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Diterpene constituents of leaves from *Juniperus brevifolia*

Ana M.L. Seca [a], Artur M.S. Silva [b,*], Isabel L. Bazzocchi [c], Ignacio A. Jimenez [c]

[a] *Department of Technologic Sciences and Development, University of Azores, Rua Mãe de Deus, 9501-801 Ponta Delgada, Azores, Portugal*
[b] *Department of Chemistry, University of Aveiro, Campus de Santiago, 3810-193 Aveiro, Portugal*
[c] *Instituto Universitario de Bio-Orgânica ''António González'', Universidad de La Laguna, Av. Astrofisico Francisco Sanchez, 2, 38206 La Laguna, Tenerife, Spain*

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Abstract

The dichloromethane extract from leaves of *Juniperus brevifolia*, through chromatographic fractionations yield six compounds: 3β-hydroxy-abieta-8,11,13-trien-7-one, 18-hydroxy-sandaracopimara-8(14),15-dien-7-one, sandaracopimara-8(14),15-dien-18-yl formate; and the first examples of sandaracopimaranes and abieta-8,11,13-triene diterpenoids with a large aliphatic chain on C-18, abieta-8,11,13-trien-18-yl hexadecanoate, 7-oxoabieta-8,11,13-trien-18-yl hexadecanoate, sandaracopimara-8(14),15-dien-18-yl hexadecanoate. Moreover fifteen known compounds were also isolated, some of them for the first time identified on *Juniperus* genus. The compound abieta-8,11,13-trien-18-yl formate is reported for the first time as a natural product. All the structures were established by spectroscopic methods. 2D NMR techniques have allowed the revision of certain previously reported ^{13}C NMR assignments. Studies on the isolated new compounds showed those possessing a diterpenol ester of a long-chain fatty acid present lipophilicity very distinct from other diterpenoid compounds.

Keywords: *Juniperus brevifolia*; Cupressaceae; Cedro do mato; Abietanes; Sadaracopimaranes; Fatty acid diterpenol ester

1. Introduction

Being isolated in the middle of the Atlantic Ocean and having several natural resources, Azores becomes very interesting as a source of possible new bioactive compounds and/or with chemosystematic significance. One of these sources may be the *Juniperus brevifolia* (Seub.) Antoine (Cupressaceae), locally known as ''cedro-do-mato'' and well known for its durability and resistance to rotting. This species is the unique conifer tree endemic of Azores and it is a typical component of the primitive *laurisilva* forest (Schäfer, 2002). No evidence has been found for the use of *J. brevifolia* in traditional medicine. However, the wide range of biological activities reported for other species of this genus as well as for their constituents (Seca and Silva, 2006) stimulated our interest to study the chemical composition of *J. brevifolia*. Previous studies on this plant described the components of its essential oil (Adams, 1999; Da Silva et al., 2000) and hexane extract (Seca and Silva, in press). We report herein on the isolation and structural elucidation of six new diterpenes and fifteen known compounds from the dichloromethane extract of *J. brevifolia* leaves and on the correction of some literature ^{13}C NMR assignments for abieta-8,11,13-trien-18-yl formate.

2. Results and discussion

The analysis of the dichloromethane extract of the leaves of *J. brevifolia* led to the isolation of three new abietanes (**1**–**3**) and three new pimaranes (**4**–**6**); three of them being esters of the long-chain fatty hexadecanoic acid and other ester of formic acid (Fig. 1). Moreover, fifteen known compounds were also identified, by comparison

* Corresponding author. Tel.: +351 234 370714; fax: +351 234 370084.
E-mail address: artur.silva@ua.pt (A.M.S. Silva).

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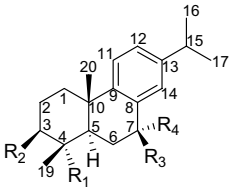
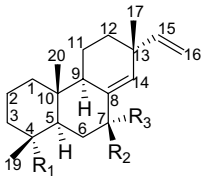


	R_1	R_2	R_3	R_4
1	$CH_2OCO(CH_2)_{14}CH_3$	H	H	H
2	$CH_2OCO(CH_2)_{14}CH_3$	H	=O	
3	CH_3	OH	=O	
7	CH_2OCHO	H	H	H



	R_1	R_2	R_3
4	CH_2OCHO	H	H
5	$CH_2OCO(CH_2)_{14}CH_3$	H	H
6	CH_2OH	=O	

Fig. 1. Structures of compounds isolated from *Juniperus brevifolia* leaves.

their spectra data with those reported in the literature, as 15,16-bisnor-13-oxo-labda-8(17),11*E*-dien-19-oic acid (**8**) (Inoue et al., 1985; Muhammad et al., 1996), *E*- and *Z*-communic acid (**9**) (Muhammad et al., 1995), hinokiol (**10**) (Fang et al., 1996; Wang et al., 2002), 18-hydroxy-dehydroabietane (pomiferin A) (**11**) (San Feliciano et al., 1992; Fraga et al., 2003), sugiol (**12**) (Ara et al., 1988; Fang et al., 1993), sandaracopimara-8(14),15-dien-18-ol (**13**) (San Feliciano et al., 1992; Barrero et al., 2004), sandaracopimaric acid (**14**) (Wenkert and Buckwalter, 1972; Sakar and San Feliciano, 1994), nootkatone (**15**) (Miyazawa et al., 2000; Schneider et al., 2004), stigma-4-en-3-one (**16**), β-sitosterol (**17**) (Seca et al., 2000); and for the first time in *Juniperus* species, sandaracopimara-8(14),15-diene (**18**) (Kenmoku et al., 2004), methyl ester of 15-agathic acid (**19**) (Richomme et al., 1991), 7-oxo-abieta-8,11,13-trien-18-ol (**20**) (Tanaka et al., 1997) and eicosanyl-*trans*-*p*-coumarate (**21**) (Mahmood et al., 2003). Abieta-8,11,13-trien-18-yl formate (**7**) is a synthetic dehydroabietane derivative with gastroprotective effect (Sepúlveda et al., 2005) which is identified for the first time as a natural product. A detailed analysis of the COSY, HSQC and HMBC spectra of **7** have shown that the literature assignment resonances of C-4 and C-10 (Sepúlveda et al., 2005) must be interchanged. In fact, the HMBC spectrum of **7** showed correlations between the proton resonance at δ 7.18 (1H, *d*, $J = 8.2$ Hz, H-11) with the quaternary carbons at δ_C 37.3 (C-10), 134.6 (C-8) and 145.7 (C-13) and also between that of the methyl group at δ_H 0.96 (3H, *s*, H-19) with the carbons at δ_C 71.8 (C-18), 43.9 (C-5), 36.7 (C-4) and 35.4 (C-3). These data allowed the unequivocal assignment of C-4 and C-10 which proves the previous assignments must be interchanged.

The HR-ESIMS data of **1** exhibited a sodiated molecular ion peak at *m*/*z* 547.4463 establishing the molecular formula $C_{36}H_{60}O_2$. The IR spectrum showed absorption bands at ν_{max} 1738, 1166 and 2924 cm^{-1}, suggesting the presence of an ester group of an aliphatic long-chain acid. This was also supported by the resonance at δ_C 174.1 and a large number of signals between δ_C 29.2 and 29.7 in its ^{13}C NMR spectrum. The ^1H NMR spectrum of **1** showed the presence of three protons in a trisubstituted aromatic ring at δ_H 7.18 (1H, *d*, $J = 8.2$ Hz), 7.00 (1H, *dd*, $J = 8.2$, 1.8 Hz) and 6.89 (1H, *brs*), two quaternary methyl groups at δ_H 1.22 and 0.93, and one hydroxymethylene group at δ_H 3.96 and 3.69 (AB system, 1H each, *d*, $J = 11.0$ Hz). The presence of an isopropyl group linked to a quaternary carbon was supported by the signals at δ_H 2.83 (1H, *sept*, $J = 6.9$ Hz) and 1.23 (6H, *d*, $J = 6.9$ Hz). Comparison of the ^{13}C NMR data of **1** and those of abieta-8,11,13-trien-18-yl formate (**7**) showed a good agreement except for the chemical shift of C-1′ (shifted downfield, $\Delta\delta$ +12.9 ppm, due to the substitution of H–COO to RCOO group) and the presence of one methyl and fourteen methylene additional groups (confirmed by ^{13}C DEPT NMR spectra). The presence of a long-chain aliphatic acid esterified with the dehydroabietane derivative on C-18 was confirmed by the important connectivities found in the HMBC spectrum (as shown in Fig. 2) and the NOE cross peaks observed in the NOESY spectrum between the signal at δ_H 0.93 (3H, *s*, H-19) with that at 1.22 (3H, *s*, H-20). The equatorial position of the hydroxymethylene group was confirmed by the shift of the 4-*C*H_3 at δ_C 17.5 corresponding to axial position (Chamy et al., 1987; Ulubelen and Topcu, 1992). All of these data established the structure of **1** (Fig. 1) as abieta-8,11,13-trien-18-yl hexadecanoate.

Compound **2** was shown to have the molecular formula $C_{36}H_{58}O_3$ according to the HR-ESIMS quasi-molecular ion peak observed at *m*/*z* 539.4446 $[M+H]^+$. Its IR spectrum revealed absorptions for two types of carbonyl groups (ν_{max} 1683 and 1737 cm^{-1}). The ^{13}C NMR spectrum (Table 1) was similar to those of **1**, being the presence of a carbonyl group (δ_C 199.0), lack of one methylene group and the deshielding effect on C-6 (from δ_C 18.9 to 36.0) the major differences. The downfield shift in the resonances of the 1,2,4-trisubstituted aromatic ring (δ_H 7.31, *d*, $J = 8.2$ Hz, H-11; 7.42, *dd*, $J = 2.1$ and 8.2 Hz, H-12; 7.88, *d*, $J = 2.1$ Hz, H-14) of **2** compared to those of **1** are the main differences in their ^1H NMR spectra. These data were compatible with the presence of a carbonyl group at C-7 conjugated with the aromatic ring which resulted in a mesomeric deshielding effect on H-12, and H-14, and mainly anisotropic deshielding effect on H-14 and H-6. This was confirmed by the connectivities found in the HMBC spectrum (Fig. 2). The stereochemistry of **2** was established based on chemical shift resonances of the 4-methyl group

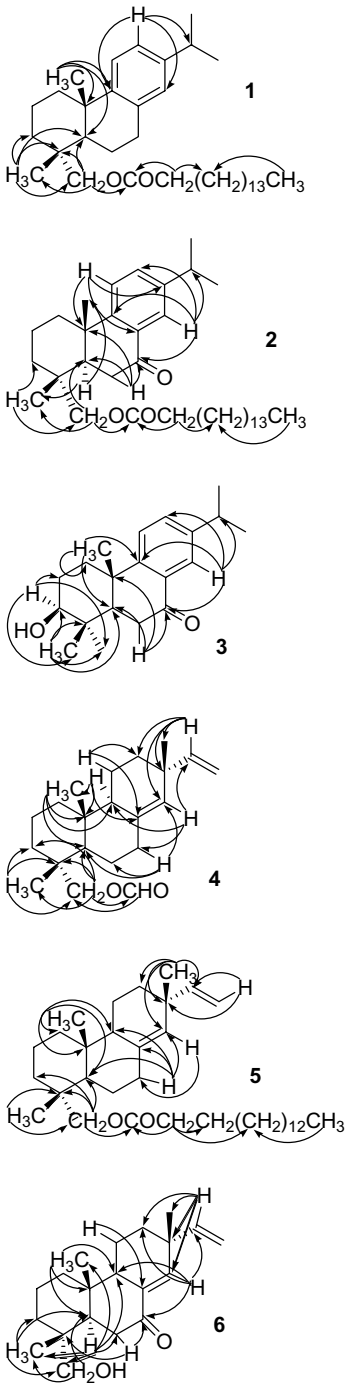


Fig. 2. Major HMBC connectivities observed for new compounds.

(δ_C 17.3), and a NOESY experiment, showing that the axial methyl protons at C-19 gave cross peaks with the 20-methyl protons and the equatorial hydroxymethylene protons giving cross peak with H-5. From the described spectroscopic observations, one can conclude that compound **2** is 7-oxo-abieta-8,11,13-trien-18-yl hexadecanoate (Fig. 1) identified here for the first time.

Spectroscopic data of **3** showed that it is related to **2** (see Table 1 and Section 3), being the absence of the long-chain alkyl ester the most notable difference, suggested by the

Table 1
^{13}C NMR spectroscopic data for **1–6** in $CDCl_3$

C	1	2	3	4	5	6
1	38.2	37.4	36.2	38.7	38.8	38.3
2	18.2	18.1	27.5	18.1	18.2	18.0
3	35.5	35.3	78.1	35.8	36.0	35.1
4	37.4	36.6	38.8	36.6	36.7	35.7
5	44.2	43.3	48.5	48.4	48.5	42.7
6	18.9	36.0	35.8	22.5	22.5	37.0
7	30.2	199.0	199.5	35.5	35.6	200.7
8	134.7	130.6	130.4	136.6	136.7	135.2
9	147.1	153.3	152.9	50.5	50.6	50.9
10	36.7	37.6	37.6	38.1	38.1	37.6
11	124.3	123.6	123.9	18.7	18.7	18.9
12	123.9	132.6	132.6	34.5	34.5	33.9
13	145.6	146.8	146.9	37.4	37.4	38.6
14	126.8	125.0	124.9	128.9	128.8	144.5
15	33.4	33.6	33.6	149.0	149.0	146.3
16	24.0	23.8	23.8	110.1	110.0	111.8
17	24.0	23.8	23.8	25.9	26.0	25.8
18	72.2	71.4	27.5	72.4	72.6	70.8
19	17.5	17.3	14.9	17.8	17.9	17.1
20	25.4	23.9	23.4	15.5	15.5	14.4
1′	174.1	173.8	–	161.3	174.0	–
2′	34.4	34.3	–	–	34.5	–
3′	25.1	24.9	–	–	25.1	–
4′–13′	29.7–29.2	29.7–29.1	–	–	29.7–29.2	–
14′	31.9	31.9	–	–	31.9	–
15′	22.7	22.7	–	–	22.7	–
16′	14.1	14.1	–	–	14.1	–

disappearance of the great number of signals at δ_C ca. 29, assigned to the methylene groups, and that at δ_C 173.8 due to the carbonyl ester group. The HR-EIMS of **3** showed a $[M]^+$ ion at *m/z* 300.2094 consistent with the molecular formula $C_{20}H_{28}O_2$. The peaks at *m/z* 285 ($[M-15]^+$), 267 ($[M-33]^+$) and 199 ($[M-101]^+$) are characteristics of a 3-hydroxy-abieta-8,11,13-triene diterpene (Enzell and Ryhage, 1967). The structure of **3** was also supported by the presence of another 4-methyl group and a C-3 hydroxymethine group (δ_C 78.1, δ_H 3.35, 1H, *dd*, $J = 4.5$, 11.4 Hz), confirmed by ^{13}C DEPT-135 NMR spectrum, instead of a hydroxymethylene (δ_C 71.4) and methylene groups in **2**. The coupling constant of the signal due to H-3 suggests their α-axial orientation as reported for hinokiol (Wang et al., 2002), which was confirmed by comparison with the spectral data of its 3α-OH isomer previously synthesised (Burnell et al., 1993). The stereochemistry of chiral carbon C-10 and of the C-4 methyl substituents were assigned by NOE effects between δ_H 3.35 (1H, *dd*, $J = 4.5$, 11.4 Hz, H-3) and the protons at δ_H 1.05 (3H, *s*, H-18) and 1.84–1.90 (3H, *m*, H-5 and H-2), and between δ_H 0.97 (3H, *s*, H-19) and 1.24 (3H, *s*, H-20), found in the NOESY spectrum of **3**. All these spectral data and the connectivities found in the HMBC spectrum (Fig. 2) are only compatible with the structure of 3β-hydroxy-abieta-8,11,13-trien-7-one **3**.

Compound **4** had the molecular formula $C_{21}H_{32}O_2$, as inferred from molecular ion peak on HR-EIMS. The peaks at *m/z* 121 and 135 are frequently found on sandaracopimara-8(14),15-diene diterpenes (Audier et al., 1966). The

peaks at *m/z* 288, 257, 255 and 241 are correspondents to the fragment ions $[M-28]^+$, $[M-Me-44]^+$, $[M-Me-46]^+$ and $[M-Me-CH_2OCHO]^+$ (Audier et al., 1966) and were an evidence for the presence of a C-4 hydroxymethyl group esterified with formic acid. The presence of an ester group and a double bond was inferred by the absorptions found in the FTIR spectrum of **4** appearing at ν_{max} 1728, 1634, and 1167 cm^{-1}. The ^1H, ^{13}C and DEPT NMR spectra showed signals due to the protons of three methyl groups, appearing as singlets at δ_H 0.84, 0.89 and 1.04, an hydroxymethylene group (δ_C 72.4; δ_H 3.75 and 3.99, *d*, $J = 10.9$ Hz), an ABX spin system corresponding to a vinyl moiety on a quaternary carbon (δ_H 4.88, *dd*, $J = 1.5$, 10.6 Hz; δ_H 4.91, *dd*, $J = 1.5$, 17.4 Hz; δ_H 5.77, *dd*, $J = 10.6$, $J = 17.4$ Hz), and an olefinic proton (δ_H 5.22, *brs*, H-14). These NMR data suggests a pimarane skeleton for compound **4** (San Feliciano et al., 1992; Chang et al., 2000), and are similar to those of sandaracopimara-8(14),15-dien-18-ol **13** (San Feliciano et al., 1992). The assignment of the olefinic proton at C-14 and the formyl group (δ_C 161.3; δ_H 8.11, *s*) attached to the hydroxymethylene carbon at C-18 were determined from the correlations observed on the HMBC experiment (Fig. 2). The identification of **4** as belonging to the sandaracopimarane series was based on the literature (Cambie et al., 1975; Chang et al., 2000), being the C-17 carbon resonance nearly δ_C 26 an indication of the axial 13-methyl group in opposition of that at δ_C ca. 30 which is indicative of an equatorial orientation. The configuration of C-4, C-5, C-9 and C-10 were assigned on the basis of NOESY experiment. Thus, the signal due to the axial methyl protons at δ_H 0.84 (H-20) gave cross peaks with those of the methyl protons at δ_H 0.89 (H-19) and δ_H 1.04 (H-17), while that of H-5 gave intense cross peak with H-9. Moreover, the NOE effect between δ_H 1.04 (H-17) and δ_H 5.22 (H-14) strongly supports the axial orientation of the 13-methyl group. The carbon chemical shift of the 4-methyl group (δ_C 17.9) confirms its β-position. Accordingly, compound **4** is a new sandaracopimarane derivative identified as sandaracopimara-8(14),15-dien-18-yl formate.

Compound **5** was identified as sandaracopimara-8(14),15-dien-18-yl hexadecanoate, since their ^1H and ^{13}C NMR data were similar to those of compounds **4** and **13**. The differences are limited to the absence of the signals at δ_H 8.11 (formyl proton resonance), the appearance of a strong broad singlet at δ_H ca. 1.25 due to a great number of protons and a triplet at δ_H 2.32. The singlet at δ_H ca. 1.25 correlates with the signals of a large number of carbon (at δ_C ca. 29 ppm), as observed in the HSQC spectrum, and were assigned to methylene carbons and confirmed by a ^{13}C DEPT-135 NMR experiment. The triplet at δ_H 2.32 was also assign to a methylene group but the resonance of this carbon was shifted downfield to δ_C 34.5. These proton and carbon resonances suggest, like in compound **1** and **2**, the presence of a long aliphatic chain. The HMBC connectivities (see Fig. 2) allowed the identification of compound **5** as an ester formed from a long-chain fatty acid and the sandaracopimara-8(14),15-dien-18-ol. The HR-ESIMS exhibited a quasi-molecular ion $[M+H]^+$ at *m/z* 527.4846 establishing a molecular formula of $C_{36}H_{62}O_2$ to compound **5**, and allowed the identification of the aliphatic chain as an hexadecanoic acid derivative. The stereochemistry of the chiral carbons C-4, C-5, C-10 and C-13 were assigned based on the chemical shifts of the 4- and 13-methyl groups (as explained to compound **4**) and on the NOE effects observed on a NOESY spectrum: e.g. H-5 gave cross peak with the hydroxymethylene proton, while NOE cross peaks were also observed between H-17 and the protons H-20 and H-14. The configuration of C-9 was assumed to be the same as all the compounds isolated from *J. brevifolia* (H-9 as α-proton) because of the co-occurrence in the plant.

The molecular formula $C_{20}H_{30}O_2$ of compound **6** was deduced from its molecular ion peak in the HR-EIMS spectrum, which confirms a diterpene skeleton. The peaks at *m/z* 121 and 133 supports a sandaracopimara-8(14),15-diene diterpene structure and the fragments $[M-Me-H_2O]^+$ and $[M-CH_2OH]^+$ at *m/z* 269 and 271, respectively (Audier et al., 1966), supports the presence of a hydroxymethyl group. The ^1H, ^{13}C and DEPT NMR spectra of **6** are similar to those of **13** except the presence of a carbonyl group (δ 200.7) and lack of one methylene group. The ^1H and ^{13}C NMR spectra of **6** differs from that of **4**, **5** and **13** in the resonance of C-14 and of an olefinic proton which shifted from δ_C ca. 128 to 144.5 and from δ_H 5.22 to 6.72, respectively, and suggests a structural feature of a carbonyl group conjugated with a double bond. The anisotropic deshielding effect of the carbonyl group also justify the downfield shift of the methylene protons adjacent to the carbonyl group (δ_H 2.21, *dd*, $J = 13.6$, 18.6 Hz, and δ_H 2.49, *dd*, $J = 5.2$, 18.6 Hz). The resonances at δ_H 3.09, 3.37 (2*d*, $J = 10.9$ Hz) and δ_C 70.8 support the presence of a secondary alcohol group. The presence of the alcohol and of the α,β-unsaturated carbonyl system in the structure **6** was confirmed by the absorption bands at 3449 and 1676, 1607 cm^{-1} in its FTIR spectrum. The detailed analysis of the 2D NMR experiments of **6** allowed the unequivocal assignment of all proton and carbon resonances (Fig. 2). The stereochemistry of **6** was deduced by analogy with those of **4** and **5**, by opposition of its isomer 19-hydroxy-sandaracopimara-8(14),15-dien-7-one (Yang et al., 1998), and supported by the chemical shifts of the substituents on chiral carbons and by the NOE effects observed in their NOESY spectrum: (i) between H-20 (δ_H 0.87) and H-17 (δ_H 1.10); (ii) between H-5 (δ_H 1.86) and H-9 (δ_H 2.10); (iii) H-17 (δ_H 1.10) and H-14 (δ_H 6.72) and (iv) between H-6α (δ_H 2.49) and H-18 (δ_H 3.37). The structure **6** was fully characterised and identified as 18-hydroxy-sandaracopimara-8(14),15-dien-7-one.

The communic acid (*E* and *Z* isomers **9**), sandaracopimaric acid (**14**) and sugiol (**12**) were the major diterpene constituents of the dichloromethane extract of the leaves of *J. brevifolia*. This result agrees with literature data where these three compounds are the most frequently reported (Seca

and Silva, 2006). The occurrence of sugiol, and other phenolic abietanes, have a chemosystematic significance since they are indicated as chemosystematic markers of Cupressaceae family (Otto and Wilde, 2001), while sandaracopimaric and communic acids occurring in all conifer families.

It is known that lipophilicity of the compounds play an important role in several biological processes as absorption, metabolism, distribution as well as toxicity and it is correlating with cytotoxicity (Maliepaard et al., 1992; Bajda et al., 2007). The new compounds described above **1**–**6** showed significant differences in lipophilicity (from 3.92 to **6** to major than 12.09 to **5**) suggesting differences in their function, biolocalization and biosynthetic role, particularly for **1**, **2** and **5**. The isolation of diterpene fatty acid esters is very unusual. The literature report the isolation of 7α-docosanyl- and 7α-stearylhorminone from *Salvia lanigera* (Aboul-Ela, 2006) and the isolation of a mixture of fatty acid esters of 7α-acyloxy-6β-hydroxyroylleanone and their biological activities (Teixeira et al., 1997; Marques et al., 2002; Cerqueira et al., 2004). Compounds **1**, **2** and **5** represent the first examples of diterpenes possessing an esterified fatty acid at C-18. These findings encourage further pharmacological studies of their bioactivities, mechanism of action and interaction with the biomembranes.

3. Experimental

3.1. General experimental procedures

NMR experiments were performed on either Bruker Avance 300 or Avance 500 spectrometers. EIMS were obtained at 70 eV electron impact ionisation using a VG Autospec M mass spectrometer and ESI-MS at positive mode in a Q-TOF 2 instruments. IR spectra were obtained with a MATTSON 7020 FTIR spectrometer with KBr pellets. Prep. TLC was carried out on silica gel (Merck silica gel 60 F_{254}) and CC on silica gel 60, 70–230 mesh, and sephadex LH-20; spots were visualized by heating silica gel plates sprayed with CH_3COOH:H_2O:H_2SO_4 (80:16:4) or under UV lamp (at 254 and/or 366 nm). The lipophilicity of the new compounds was calculated using Chemoffice 2004 version 8.0 software. The parameter is presented as log *P*.

3.2. Plant material

Leaves of *J. brevifolia* were harvested in Pedreira, São Miguel, Azores, in June of 2001 and voucher specimen was identified by Prof Eduardo Dias of University of Azores where a voucher was deposited under the reference DCTD-2001/Jb/01.

3.3. Extraction and isolation

Powdered leaves (1239 g) were extracted with CH_2Cl_2 using a Soxhlet apparatus. Evaporation to dryness gave 108.6 g of residue which was subject to column chromatography (CC) on silica gel and eluted with gradient systems of increasing polarity hexane:EtOAc (0–100%) and finally MeOH. 50 Fractions were collected and combined on the basis of their TLC profiles to afford 19 main fractions 1–19, among which fractions 8 (6.9 g), 9 (5.4 g) and 13 (5.7 g) reveals to be interesting from the point of view of terpene analysis when sprayed with an appropriate reagent mixture (see Section 3.1), and were purified by sephadex CC (hexane:$CHCl_3$:MeOH, 2:1:1). The semi-purified fr. 8.1 (2.6 g) was rechromatographed on sephadex CC (hexane:$CHCl_3$:MeOH, 2:1:1) to give fr. 8.1.1–8.1.4. Fr 8.1.2 was submitted to prep. TLC (hexane:Et_2O, 6:4) to give **16** (2 mg) and **17** (4.1 mg). Fr. 8.2 (590 mg) was chromatographed on silica gel CC (hexane:EtOAc of increasing polarity, 2.5–15%) to give fr. 8.2.1–8.2.8. Fr. 8.2.2 was purified by prep. TLC (hexane:*iso*-PrOH, 6.5%) to give **21** (1.5 mg). Fr. 8.2.3 was purified by prep. TLC (hexane:*iso*-PrOH, 93.5:6.5) to give **13** (11.3 mg) and **15** (3 mg). Fr. 8.2.7 was purified by silica gel CC (hexane:EtOAc, 88:12) to give **18** (3 mg) and **17** (5 mg). Fr. 8.3 (213 mg) was chromatographed on prep. TLC (hexane:dioxane, 9:1, 3×) to give 2 impure fr. (8.3.1–8.3.2) and the pure compound **20** (17 mg). Fr. 8.3.1 was submitted to prep. TLC (hexane:Et_2O, 35:1, 4×) to give **1** (7.1 mg), **4** (5.7 mg), **5** (18 mg) and **7** (3.2 mg). Compound **2** (2 mg) and **15** (1.7 mg) were isolated from fr. 8.3.2 after purification by prep. TLC (CH_2Cl_2:$CHCl_3$, 9:1). Fr. 8.4 (113 mg) was chromatographed on prep. TLC (hexane:dioxane, 9:1, 3×) to give 8.4.1–8.4.4. Fr. 8.4.3 was purified by prep. TLC (CH_2Cl_2:EtOAc, 9:1) to give **14** (12.4 mg). Fr. 8.4.4 was submitted to prep. TLC (CH_2Cl_2:EtOAc, 9:1, 2×) to give **3** (1.8 mg), **6** (5 mg) and **20** (4 mg). Fr. 8.5 (2.1 g) was chromatographed on silica gel CC (hexane:Et_2O of increasing polarity, 0–100%) to give fr. 8.5.1–8.5.5. The mixture of isomers *E* and *Z* of **9** (264 mg) was obtained from fr. 8.5.1 by crystallisation from hexane. Compound **12** (54 mg) was obtained from fr. 8.5.3 by crystallization in EtOH. Fr. 8.5.4 (300 mg) was chromatographed on silica gel CC (hexane:Et_2O of increasing polarity, 5–25%) to give **13** (25 mg), **14** (51 mg), one impure fr., which after purification by prep. TLC (hexane:Et_2O, 7:3) gave **11** (37.8 mg) and other impure fr. that gave **19** (10 mg) after purification by prep. TLC (hexane:Et_2O, 41:9). Fr. 8.6 (650 mg) was chromatographed on prep. TLC (CH_2Cl_2:EtOAc, 3:2) to give **12** (10 mg), the mixture of isomers *E* and *Z* of **9** (14 mg) and **14** (9.7 mg). Fr. 9.3 was chromatographed on prep. TLC (CH_2Cl_2, 4×) to give **8** (11 mg). Compound **10** (3.8 mg) was obtained from fr. 13.4 after purification by prep. TLC (CH_2Cl_2:Et_2O, 9:1, 3×).

3.4. Abieta-8,11,13-trien-18-yl hexadecanoate (*1*)

Yellowish oil, IR ν_{max} (KBr) cm^{-1}: 2924 and 2853 (C–H alkanes), 1738 (>C=O), 1497 (C=C), 1166 (C–O). ^1H NMR spectral data (300 MHz, $CDCl_3$): δ 0.87 (3H, *t*,

$J = 6.9$ Hz, H-16′), 0.93 (3H, *s*, H-19), 1.22 (3H, *s*, H-20), 1.23 (6H, *d*, $J = 6.9$ Hz, H-16 and H-17), 1.21–1.35 (22H, *m*, H-4′ to H-14′), 1.35–1.44 (3H, *m*, H-3 and H-1), 1.58–1.61 (2H, *m*, H-3′), 1.61–1.67 (1H, *m*, H-5), 1.67–1.70 (2H, *m*, H-2), 1.71–1.77 (2H, *m*, H-6), 2.25–2.32 (1H, *m*, H-1), 2.27 (2H, *t*, $J = 7.5$ Hz, H-2′), 2.83 (1H, *sept*, $J = 6.9$ Hz, H-15), 2.80–2.87 (2H, *m*, H-7), 3.69 (1H, *d*, $J = 11.0$ Hz, H-18), 3.96 (1H, *d*, $J = 11.0$ Hz, H-18), 6.89 (1H, *brs*, H-14), 7.00 (1H, *dd*, $J = 1.7$, 8.2 Hz, H-12), 7.18 (1H, *d*, $J = 8.2$ Hz, H-11). ^{13}C NMR spectral data (75 MHz, $CDCl_3$): see Table 1. Positive HR-ESIMS *m/z*: 547.4463, $[M+Na]^+$ (calculated for $C_{36}H_{60}NaO_2$: 547.4485).

3.5. 7-Oxo-abieta-8,11,13-trien-18-yl hexadecanoate (**2**)

Yellowish oil, IR ν_{max} (KBr) cm^{-1}: 2926 and 2853 (C–H alkanes), 1737 (>C=O ester), 1683 (C=O ketone), 1604 (C=C aromatic), 1460, 1379, 1251, 1164 (C–O ester). ^{1}H NMR spectral data (500 MHz, $CDCl_3$): δ 0.88 (3H, *t*, $J = 6.8$ Hz, H-16′), 1.02 (3H, *s*, H-19), 1.25 (3H, *s*, H-20), 1.26 (6H, *d*, $J = 6.8$ Hz, H-16 and H-17), 1.22–1.33 (22H, *m*, H-4′ to H15′), 1.46–1.48 (2H, *m*, H-3), 1.55–1.69 (2H, *m*, H-1 and H-3′), 2.20 (1H, *dd*, $J_{5\alpha,6\alpha} = 7.3$ Hz, $J_{5\alpha,6\beta} = 10.7$ Hz, H-5), 2.26 (2H, *t*, $J = 7.5$ Hz, H-2′), 2.35 (1H, *dt*, $J = 2.9$, 12.7 Hz, H-1), 2.65 (1H, *dd*, $J_{6\beta,5\alpha} = 10.7$ Hz, $J_{6\beta,6\alpha} = 18.5$ Hz, H-6), 2.68 (1H, *dd*, $J_{6\alpha,5\alpha} = 7.3$ Hz, $J_{6\alpha,6\beta} = 18.5$ Hz, H-6), 2.93 (1H, *sept*, $J = 6.8$ Hz, H-15), 3.74 (1H, *d*, $J = 11.2$ Hz, H-18), 3.83 (1H, *d*, $J = 11.2$ Hz, H-18), 7.31 (1H, *d*, $J = 8.2$ Hz, H-11), 7.42 (1H, *dd*, $J = 2.1$, 8.2 Hz, H-12), 7.88 (1H, *d*, $J = 2.1$ Hz, H-14). ^{13}C NMR spectral data (125 MHz, $CDCl_3$): see Table 1. Positive HR-ESIMS *m/z*: 539.4446, $[M+H]^+$ (calculated for $C_{36}H_{59}O_3$: 539.4459).

3.6. 3β-Hydroxy-abieta-8,11,13-trien-7-one (**3**)

Colorless oil, IR ν_{max} (KBr) cm^{-1}: 3439 (O–H), 2928 and 2867 (C–H alkanes), 1677 (C=O ketone), 1607 (C=C aromatic), 1459, 1384, 1266, 1234. ^{1}H NMR spectral data (500 MHz, $CDCl_3$): δ 0.97 (3H, *s*, H-19), 1.05 (3H, *s*, H-18), 1.24 (3H, *s*, H-20), 1.24 (3H, *d*, $J = 6.8$ Hz, H-16), 1.25 (3H, *d*, $J = 6.8$ Hz, H-17), 1.70–1.75 (1H, *m*, H-1), 1.84–1.90 (3H, *m*, H-2 and H-5), 2.38 (1H, *dt*, $J = 2.1$, 7.8 Hz, H-1), 2.70 (1H, *dd*, $J_{6\alpha,5\alpha} = 5.5$ Hz, $J_{6\alpha,6\beta} = 18.1$ Hz, H-6), 2.74 (1H, *dd*, $J_{6\beta,5\alpha} = 12.2$ Hz, $J_{6\beta,6\alpha} = 18.1$ Hz, H-6), 2.93 (1H, *sept*, $J = 6.9$ Hz, H-15), 3.35 (1H, *dd*, $J_{3\alpha,2\alpha} = 4.5$ Hz, $J_{3\alpha,2\beta} = 11.4$ Hz, H-3), 7.27 (1H, *d*, $J = 8.4$ Hz, H-11), 7.40 (1H, *dd*, $J = 2.1$, 8.4 Hz, H-12), 7.87 (1H, *d*, $J = 2.1$ Hz, H-14). ^{13}C NMR spectral data (125 MHz, $CDCl_3$): see Table 1. EIMS (probe) 70 eV, *m/z* (rel. int.): 300 $[M]^+$ (18), 285 $[M-Me]^+$ (30), 267 $[M-Me-H_2O]^+$ (87), 225 $[M-Me-H_2O-(CH_3)_2C]^+$ (25), 199 $[M-Me-C_5H_9OH]^+$ (100), 185 (23), 157 (21), 129 (32), 69 (81). Positive mode HR-EIMS *m/z*: 300.2094, $[M]^+$ (calculated for $C_{20}H_{28}O_2$: 300.2089).

3.7. Sandaracopimara-8(14),15-dien-18-yl formate (**4**)

Colorless oil, IR ν_{max} (KBr) cm^{-1}: 2926 and 2868 (C–H alkanes), 1728 (C=O ester), 1634 (C=C alkene), 1462, 1384, 1167 (C–O ester). ^{1}H NMR spectral data (300 MHz, $CDCl_3$): δ 0.84 (3H, *s*, H-20), 0.89 (3H, *s*, H-19), 1.04 (3H, *s*, H-17), 0.97–1.05 (1H, *m*, H-1), 1.29–1.32 (1H, *m*, H-5), 1.33–1.48 (6H, *m*, H-3, H-6 and H-12), 1.48–1.54 (2H, *m*, H-2), 1.54–1.59 (2H, *m*, H-11), 1.70–1.75 (2H, *m*, H-1), 1.74–1.78 (1H, *m*, H-9), 2.02–2.09 (1H, *m*, H-7), 2.21–2.27 (1H, *m*, H-7), 3.75 (1H, *d*, $J = 10.9$ Hz, H-18), 3.99 (1H, *d*, $J = 10.9$ Hz, H-18), 4.88 (1H, *dd*, $J = 1.5$, 10.6 Hz, H-16), 4.91 (1H, *dd*, $J = 1.5$, 17.4 Hz, H-16), 5.22 (1H, *brs*, H-14), 5.77 (1H, *dd*, $J = 10.6$, 17.4 Hz, H-15), 8.11 (1H, *s*, H-1′). ^{13}C NMR spectral data (75 MHz, $CDCl_3$): see Table 1. EIMS (probe) 70 eV, *m/z* (rel. int.): 316 $[M]^+$ (13), 301 $[M-Me]^+$ (33), 288 $[M-28]^+$ (6), 257 $[M-Me-44]^+$ (47), 255 $[M-Me-HCOOH]^+$ (17), 241 $[M-Me-60]^+$ (6), 135 (100), 121 (34), 107 (46), 93 (38). Positive HR-EIMS *m/z*: 316.2399, $[M]^+$ (calculated for $C_{21}H_{32}O_2$: 316.2402).

3.8. Sandaracopimara-8(14),15-dien-18-yl hexadecanoate (**5**)

Colorless oil, IR ν_{max} (KBr) cm^{-1}: 2925 and 2853 (C–H alkanes), 1737 (C=O ester), 1465, 1379, 1169 (C–O ester). ^{1}H NMR spectral data (300 MHz, $CDCl_3$): δ 0.84 (3H, *s*, H-20), 0.87 (3H, *s*, H-19), 0.80 (3H, *t*, $J = 6.7$ Hz, H-16′), 1.04 (3H, *s*, H-17), 1.00–1.06 (1H, *m*, H-1), 1.25 (26H, *m*, H-4′ to H-15′), 1.28–1.32 (1H, *m*, H-5), 1.31–1.40 (5H, *m*, H-2, H-3, H-6 and H-12), 1.43–1.54 (3H, *m*, H-2, H-6 and H-12), 1.54–1.65 (4H, *m*, H-11 and H-3′), 1.71–1.76 (2H, *m*, H-1 and H-9), 2.00–2.04 (1H, *m*, H-7), 2.24 (1H, *brd*, $J = 12.6$ Hz, H-7), 2.32 (3H, *t*, $J = 7.4$ Hz, H-2′), 3.62 (1H, *d*, $J = 10.9$ Hz, H-18), 3.88 (1H, *d*, $J = 10.9$ Hz, H-18), 4.88 (1H, *dd*, $J = 1.5$, 10.5 Hz, H-16), 4.91 (1H, *dd*, $J = 1.5$, 17.4 Hz, H-16), 5.22 (1H, *brs*, H-14), 5.77 (1H, *dd*, $J = 10.5$, 17.4 Hz, H-15). ^{13}C NMR spectral data (75 MHz, $CDCl_3$): see Table 1. Positive HR-ESIMS *m/z*: 527.4846 $[M+H]^+$ (calculated for $C_{36}H_{63}O_2$: 527.4823).

3.9. 18-Hydroxy-sandaracopimara-8(14),15-dien-7-one (**6**)

Colorless oil, IR ν_{max} (KBr) cm^{-1}: 3449 (O–H), 2928 and 2867 (C–H alkanes), 1676 and 1607 (α,β-unsaturated carbonyl system), 1459, 1384, 1266, 1045 (C–O alcohol). ^{1}H NMR spectral data (300 MHz, $CDCl_3$): δ 0.84 (3H, *s*, H-19), 0.87 (3H, *s*, H-20), 1.10 (3H, *s*, H-17), 1.05–1.15 (1H, *m*, H-1), 1.34–1.39 (2H, *m*, H-3), 1.39–1.66 (4H, *m*, H-2, H-11 and H-12), 1.64–1.67 (1H, *m*, H-12), 1.70–1.77 (1H, *m*, H-11), 1.77–1.81 (1H, *m*, H-1), 1.86 (1H, *dd*, $J_{5\alpha,6\alpha} = 5.2$ Hz, $J_{5\alpha,6\beta} = 13.6$ Hz, H-5), 2.10 (1H, *m*, H-9), 2.21 (1H, *dd*, $J_{6\beta,5\alpha} = 13.6$ Hz, $J_{6\beta,6\alpha} = 18.6$ Hz, H-6), 2.49 (1H, *dd*, $J_{6\alpha,5\alpha} = 5.2$ Hz, $J_{6\alpha,6\beta} = 18.6$ Hz, H-6), 3.09 (1H, *d*, $J = 10.9$ Hz, H-18), 3.37 (1H, *d*, $J = 10.9$ Hz, H-18), 4.98 (1H, *dd*, $J = 0.9$, 10.6 Hz, H-16), 5.01 (1H, *dd*,

$J = 0.9$, 17.5 Hz, H-16), 5.81 (1H, *dd*, $J = 10.6$, 17.5 Hz, H-15), 6.72 (1H, *brs*, H-14). ^{13}C NMR spectral data (75 MHz, $CDCl_3$): see Table 1. EIMS (probe) 70 eV, *m/z* (rel. int.): 302 $[M]^+$ (24), 287 $[M-Me]^+$ (13), 271 $[M-CH_2OH]^+$ (13), 269 $[M-Me-H_2O]^+$ (22), 133 $[M-169]^+$ (100), 121 (95), 105 (90), 93 (48). Positive HR-EIMS *m/z*: 302.2245, $[M]^+$ (calculated for $C_{20}H_{30}O_2$: 302.2246).

3.10. Abieta-8,11,13-trien-18-yl formate (7)

Yellowish oil, IR ν_{max} (KBr) cm^{-1}: 2932 and 2854 (C–H alkanes), 1738 (>C=O ester), 1606 (C=C aromatic), 1461, 1375, 1245, 1163 (C–O ester). ^{1}H NMR spectral data (300 MHz, $CDCl_3$): δ0.96 (3H, *s*, H-19), 1.22 (3H, *s*, H-20), 1.22 (6H, *d*, $J = 7.0$ Hz, H-16 and H-17), 1.39–1.42 (1H, *m*, H-1), 1.43–1.47 (2H, *m*, H-3), 1.63–1.79 (5H, *m*, H-2, H-6 and H-5), 2.29 (1H, *dt*, $J = 3.2$, 15.6 Hz, H-1), 2.82 (1H, *sept*, $J = 6.9$ Hz, H-15), 2.85–2.94 (2H, *m*, H-7) 3.79 (1H, *d*, $J = 11.1$ Hz, H-18), 4.10 (1H, *d*, $J = 11.1$ Hz, H-18), 6.90 (1H, *brs*, H-14), 7.00 (1H, *dd*, $J = 2.0$, 8.2 Hz, H-12), 7.18 (1H, *d*, $J = 8.2$ Hz, H-11), 8.06 (1H, *s*, H-1′). ^{13}C NMR spectral data (75 MHz, $CDCl_3$): 17.4 (C-19), 18.5 (C-2), 18.9 (C-6), 24.0 (C-16, C-17), 25.2 (C-20), 30.0 (C-7), 33.4 (C-15), 35.4 (C-3), 36.7 (C-4), 37.3 (C-10), 38.2 (C-1), 43.9 (C-5), 71.8 (C-18), 123.9 (C-12), 124.2 (C-11), 126.8 (C-14), 134.6 (C-8), 145.7 (C-13), 147.0 (C-9), 161.2 (C-1′). EIMS (probe) 70 eV, *m/z* (rel. int.): 314 $[M]^+$ (3), 299 $[M-Me]^+$ (9), 253 $[M-Me-HCOOH]^+$ (100), 211 (12), 185 (3), 141 (5). Positive HR-EIMS *m/z*: 314.2245, $[M]^+$ (calculated for $C_{21}H_{30}O_2$: 314.2246).

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References

Aboul-Ela, M.A., 2006. New abietane diterpene quinone fatty acid esters from *Salvia lanigera*. Alex. J. Pharm. Sci. 20, 32–34.

Adams, R.P., 1999. Systematics of multi-seeded eastern hemisphere *Juniperus* based on leaf essential oils and RAPD DNA fingerprinting. Biochem. Sys. Ecol. 27, 709–725.

Ara, I., Siddiqui, B.S., Faizi, S., Siddiqui, S., 1988. Tricyclic diterpenoids from the stem bark of *Azadirachta indica*. J. Nat. Prod. 51, 1054–1061.

Audier, H.E., Bory, S., Fétizon, M., Anh, N.-T., 1966. Spectres de mass des terpènes. III – influence dês liaisons éthyléniques sur la fragmentation des diterpénes. Bull. Soc. Chim. Fr., 4002–4010.

Bajda, M., Boryczka, S., Wietrzyk, J., Malawska, B., 2007. Investigation of lipophilicity of anticancer-active thioquinoline derivatives. Biomed. Chromatogr. 21, 123–131.

Barrero, A.F., del Moral, J.F.Q., Herrador, M.M., Akssira, M., Bennamara, A., Akkad, S., Aitigri, M., 2004. Oxygenated diterpenes and other constituents from Moroccan *Juniperus phoenicea* and *Juniperus thurifera* var. *Africana*. Phytochemistry 65, 2507–2515.

Burnell, R.H., Côté, C., Théberge, N., 1993. Approaches to the synthesis of aromatic diterpenes oxygenated in the A ring. Synthesis of margocin. J. Nat. Prod. 56, 1459–1467.

Cambie, R.C., Burfitt, I.C., Goodwin, T.E., Wenkert, E., 1975. Carbon-13 nuclear magnetic resonance spectroscopy of naturally occurring substances. XXXVII. Structure of hallol. J. Org. Chem. 40, 3789–3791.

Cerqueira, F., Cordeiro-da-Silva, A., Gaspar-Marques, C., Simões, F., Pinto, M.M.M., Nascimento, M.S.J., 2004. Effect of abietane diterpenes from *Plectranthus grandidentatus* on T- and B-lymphocyte proliferation. Bioorg. Med. Chem. 12, 217–223.

Chamy, M.C., Piovano, M., Gambaro, V., Garbarino, J.A., Nicoletti, M., 1987. Dehydroabietane diterpenoids from *Calceolaria ascendis*. Phytochemistry 26, 1763–1765.

Chang, L.C., Song, L.L., Park, E.J., Luyengi, L., Lee, K.J., Farnsworth, N.R., Pezzuto, J.M., Kinghorn, A.D., 2000. Bioactive constituents of *Thuja occidentalis*. J. Nat. Prod. 63, 1235–1238.

Da Silva, J.A., Pedro, L.G., Santos, P.A.G., Figueiredo, A.C., Barroso, J.G., Tenreiro, R.P., Ribeiro, C.A., Deans, S.G., Looman, A., Scheffer, J.J.C., 2000. Essential oils from seven populations of *Juniperus brevifolia* (Seub.) Antoine, an endemic species of the Azores. Flavour Fragr. J. 15, 31–39.

Enzell, C.R., Ryhage, R., 1967. Mass spectrometric studies of diterpenes. 5 Aromatic diterpenes. Arkiv Kemi 27, 213–229.

Fang, J.-M., Sou, Y.-C., Chiu, Y.-H., Cheng, Y.-S., 1993. Diterpenes from the bark of *Juniperus chinensis*. Phytochemistry 34, 1581–1584.

Fang, J.-M., Chen, Y.-C., Wang, B.-W., Cheng, Y.-S., 1996. Terpenes from heartwood of *Juniperus chinensis*. Phytochemistry 41, 1361–1365.

Fraga, B.M., Hernández, M.G., Artega, J.M., Suárez, S., 2003. The microbiological transformation of the diterpenes dehydroabietanol and teideadiol by *Mucor plumbeus*. Phytochemistry 63, 663–668.

Inoue, M., Hasegawa, S., Hirose, Y., 1985. Terpenoids from the seed of *Platyclaudus orientalis*. Phytochemistry 24, 1602–1604.

Kenmoku, H., Tanaka, M., Ogiyama, K., Kato, N., Sassa, T., 2004. Identification of (+)-phyllocladene, (−)-sandaracopimaradiene, and (+)-kaurene as new fungal metabolites from fusicoccin-producing *Phomopsis amygdali* F6. Biosci. Biotech. Biochem. 68, 1574–1577.

Mahmood, U., Kaul, V.K., Acharya, R., Jirovetz, L., 2003. *p*-Coumaric acid from *Tanacetum longifolium*. Phytochemistry 64, 851–853.

Maliepaard, M., De Mol, N.J., Janssen, L.H.M., Van der Neut, W., Verboom, W., Reinhoudt, D.N., 1992. Role of lipophilicity in the in vitro antitumor activity of a series of new mitosene compounds. Anticancer Drug Des. 7, 415–425.

Marques, C., Pedro, M., Simões, M., Nascimento, M.S.J., Pinto, M., Rodríguez, B., 2002. Effect of abietane diterpenes from *Plectranthus grandidentatus* on the growth of human cancer cell lines. Planta Med. 68, 839–840.

Miyazawa, M., Nakamura, Y., Ishikawa, Y., 2000. Insecticidal sesquiterpene from *Alpinia oxyphylla* against *Drosophila melanogaster*. J. Agric. Food Chem. 48, 3639–3641.

Muhammad, I., Mossa, J.S., Al-Yahya, M.A., Ramadan, A.F., El-Feraly, F.S., 1995. Futher antibacterial diterpenes from the bark and leaves of *Juniperus procera* Hochst. ex Endl. Phytother. Res. 9, 584–588.

Muhammad, I., Mossa, J.S., El-Feraly, F.S., 1996. Additional antibacterial diterpenes from the bark of *Juniperus procera*. Phytother. Res. 10, 604–607.

Otto, A., Wilde, V., 2001. Sesqui-, di-, and triterpenoids as chemosystematic markers in extand conifers – a review. Bot. Rev. 67, 141–238.

Richomme, P., Godet, M.-C., Foussard, F., Toupet, L., Sévenet, T., Bruneton, J., 1991. A novel leishmanicidal labdane from *Polyalthia macropoda*. Planta Med. 57, 552–554.

Sakar, M.K., San Feliciano, A., 1994. Diterpenoids of *Juniperus foetidissima* unripe berries. Fitoterapia 65, 304–306.

San Feliciano, A., del Corral, J.M.M., Gordaliza, M., Salinero, M.A., 1992. Diterpenoides neutros y compuestos aromaticos de las hojas de *Juniperus phoenicea* subsp. *turbinate*. An. Quim. 88, 512–516.

Schäfer, H., 2002. Flora of the Azores: A Field Guide. Margraf Verlag, Weikersheim.

Schneider, I., Gibbons, S., Bucar, F., 2004. Inhibitory activity of *Juniperus communis* on 12(*S*)-HETE production in human platelets. Planta Med. 70, 471–474.

Seca, A.M.L., Silva, A.M.S., Silvestre, A.J.D., Cavaleiro, J.A.S., Domingues, F.M.J., Neto, C.P., 2000. Chemical composition of the light petroleum extract of *Hibiscus cannabinus* bark and core. Phytochem. Anal. 11, 345–350.

Seca, A.M.L., Silva, A.M.S., 2006. The chemical composition of *Juniperus* genus (1970–2004). In: Govil, J.N., Singh, V.K. (Eds.), In: Recent Progress in Medicinal Plants, vol. 16. Studium Press LLC, Houston, pp. 401–522.

Seca, A.M.L., Silva, A.M.S., in press. The chemical composition of hexane extract from bark of *Juniperus brevifolia*. Nat. Prod. Res.

Sepúlveda, B., Astudillo, L., Rodríguez, J.A., Yáñez, T., Theoduloz, C., Schmeda-Hirschmann, G., 2005. Gastroprotective and cytotoxic effect of dehydroabietic acid derivatives. Pharmacol. Res. 52, 429–437.

Tanaka, R., Ohtsu, H., Matsunaga, S., 1997. Abietane diterpene acids and other constituents from the leaves of *Larix kaempferi*. Phytochemistry 46, 1051–1057.

Teixeira, A.P., Batista, O., Simões, F., Nascimento, J., Duarte, A., de la Torre, M.C., Rodríguez, B., 1997. Abietane diterpenoids from *Plectranthus grandidentatus*. Phytochemistry 44, 325–327.

Ulubelen, A., Topcu, G., 1992. Abietane diterpenoids from *Salvia pomifera*. Phytochemistry 31, 3949–3951.

Wang, S.-Y., Wu, J.-H., Shyur, L.-F., Kuo, Y.-H., Chang, S.-T., 2002. Antioxidant activity of abietane-type diterpenes from heartwood of *Taiwania cryptomerioides* Hayata. Holzforschung 56, 487–492.

Wenkert, E., Buckwalter, B.L., 1972. Cabon-13 nuclear magnetic resonance spectroscopy of naturally occurring substances. X. Pimaradienes. J. Am. Chem. Soc. 94, 4367–4369.

Yang, S.-J., Fang, J.-M., Cheng, Y.-S., 1998. Diterpenes from *Taxus mairei*. Phytochemistry 49, 2037–2043.