**, *69*, 8-11.
14. Le Cocq, C.; Lallemand, J.Y. *J. Chem. Soc., Chem. Commum.* **1981**, 150-152.
15. Pettit, G.R.; Cragg, G.M.; Polonsky, J.; Herald, D.L.; Goswami, A.; Smith, C.R.; Moretti, C.; Schmidt, J.M.; Weisleder, D. *Can. J. Chem.* **1987**, *65*, 1433-1435.
16. Jolad, S.D.; Hoffmann, J.J.; Cole, J.R.; Barry III, C.E.; Bates, R.B.; Linz, G.S.; Konig, W. *J. Nat. Prod.* **1985**, *48*, 644-645.
17. Hoye, T.R.; Zhuang, Z.P. *J. Org Chem.* **1988**, *53*, 5578-5580.
18. Kawazu, K.; Alcantara, J.P.; Kobayashi, A. *Agric. Biol Chem.* **1989**, *53*, 2719-2722.
19. Myint, S.H.; Laurens, A.; Hocquemiller, R.; Cavé, A.; Davoust, D.; Cortes, D. *Heterocycles* **1990**, *31*, 861-867.
20. Myint, S.H.; Cortes, D; Laurens, A.; Hocquemiller, R.; Leboeuf, M.; Cavé, A.; Cotte, J.; Quéro, A.-M. *Phytochemistry* **1991**, in press.