# New Sesquiterpenes from *Euonymus europaeus* (Celastraceae)

Charles Descoins Jr., Isabel López Bazzocchi,* and Angel Gutiérrez Ravelo

*Instituto Universitario de Bio-Orgánica Antonio González, Universidad de La Laguna, Avenida Astrofísica Francisco Sánchez 2, La Laguna 38206, Tenerife, Canary Islands, Spain.* Received August 15, 2001; accepted October 9, 2001

**A new sesquiterpene evoninate alkaloid (1), and two sesquiterpenes (2, 3) with a dihydro-$\beta$-agarofuran skeleton, along with three known sesquiterpenes (4—6), were isolated from the seeds of *Euonymus europaeus*. Their structures were elucidated by high resolution mass analysis, and one- and two-dimensional (1D and 2D) NMR spectroscopy, including homonuclear and heteronuclear correlation [correlation spectroscopy (COSY), rotating frame Overhauser enhancement spectroscopy (ROESY), heteronuclear single quantum coherence (HSQC), and heteronuclear multiple bond correlation (HMBC)] experiments.**

**Key words** *Euonymus europeaus*; Celastraceae; sesquiterpene alkaloid; dihydro-$\beta$-agarofuran sesquiterpene

Plants from the Celastraceae family have been used for centuries in South America, China and Africa in traditional medicine for the treatment of rheumatism, cancer and to protect plants against insect attacks (insecticidal and antifeedant properties).[1)] Sesquiterpene and alkaloid polyesters, based on the dihydro-$\beta$-agarofuran [5,11-epoxy-5$\beta$,10$\alpha$-eudesman-4(14)-ene] skeleton, are chemotaxonomic indicators of the family,[2)] and they have attracted a great deal of interest because of their cytotoxic,[3)] antitumor-promoting,[4)] reverse multidrug-resistance,[5)] antifeedant and insecticidal activities.[6)] Anti-human immunodeficiency virus (HIV)[7)] and immunosuppressive[8)] sesquiterpenes of this type have also been reported. Recently, the first enantioselective synthesis of a dihydroagarofuran triol (isocelorbicol) isolated from *Celastrus orbiculatus* was described.[9)]

*Euonymus europaeus* L., the sole representative of this genus in central and western Europe contains sesquiterpene polyesters belonging to the alatol, 3-deoxymaytol and 3,4-dideoxymaytol families,[10—13)] and alkaloid polyesters with the evoninate skeleton.[14—17)] Subsequent to an extensive reinvestigation of the sesquiterpene content of the seeds of *E. europaeus*, we report here on the isolation and structure elucidation of a evoninate alkaloid (**1**), and two dihydro-$\beta$-agarofuran polyesters (**2**, **3**), along with three known sesquiterpenes (**4**—**6**). Their structures were determined by high resolution-mass spectrometry (HR-MS), and application of spectroscopic techniques, including $^1$H–$^{13}$C heteronuclear correlation [heteronuclear single quantum coherence (HSQC)], long-range correlation with inverse detection [heteronuclear multiple bond correlation (HMBC)], and [rotating frame Overhauser enhancement spectroscopy (ROESY)] NMR experiments.

Repeated chromatography of the *n*-hexane–$Et_2O$ (1 : 1) extract of the seeds of *E. europaeus* on Sephadex LH-20 and Si gel yielded three new sesquiterpenes (**1**—**3**), along with the known compounds (**4**—**6**) (Chart 1).

Compound **1** was assigned the molecular formula $C_{32}H_{39}NO_{15}$ by HR-MS analysis, pointing to an alkaloidic structure. The IR spectrum showed a hydroxyl-type absorption band at 3450 cm$^{-1}$, ester carbonyl bands at 1750, 1730 cm$^{-1}$, and a ketone type at 1715 cm$^{-1}$. The presence of three hydroxyl groups was determined by chemical ionization (CI)-MS studies using $ND_3$ (*m/z* 682, 3 hydroxyls, $[MD]^+$, 100%).

The $^1$H-NMR spectrum (Table 1) showed three acetyl singlets at $\delta$ 1.98, 2.09 and 2.19; two methyl singlets at $\delta$ 1.58 and 1.87; five methine protons at $\delta$ 4.53 (d, $J$=3.2 Hz), $\delta$ 4.80 (m, overlapping signal), $\delta$ 5.17 (t, $J$=3.2 Hz), $\delta$ 5.34 (s) and $\delta$ 5.74 (s), and two sets of methylene protons at $\delta$ 4.29, 4.94 ($AB_q$, $J$=13.0 Hz, $H_{15}$) and $\delta$ 3.70, 6.08 ($AB_q$, $J$=12.3 Hz, $H_{12}$). On the basis of a $^1$H–$^1$H correlation spectroscopy (COSY) experiment, signals at $\delta$ 4.53, 5.17 and 4.80 were assigned to H-1, H-2 and H-3, respectively.

The presence of an evoninic acid moiety was determined by the signals of three aromatic protons corresponding to the 2,3-disubstituted pyridine unit at $\delta$ 8.70 (H-6′), $\delta$ 8.14 (H-4′), and $\delta$ 7.28 (H-5′) as double doublets, and by two secondary methyl doublets at $\delta$ 1.15 (Me-10′) and $\delta$ 1.41 (Me-9′) with the geminal protons at $\delta$ 4.80 (H-7′) and $\delta$ 2.43 (H-8′), respectively. The mass spectrum exhibited peaks consis-

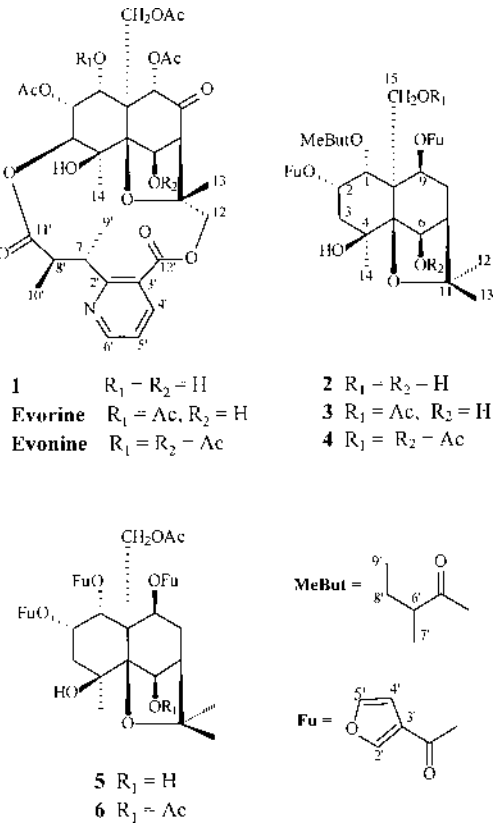


Chart 1. Structure of Compounds **1**—**6**

* To whom correspondence should be addressed. e-mail: ilopez@ull.es

Table 1. NMR Data for Compounds **1**—**4**

| | **1** | | **2** | | **3** | | **4** |
|---|---|---|---|---|---|---|---|
| | $\delta_H$ [a] | $\delta_C$ [b] | $\delta_H$ [a] | $\delta_C$ [b] | $\delta_H$ [a] | $\delta_C$ [b] | $\delta_H$ [a] |
| 1 | 4.53 (d, 3.2) | 71.4 (d) | 5.72 (d, 3.6) | 71.1 (d) | 5.68[c] (m) | 68.4 (d) | 5.65 (m) |
| 2 | 5.17 (t, 3.2) | 71.0 (d) | 5.65 (m) | 68.8 (d) | 5.68[c] (m) | 69.5 (d) | 5.65 (m) |
| 3 | 4.80[c] (m) | 74.5 (d) | | 41.0 (t) | 2.31 (m) | 41.6 (t) | |
| 4 | | 72.0 (s) | | 72.0 (s) | | 72.1 (s) | |
| 5 | | 94.0 (s) | | 91.1 (s) | | 91.0 (s) | |
| 6 | 5.34 (s) | 78.9 (d) | 4.84 (d, 5.4) | 78.6 (d) | 4.84 (d, 5.4) | 78.7 (d) | 6.13 (s) |
| 7 | 3.15 (s) | 63.8 (d) | | 50.1 (d) | | 50.1 (d) | |
| 8 | | 198.4 (s) | 2.24 (m) | 33.5 (t) | 2.31 (m) | 33.8 (t) | 2.56 (m) |
| 9 | 5.74 (s) | 76.0 (d) | 5.56 (d, 7.0) | 68.1 (d) | 5.56 (d, 7.0) | 68.2 (d) | 5.27 (d, 6.9) |
| 10 | | 52.5 (s) | | 55.0 (s) | | 53.7 (s) | |
| 11 | | 86.0 (s) | | 84.7 (s) | | 84.9 (s) | |
| 12 | 3.70, 6.08 (d, 12.3) | 21.1 (t) | 1.51 (s) | 26.5 (q) | 1.54 (s) | 26.4 (q) | 1.49 (s) |
| 13 | 1.58 (s) | 71.0 (q) | 1.62 (s) | 30.0 (q) | 1.61 (s) | 29.7 (q) | 1.56 (s) |
| 14 | 1.87 (s) | 24.0 (q) | 1.77 (s) | 24.8 (q) | 1.78 (s) | 24.8 (q) | 1.78 (s) |
| 15 | 4.29, 4.94 (ABq, 13.0) | 65.0 (t) | 4.11, 4.52 (ABq, 12.0) | 64.0 (t) | 4.52, 5.10 (ABq, 13.0) | 66.0 (t) | 4.42, 5.21 (ABq, 12.9) |

*a*) $\delta$, $CDCl_3$, *J* values in Hertz. *b*) Data are based on DEPT and HSQC experiments. *c*) Overlapping signals.

tent with losses of hydroxyl group *m/z* 661 ($M^+-18$), acetic acid *m/z* 617 ($M^+-CH_3COOH$), and fragments at *m/z* 206 ($C_{10}H_{12}NO_3$), *m/z* 178 ($C_9H_{12}NO_2$) and *m/z* 160 ($C_9H_{10}NO$), corresponding to an evoninate moiety.

The $^{13}$C-NMR spectrum (Table 1) confirmed the presence of five carboxyl carbons at $\delta$ 173.0, 170.4, 170.1, 169.7 and 168.8, and five aromatic carbons of the 2,3-disubstituted pyridine at $\delta$ 165.4 (C-2′), 152.0 (C-6′), 138.4 (C-4′), 124.0 (C-3′), and 121.0 (C-5′); also were observed four quaternary carbons at $\delta$ 52.5, 72.0, 86.0 and 94.0, and a carbonyl group at $\delta$ 198.4. All these data indicate that **1** was a sesquiterpene pyridine alkaloid with a dihydro-$\beta$-agarofuran skeleton.

The regiosubstitution of two of the acetate groups was deduced unambiguously from an HMBC experiment (Fig. 1), which showed three-bond coupling between the carboxyl signals of the acetate groups and the signals at $\delta_H$ 5.74 (H-9) and 4.29, 4.94 (H-15), while the carbonyl group was sited at C-8 as the signal at $\delta_C$ 198.4 was correlated with the signals at $\delta_H$ 3.15 (H-7) and 5.74 (H-9). The remaining acetyl group was located at C-2 as the signal at $\delta_H$ 5.17 (H-2) was coupled in the COSY experiment with that at $\delta_H$ 4.80 (H-3), which in its turn showed long-range correlation with the carboxyl group at $\delta_C$ 173.0 (C-11′). A ROESY experiment showing nuclear Overhauser effect (NOE) effects between H-1 and H-2 and H-9; between Me-14 and H-3, H-6 and H-15, and finally between H-9 and Me-13, enabled the relative position of the substituent groups to be determined. All these data and comparison with those given in the literature for evorine and evonine[16] (Chart 1) revealed that **1** was 1,6-*bis*-deacetylevonine, and had not been previously described. This was confirmed by acetylation of **1** (see Experimental part).

Compounds **2** and **3** have the molecular formula $C_{30}H_{38}O_{12}$ and $C_{32}H_{40}O_{13}$, respectively, by analysis of the HR-MS data (*m/z* 590.2349 and 632.2443), and they exhibited common hydroxyl and ester-type absorption bands in their IR spectra.

The $^1$H-NMR spectrum of **2** (Table 1) showed one group of two broad singlets ($\delta$ 8.06, 8.00, each 1H) and two groups of double doublets ($\delta$ 7.47, 7.41, 6.47, 6.29, each 1H), which were attributed to the protons of two furancarbonyloxy substituents; one doublet ($\delta$ 0.83, 3H), one triplet ($\delta$ 0.61, 3H),

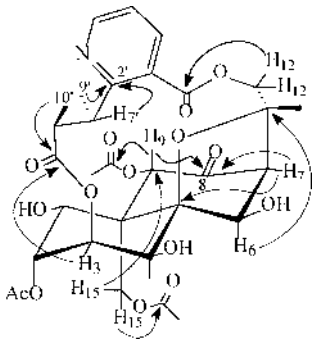


Fig. 1. $^1$H–$^{13}$C (HMBC) Couplings for **1**

and two multiplets ($\delta$ 1.98, 1H, $\delta$ 1.25, 2H) were assigned to a 2-methylbutanoyloxy group; all these data were confirmed by the $^{13}$C-NMR spectrum (Table 1). In addition, the $^1$H-NMR spectrum contained signals assignable to protons on carbon atoms carrying three secondary ester groups at $\delta$ 5.72 (d, $J$=3.6 Hz, H-1), $\delta$ 5.65 (m, H-2), and $\delta$ 5.56 (d, $J$=7.0 Hz, H-9); one primary hydroxyl group at $\delta$ 4.11, 4.52 ($AB_q$, $J$=12.0 Hz, H-15); one secondary hydroxyl group at $\delta$ 4.84 (d, $J$=5.4 Hz, H-6); a tertiary methyl group a $\delta$ 1.77 (s, Me-14) attached to a carbon at $\delta$ 72.0 bearing a hydroxyl group, and two angular methyl groups at $\delta$ 1.15 and 1.62.

All these data and two-dimensional (2D) experiments (COSY, HSQC, ROESY) indicated that **2** is a dihydro-$\beta$-agarofuran sesquiterpene with two furancarbonyloxy, one 2-methylbutanoyloxy, and three hydroxyl groups, positioned at 1$\alpha$, 2$\alpha$, 4$\beta$, 6$\beta$, 9$\alpha$, 15. The regiosubstitution characteristics were deduced from an HMBC experiment, showing three-bond coupling between the carboxyl signals of the furoate groups at $\delta_C$ 161.9 and 161.7, and the signals at $\delta_H$ 5.65 (H-2) and 5.56 (H-9), and between the carboxyl signal of the 2-methylbutyrate group at $\delta_C$ 174.1 and the signal at $\delta_H$ 5.72 (H-1). Thus, the proposed structure for **2** was 2$\alpha$,9$\beta$-di-($\beta$-furancarbonyloxy)-4$\beta$,6$\beta$-15-trihydroxy-1$\alpha$-(2)-methylbutanoyloxy-dihydro-$\beta$-agarofuran.

The data for compounds **3** and **4** were very similar to those of **2**, with the most notable differences being in their $^1$H-

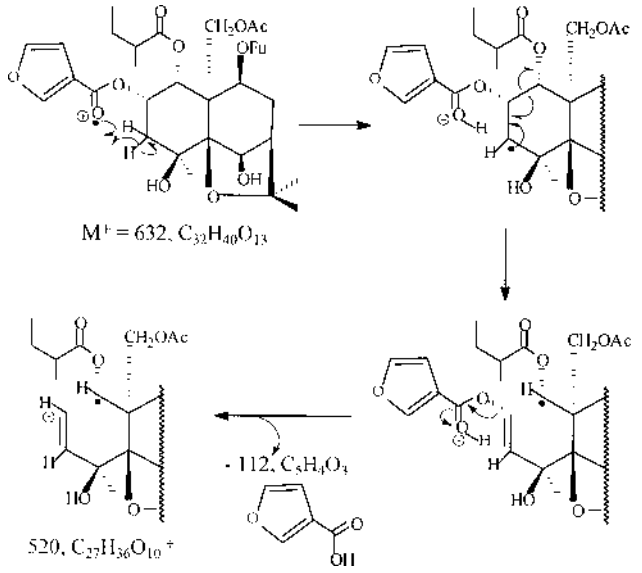


Chart 2. Mass Fragmentation of Compound **3**

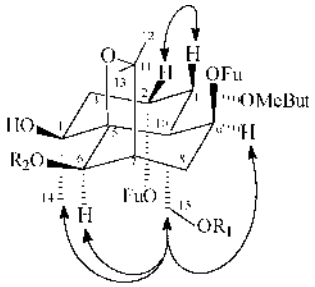


Fig. 2. ROESY Experiment of Compounds **2** and **3**

NMR spectra (Table 1), the presence of one acetyl singlet at $\delta$ 2.25, and the high field displacement of the AB system for the H-15 protons in **3**, the presence of two acetyl singlets at $\delta$ 2.22 and 2.10, and the high field displacement of the H-15 and H-6 protons, for **4**. Thus, **2**—**4** have in common one methylbutanoyloxy, and two furancarbonyloxy substituents, but differ in the number of acetate groups.

The location of the different substituents was realized by comparison of the MS data of **2**—**4** with those of two agarofurans, previously isolated from the seeds of *E. bungeanus* by Tu *et al.*[18] In case of a furancarbonyloxy (FuOH) group at C-2 and a 2-methylbutanoyloxy (MeBuOH) group at C-1, the MS spectrum shows, as described by the authors, an intense signal at *m/z* 502, corresponding to the loss of the FuOH and acetate groups, and another one of weak intensity at *m/z* 512 due to the loss of the MeBuOH and acetate groups. This difference is due to a favourable McLafferty rearrangement between the proton H-3 of the decalinic ring and the carboxyl of the FuOH group in C-2. However, the presence of FuOH and MeBuOH groups at C-1 and C-2, respectively, shows the same type of fragmentation although with inverse intensities.

In our case the MS spectrum of **4** matches the data of one of the compounds described by Tu and co-workers.[18] The MS spectrum of compound **3** presents an abundant fragment at *m/z* 520 (72%), corresponding to the favourable loss of the furancarbonyloxy group located at C-2 (Chart 2). In its $^1$H-NMR spectrum, the disappearance of the acetate singlet ($\delta$ 2.10) and upfield displacement of the geminal proton H-6 from a singlet at $\delta$ 6.13 in **4** to a doublet at $\delta$ 4.84 ($J$=5.4 Hz), coupled with a proton interchangeable with $D_2O$ in **3**, confirm the presence of a hydroxyl at C-6. These data together with $^1$H–$^{13}$C correlations (HSQC, HMBC) and a ROESY experiment (Fig. 2) allowed us to establish the structure of **3** as 15-acetoxy-2$\alpha$,9$\beta$-di-($\beta$-furancarbonyloxy)-4$\beta$,6$\beta$-dihydroxy-1$\alpha$-(2)-methylbutanoyloxy-dihydro-$\beta$-agarofuran. On the other hand, compounds **2** and **3** by acetylation gave **4**, which demonstrated that these three compounds are structurally related.

Compounds **5** and **6**, previously described in *E. bungeanus*[18] and *E. sieboldanus*,[19] respectively, are reported here for the first time in *E. europaeus*.

## Experimental

**General Experimental Procedures** Optical rotations were measured on a Perkin-Elmer 241 automatic polarimeter and $[\alpha]_D^{25}$ are given in $10^{-1}$ deg·cm$^2$·g$^{-1}$. UV spectra were collected on a Perkin-Elmer model 550-SE. IR spectra were recorded on a Nicolet Avatar. $^1$H, $^{13}$C, HMBC, HSQC and ROESY NMR spectra were recorded on a Bruker Avance 400 (400 MHz) in $CDCl_3$ with tetramethylsilane (TMS) as internal reference. MS were recorded on a MS-VG Micromass LTD-ZAB-2F and/or an HP 5930 A at 70 eV. CI-MS spectra were recorded on a Nermag R30-10 either by direct introduction or through a gas chromatograph (Varian GX 3000), and HR-MS spectra on a Finnigan MAT95Q or a VG Autospec.

**Plant Material** Seeds of *E. europaeus* were collected in November, 1997 along the railroad track from Bailly to St-Cyr-l'Ecole (France) and a voucher specimen (n° 19962707701-CD) has been deposited in the INRA herbarium. Dried and powdered seeds (750 g) were extracted with $EtO_2$–*n*-hexane (1 : 1, 3 l) in a Soxhlet apparatus. Removal of the solvent under reduced pressure left a reddish-brown oil (300 g). Repeated chromatography of this crude extract on Sephadex LH-20 (*n*-hexane–$CHCl_3$–MeOH, 2 : 1 : 1) and Si gel ($\phi$ 0.20 to 0.63 mm, mixtures of *n*-hexane–EtOAc of increasing polarity), followed by preparative TLC (Shleicher-Schull F-100/LS254, $CH_2Cl_2$–acetone, 8.5 : 1.5) yield compound **1** (7 mg, *Rf*=0.35), and by preparative high performance thin-layer chromatography (HPTLC) (HPTLC-Platten-SIL 20 $UV_{254}$, Panreac, *n*-hexane–1,4-dioxan, 3 : 7), compounds **2** (15 mg, *Rf*=0.37), **3** (5 mg, *Rf*=0.42), **4** (12 mg, *Rf*=0.49), **5** (3 mg, *Rf*=0.40), and **6** (4 mg, *Rf*=0.47), were obtained.

**1,6-Bis-deacetyl Evonine (1)** Colorless oil, $[\alpha]_D^{25}$ +20.0° ($c$=0.13, $CHCl_3$). UV $\lambda_{max}$ (MeOH) nm: 263, 230, 229. IR ($CHCl_3$) $\nu_{max}$ cm$^{-1}$: 3450, 2900, 2850, 1750, 1730, 1715, 1560 , 1455, 1210, 1110. $^1$H-NMR ($CDCl_3$) $\delta$: 1.15 (3H, d, $J$=7.0 Hz, H-10′), 1.41 (3H, d, $J$=7.0 Hz, H-9′), 1.98 (3H, s, Ac-2), 2.09 (3H, s, Ac-9), 2.19 (3H, s, Ac-15), 2.43 (1H, q, $J$=7 Hz, H-8′), 4.80 (2H, m, H-3 and H-7′, overlapping signals), 7.28 (1H, dd, $J$=7.8, 4.8 Hz, H-5′), 8.14 (1H, dd, $J$=7.8, 1.7 Hz, H-4′), 8.70 (1H, dd, $J$=4.8, 1.7 Hz, H-6′), for other signals, see Table 1. $^{13}$C-NMR ($CDCl_3$) $\delta$: 9.7 (q, C-10′), 11.5 (q, C-9′), 20.6, 20.4, 20.0 (s, 3×$CH_3$, OAc), 36.0 (d, C-7′), 43.3 (d, C-8′), 121.0 (d, C-5′), 124.0 (s, C-3′), 138.4 (d, C-4′), 152.0 (d, C-6′), 165.4.0 (s, C-2′), 168.8 (s, C-12′), 173.0 (s, C-11′), 170.4, 170.1, 169.7 (s, 3×C=O, OAc), for other signals, see Table 1. MS *m/z*: 677 ($M^+$) (100), 649 (8), 238 (10), 220 (35), 206 (evonic acid$^+$, 60), 178 (35), 149 (43), 107 (60); HR-MS *m/z*: 677.2320 (Calcd for $C_{32}H_{39}O_{15}N$: 677.2310). CI-MS ($NH_3$) *m/z*: 678 $(MH)^+$ (100), 660 ($MH^+-H_2O$) (14), 206 (33). CI-MS ($ND_3$) *m/z*: 682 $(MD)^+$ (100).

**Acetylation of 1** $Ac_2O$ (4 drops) was added to compound **1** (2 mg) dissolved in pyridine (2 drops), and the mixture left at room temperature for 16 h. EtOH (3×2 ml) was added and carried almost to dryness in a rotavapor, and this process was repeated with $CHCl_3$ (3×2 ml), to give evonine[16] (2 mg).

**2$\alpha$,9$\beta$-Di-($\beta$-furancarbonyloxy)-4$\beta$,6$\beta$,15-trihydroxy-1$\alpha$-(2)-methylbutanoyloxy-dihydro-$\beta$-agarofuran (2)** Colorless oil, $[\alpha]_D^{25}$ +30.0° ($c$= 0.13, $CHCl_3$); UV $\lambda_{max}$ (MeOH) nm: 231, 221. IR ($CHCl_3$) $\nu_{max}$ cm$^{-1}$: 3431, 2934, 1729, 1646, 1574, 1311, 1161, 1135, 874, 760. $^1$H-NMR ($CDCl_3$) $\delta$: 0.61 (3H, t, $J$=7.5 Hz, H-9′), 0.83 (3H, d, $J$=7.1 Hz, H-7′), 1.25 (2H, m, H-8′), 1.98 (1H, m, H,6′), 3.27 (1H, s, $C_4$-OH), 4.98 (1H, d, $J$=5.8 Hz, $C_6$-OH), 6.29 (1H, dd, $J$=0.7, 1.5 Hz, H-4″), 6.47 (1H, dd, $J$=0.7, 1.5 Hz, H-4′), 7.41 (1H, dd, $J$=1.5, 3.1 Hz, H-5″),7.47 (1H, dd, $J$=1.5, 3.1 Hz, H-5′), 8.00 (1H, br s, H-2″),8.06 (1H, br s, H-2′), for other signals, see Table 1. $^{13}$C-NMR ($CDCl_3$) $\delta$: 11.2 (q, C9′), 15.9 (q, C7′), 25.6 (t, C8′), 40.9 (d, C6′),

109.9, 109.5 (d, 2×C4′), 118.9 (s, 2×C-3′), 144.4, 143.7 (d, 2×C-5′), 148.8, 147.8 (d, 2×C-2′), 161.9, 161.7 (s, 2×C=O, FuOH), 174.1 (s, C=O, MeBuOH), for other signals, see Table 1. MS *m/z*: 575 ($M^+-15$) (6), 478 ($M^+-FuOH$) (2), 460 ($M^+-FuOH-H_2O$) (8), 398 (3), 348 (3), 264 (4), 249 (6), 231 (10), 189 (5), 105 (13), 95 (100), 85 (17), 57 (35). HR-MS *m/z*: 590.2363 (Calcd for $C_{30}H_{38}O_{12}$: 590.2349), 575.2128 ($M^+-15$) (Calcd for $C_{25}H_{35}O_{12}$: 575.2163).

**Acetylation of 2** $Ac_2O$ (4 drops) was added to compound **2** (2 mg) dissolved in pyridine (2 drops), and the mixture left at room temperature for 16 h. EtOH (3×2 ml) was added and carried almost to dryness in a rotavapor, and this process was repeated with $CHCl_3$ (3×2 ml), to give **4** (2 mg).

**15-Acetoxy, 2α,9β-Di-(β-furancarbonyloxy)-4β,6β-dihydroxy-1α-(2-methyl butanoyloxy)-dihydro-β-agarofuran (3)** Colorless oil, $[\alpha]_D^{25}$ +39.7° (*c*=0.39, $CHCl_3$). UV $\lambda_{max}$ (MeOH) nm: 234, 221. IR ($CHCl_3$) $\nu_{max}$ $cm^{-1}$: 3500, 2931, 1735, 1508, 1310, 1235, 1160, 1134, 874, 762. $^1$H-NMR ($CDCl_3$) $\delta$: 0.82 (3H, d, *J*=7.0 Hz, H-7′), 1.24 (3H, m, H-8′ and H-6′, overlapping signals), 2.06 (1H, m, H-6′), 2.25 (3H, s, Ac-15), 3.28 (1H, s, OH-4), 5.03 (1H, d, *J*=5 Hz, OH-6), 6.75 (1H, dd, *J*=1.0, 1.5 Hz, H-4′), 6.89 (1H, dd, *J*=1.0, 1.5 Hz, H-4′), 7.45 (1H, dd, *J*=1.0, 1.5 Hz, H-5′), 7.50 (1H, dd, *J*=1.0, 1.5 Hz, H-5′), 8.07 (1H, br s, H-2″), 8.31 (1H, br s, H-2′), for other signals, see Table 1. $^{13}$C-NMR ($CDCl_3$) $\delta$: 11.3 (q, C-7′), 16.0 (q, C-9′), 21.5 (q, $CH_3$, Ac-15), 25.5 (t, C-8′), 40.8 (d, C-6′), 108.9, 109.6 (d, 2×C-4′), 128.7, 118.7 (s, 2×C-3′), 144.3, 143.8 (d, 2×C-5′), 148.8, 148.5 (d, 2×C-2′), 161.7 (s, C=O, FuOH), 170.9 (s, C=O, OAc), 174.5 (s, C=O, MeBuOH), for other signals, see Table 1. MS *m/z*: 632 ($M^+$) (18), 617 ($M^+-15$) (40), 512 ($M^+-MeBuOH-H_2O$) (2), 520 ($M^+-FuOH$) (72), 502 ($M^+-FuOH-H_2O$) (18), 449 ($M^+-FuOH-C_4H_7O$) (83), 390 ($M^+-2FuOH-H_2O$) (18), 287 (16), 192 (17), 175 (10), 165 (20), 112 (10), 102 (3), 95 (100). HR-MS *m/z*: 632.2469 (Calcd for $C_{32}H_{40}O_{13}$: 632.2443). CI-MS ($NH_3$) *m/z*: 650 $(M+NH_4)^+$ (100), 633 $(MH)^+$ (40), 615 $(MH^+-H_2O)$ (49). CI-MS ($ND_3$) *m/z*: 656 $(M+ND4)^+$ (100), 636 $(MD)^+$ (3).

**Acetylation of 3** $Ac_2O$ (4 drops) was added to compound **3** (2 mg) dissolved in pyridine (2 drops), and the mixture left at room temperature for 16 h. EtOH (3×2 ml) was added and carried almost to dryness in a rotavapor, and this process was repeated with $CHCl_3$ (3×2 ml), to give **4** (2 mg).

**Acknowledgements** We are indebted to the DGES Project BQU2000-0870-CO2-01 and the FEDER Project 1FD1997-0747-CO4-01 for financial assistance.

**References**

1) González A. G., Bazzocchi I. L., Moujir L., Jiménez I. A., Ravelo A. G., "Studies in Natural Products Chemistry," Vol. 23, ed. by Atta-Ur-Rahman, Elsevier, The Netherlands, 2000, pp. 649—738.
2) Brüning R., Wagner H., *Phytochemistry*, **7**, 1821—1858 (1978).
3) Kuo Y.-H., King M.-L., Chen C.-F., Chen C.-H., Chen K., Lee K.-H., *J. Nat. Prod*., **57**, 263—269 (1994).
4) González A. G., Tincusi B. M., Bazzocchi I. L., Tokuda H., Nishino H., Konoshima T., Jiménez I. A., Ravelo A. G., *Bioorg. Med. Chem*., **8**, 1773—1778 (2000).
5) Pérez-Victoria J. M., Tincusi B. M., Jiménez I. A., Bazzocchi I. L., Gupta M. P., Castanys S., Gamarro F., Ravelo A. G., *J. Med. Chem*., **42**, 4388—4393 (1999).
6) González A. G., Jiménez I. A., Ravelo A. G., Coll J., González J. A., Lloria J., *Biochem. System. Ecol*., **25**, 513—519 (1997).
7) Duan H., Takaishi Y., Imakura Y., Jia Y., Li D., Cosentino L. M., Lee K.-H., *J. Nat. Prod*., **63**, 357—361 (2000).
8) Duan H., Takaishi Y., Momota H., Ohmoto Y., Taki T., Jia Y., Li D., *J. Nat. Prod*., **64**, 582—587 (2001).
9) Li W.-D., Zhou G., Gao X., Li Y., *Tetrahedron Lett*., **42**, 4649—4651 (2001).
10) Budzikiewicz H., Römer A., *Tetrahedron*, **31**, 1761—1767 (1975).
11) Römer A., Thomas H., Budzikiewicz H. Z., *Naturforsch. Teil B*, **31**, 607—610 (1976).
12) Römer A., Thomas H., Kreuels R., Budzikiewicz H. Z., *Naturforsch. Teil B*, **36**, 379—381 (1981).
13) Rozsa Z., Pelczer I., *J. Chem. Soc. Perkin Trans. 1*, **1989**, 1089—1095.
14) Doebel K., Reichstein T., *Helv. Chim. Acta*, **32**, 592—597 (1949).
15) Pailer M., Libiseller T., *Monatsh. Chem*., **1962**, 93.
16) Klasek A., Santavy F., Duffield A. M., Reichstein T., *Helv. Chim. Acta*, **54**, 2144—2158 (1971).
17) Klasek K., Samek Z., Santavy F., *Tetrahedron Lett*., **10**, 941—944 (1972).
18) Tu Y. Q., Wu D. G., Zhou J., Chen Y. Z., Pay X. F., *J. Nat. Prod*., **53**, 603—608 (1990).
19) Ujita K., Takaishi Y., Iida A., Fujita T., *Phytochemistry*, **31**, 1289—1292 (1992).