cin fraction (160 mg) was dissolved in DMSO (20 ml), and 200 × 100 μl portions were chromatographed repeatedly employing ODS column with the condition described below to afford a concentrated fraction of compound **2** Compound **2** was purified by rechromatography under same condition (1 9 mg)

LC conditions UV detector (wavelength 254 or 365 nm) Column·ODS (5 μm), 30 cm × 1 cm (YMC, Japan) or 40–45° with 0 01 M aq KH_2PO_4–MeCN–MeOH (3 1 1) or eluent The sample size was 100 μl (0 8 mg)/cycle

18,19-*Dehydrocamptothecin* (**2**) Pale yellow solid, MS (EI) *m/z* (rel int). 346 (51, M^+), 301 (100), 273 (57), 259 (11), 245 (14), 219 (18), 205 (11), HRMS *m/z* 346 0989 (for $C_{20}H_{14}N_2O_4$ = 346 0952) ^{1}H NMR ($CDCl_3$) δ 5 20 (*d*,1H,*J* = 16 1Hz, H-17), 5 33 (*brs*, 2H. H-5), 5 39 (*d*, 1H, *J* = 10 3 Hz, H-18), 5 40 (*d*, 1H, *J* = 17 6 Hz, H-18), 5 78 (*d*, 1H, *J* = 16 1 Hz, H-17), 5 89 (*dd*, 1H, *J* = 10 3 Hz, H-19), 7 69 (*dt*, 1H, *J* = 1 5 Hz and 8 1 Hz, H-10), 7 80 (*s*, 1H, H-14), 7 86 (*dt*, 1H, *J* = 1 5 Hz and 8 1 Hz, H-11), 7 96 (*d*, 1H, *J* = 8 1 Hz, H-9), 8 30(*d*, 1H, *J* = 8 1 Hz, H-12), 8 43 (*s*, 1H, H-7) CD $[\theta]_{241}$ +24400

Hydrogenation of compound **2** *to camptothecin* Compound **2** (1 85 mg) was dissolved in MeOH (50 ml) A part of the soln (10ml) was hydrogenated in presence of Pd-C as a catalyst under ambient H_2 with vigorous shaking to give camptothecin The reaction was completed within 20 min determined by HPLC CD spectrum of the filtrate of reaction solution was carried out directly

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COMPONENTS FROM *SANTOLINA ROSMARINIFOLIA*, SUBSPECIES *ROSMARINIFOLIA* AND *CANESCENS*

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Key Word Index—*Santolina rosmarinifolia*, subspp *rosmarinifolia* and *canescens*, Compositae, terpenoids, sesquiterpenes, eudesmanes, coumarins, acetylenes

Abstract—Apart from other already known components, two-eudesmane-type alcohols, two coumarins and a new spiroketalenolether-type acetylene were isolated from *S rosmarinifolia*, subspp. *rosmarinifolia* and *canescens* The structures assigned were based on their spectral properties. The relative stereochemistries of the new acetylene at C-11 and that of the known spiranic acetylenes were assigned by NMR-NOE experiments

In previous work we studied the essential oils of *S rosmarinifolia*, L , subsp. *rosmarinifolia* [1] and *S rosmarinifolia*, subsp *canescens* (Lag.) Nyman [unpublished results] In the present paper we report the results of the study on components of the neutral non-volatile fraction of both subspecies of *S rosmarinifolia*

From *S rosmarinifolia*, subsp *rosmarinifolia* we isolated 2-methyl-2,4-pentanediol (**1**), β-eudesmol (**2**), oplo-

panone (**3**), pygmol (**4**), (6*E*,3*R*,5*S*,9*S*)-9-acetoxy-5-hydroxynerolidiol (**5**), 1β,6α-dihydroxyeudesm-4(15)-ene (**6**) and capillin (**7**) the known 5(*E*)-and 5(*Z*)-isomers of polyacetylenes **8** and **9**, 6-methoxy-7,8-methylendioxycoumarin (**10**) and isoscopoletin (**11**).

Compound **1** [2] was isolated for the first time as a natural compound and coumarins are reported for the first time as components of *Santolina*; their physical and spectral data agree with those reported [3–6]. Compounds **2–5** and **7–9** were previously isolated from the essential oils of *S. rosmarinifolia* [1] or other species of *Santolina* [7–10].

From *S. rosmarinifolia*, subsp *canescens*, sitosterol (**12**), dammaradienol (**13**), dammaradienyl acetate (**14**), 6, spatulenol (**15**), caryophyll-3(12),7(13)-dien-6-ol (**16**), 1β-hydroxyeudesma-4(15),7-diene (**17**) and three isomeric spiroketalenolether type acetylenes **8**, **9** and **18**, were isolated

Compound **6** was identified as 1β,6α-dihydroxyeudesm-4(15)-ene and its IR and ^{1}H NMR spectra proved to be identical to those previously described by Bohlmann *et al* [11] for this compound. Its ^{13}C NMR spectral data are shown in Table 1.

Compound **17** was isolated by column chromatography as a monoacetate, **17a**, whose IR spectrum showed bands due to an acetoxyl group (1730, 1230 cm^{-1}), CH_2=C and CH=C unsaturations (3060, 1635, 880, 800 cm^{-1}) The ^{1}H NMR spectrum was consistent with the presence of four methyl groups, one of them on a quaternary carbon atom (0.73, 3H, *s*), two belonging to an isopropyl group (1.01, 6H, *d*, J=6.9 Hz) and one to a secondary acetoxyl group (2.04, 3H, *s*) whose geminal proton was shown at δ4.71 (1H, *dd*, J=11.8 and 4.5 Hz). The signals due to the CH_2=C group appeared at δ4.66, 4.85 and the multiplet assignable to the CH=C proton was shown at 5 27. Its ^{13}C NMR spectrum shows signals of four methyls (two of them identical), five methylenes (one of them olefinic), four methines (one olefinic and one oxygenated) and four quaternary carbons (two olefinic and one carbonyl)

These data agree with a bicyclic sesquiterpene with a cyclohexane moiety that supports an angular methyl, an olefinic methylene and an equatorial acetoxyl group, vicinal to a quaternary carbon atom, as can be deduced from their coupling constants. The trisubstituted double bond must be on the other ring together with the free rotating isopropyl group. All these groups might be assembled in an eudesmane skeleton with Δ^7-unsaturation according to the excision pattern of the olefinic proton [12, 13]

The β-disposition for acetoxyl group at C-1 was assigned by the ^{1}H NMR coupling constants of the signal due to the AcO-geminal proton All these data allowed us to assign the structure 1β-acetoxyeudesm-4(15),7-diene to **17a.**

In addition to the known acetylenes **8** and **9**, compound **18** was isolated The former compounds were previously isolated from other species of *Santolina* [11] The IR and ^{1}H NMR spectra of the three acetylenes were very similar; the IR spectrum showed the presence of double and triple carbon–carbon bonds, and one acetoxyl group; the ^{1}H NMR showed the presence of two methyl groups, one on an acetylenic carbon atom (δ1.98) and the other belonging to an acetoxyl group (2.04–2 14), together with five olefinic protons (see Table 2)

The most significant differences in the ^{1}H NMR spectra of this type of acetylenes are the signals due to the olefinic protons, which show the *E*- or *Z*-nature of the double bond at C-5 [14]. The ^{1}H and ^{13}C NMR (Table 3) assignments were confirmed by several 2D-experiments. Homonuclear H/H COSY experiments revealed the correlations between protons. The H–C connectivities and long range H/C correlations allowed us to assign the ^{13}C NMR signals, as shown in Table 3, for all the hydrogenated carbon atoms and for the spiranic one, respectively

Comparison of the ^{1}H NMR spectra of **8**, **9** and **18** (Table 2) suggested that in acetylenes **8** and **18**, the double bond was *E*, whereas in **9**, it was *Z*, hence the differences between **8** and **18** must be related with the stereochemis-

Table 1 ^{13}C NMR spectral data of compounds **6** and **17a** (50 3 MHz, $CDCl_3$, δppm)

C	**6**	**17a**
1	79 19	80 98
2	32 51	28.15
3	35.15	34.07
4	146 40	147 87
5	56 18	43 38
6	67 21	25 45
7	49 63	141,56
8	18 50	115 77
9	36 54	38 08
10	41 84	38 08
11	26 29	35 03
12	21 12	21 19
13	16 43	21 19
14	11 69	11 53
15	107 85	107 98
OAc		170 68
		21.69

Table 2 ^{1}H NMR spectral data for compounds **18**, **8** and **9** (200 MHz, $CDCl_3$ δppm)

H	**18**	**8**	**9**
1	1 98 *s*	1 98 *s*	1 98 *s*
6	5 10 *br s*	5 13 *br s*	4 78 *br s*
8	6 83 *d* (5 7)*	6 76 *d* (5 7)	6 36 *d* (5 7)
9	6.35 *dd* (5 7, 1 7)	6 34 *dd* (5.7, 1 8)	6 32 *d* (5 7)
11	5 65 *d* (2.5)	5 70 *dd* (2.0, 2 0)	5 65 *dd* (2 3, 1 6)
12	5 36 *dd* (2 8, 2 8)	5 20 *dd* (2 8, 2 8)	5 21 *dd* (2 9, 2 9)
13	6.68 *d* (2 8)	6 59 *dd* (3 1, 3 1)	6 61 *dd* (3 2, 3 2)
OAc	2.05 *s*	2 06 *s*	2 14 *s*

*Coupling constants in Hz

try at the chiral centre C-11 The ^{13}C NMR spectra of compounds **8** and **9** (Table 3) show identical shifts for C-9 to C-13, similar to those reported [15, 16] whereas compound **18** displays significant differences principally at C-11, thus confirming the aforementioned differences between **18**, **8** and **9** The NOE experiment (Fig 1) allowed us to establish the previously unknown relative *cis*-disposition of the acetoxy group at C-11 with respect to the tetrahydrofuranic oxygen atom in compounds **8** and **9** and the *trans*-disposition of the same groups in **18** According to these findings the relative stereochemistries of **8**, **9** and **18** are now definitively established

Table 3 ^{13}C NMR spectral data for compounds **18**, **8** and **9** (50 3 MHz, $CDCl_3$, δ ppm)

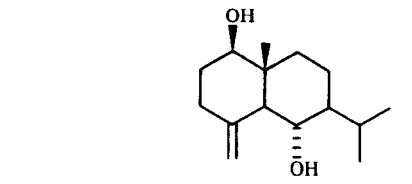
C	18	8	9
1	4 51	4 03	4 46
2	80 28	80 15	81 25
3	64 76	64 52	65 05
4	77 71	77 27	77 76
5	70 26	69 88	69 47
6	82 91	82 15	82 10
7	168 16	168 54	167 15
8	127 63	125 27	127 06
9	130 78	133 79	133 36
10	120 24	116 86	117 26
11	79 71	74 74	75 09
12	101 73	100 38	100 29
13	150.60	148.03	148 84
OAc	20.67	19.79	20 51
	169.40	169 66	170.86

EXPERIMENTAL

Plant material. Santolina rosmarinifolia, L subsp *rosmarinifolia*, was collected in June 1985 at Aldeaseca de la Armuña (Salamanca, Spain) and *Santolina rosmarinifolia*, subsp *canescens* was collected in July 1983 at Villahermosa (Cuenca, Spain) The voucher specimens are deposited in the herbarium of Biology Department, Faculty of Pharmacy

Extraction and isolation Air-dried and finely ground *S rosmarinifolia* subsp *rosmarinifolia* (5 4 kg) was extracted with MeOH. The extract was dewaxed by cooling, yielding 33 g (6 5%) of insoluble fraction H_2O was added to the solution (2 l) and the mixture extracted with hexane to yield 60 8 g (12 8%) of the hexane soluble part H_2O was added to the MeOH–H_2O-soluble fraction to give a H_2O–MeOH ratio of 2 1, and then the mixture was extracted with $CHCl_3$ to give 84 0 g of the $CHCl_3$-solubel part Finally, the $CHCl_3$ extract was extracted with Et_2O to yield 27 5 g of the Et_2O-soluble part The Et_2O fraction was chromatographed on silica gel and eluted with C_6H_6–Et_2O mixtures Six main fractions were collected and by repeated CC (silica gel, silica gel–$AgNO_3$) or crystallizations afforded 100 mg **7**, 1.07 g **8**, 450 mg **9**, 230 mg **10**, 326 mg **2**, 69 mg **1**, 480 mg **5**, 320 mg **6**, 185 mg **11**, 332 mg **3** and 1 16 g **4**

The hexane extract (150 g) of the air-dried and finely ground *S rosmarinifolia* subsp *canescens* (7.9 kg) was steam distilled to give 9 6 g (6 4%) of essential oil The non volatile fraction was defatted with MeOH to give 41 5 g of waxes, the remaining product in Et_2O soln was extracted with aq 4% NaOH to yield the neutral and acid fraction (32 12 and 10.63 g, respectively) The neutral fraction was separated by silica gel CC and eluted with hexane, C_6H_6, Et_2O and AcOEt Repeated CC (silica gel and silica gel–$AgNO_3$) or crystallizations of the fractions afforded 5 25 g **14**, 145 mg **18**, 148 mg **8**, 40 mg **9**, 1 4 g **15**, 430 mg **12**, 82 mg **13**, 50 mg **17**, 41 mg **16** and 30 mg **6**



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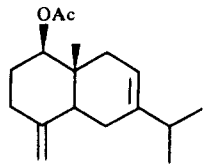
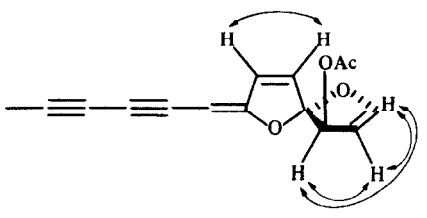
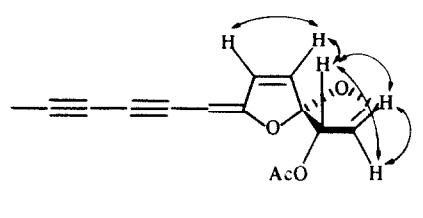


17a



8 *E* **9** *Z*



18 *E*

Fig 1 Structures of compounds **8**, **9** and **18**, with observed NOE's

Compound **1** Colourless oil IR ν_{max} cm^{-1} 3500, 1150, 1040; ^1H NMR ($CDCl_3$). δ 1 19 (3H, *d*, $J = 6.7$ Hz), 1 26 (3H, *s*), 1.32 (3H, *s*), 1 49 (1H, *dd*, $J_1 = 14.6$ $J_2 = 2.4$ Hz), 1.66 (1H, *dd*, $J_1 = 14.6$ $J_2 = 10.0$ Hz), 4.21 (1H, *ddc*, $J_1 = 12.3$ $J_2 = 10.0$ $J_3 = 6.2$ $J_4 = 2.4$ Hz)

Compound **17a** Colourless oil

λ	598	578	546	436	365
α	−2 7	−3.0	−3 3	−8.0	−18 0

($CHCl_3$, *c* 1 02%) IR ν_{max} cm^{-1}. 3060, 1730, 1635, 1230, 1020, 930, 880, 800; ^1H NMR ($CDCl_3$). δ 0 73 (3H, *s*), 1.01 (6H, *d* $J = 6.9$ Hz), 2.04 (3H, *s*), 4 71 (1H, *dd* $J_1 = 11.8$ $J_2 = 4.5$ Hz), 4.66 (1H, *d* $J = 1.4$ Hz), 4 85 (1H, *d* $J + 1.4$ Hz), 5 27 (1H, *m*) ^{13}C NMR (Table 1)

Compound **18** Semisolid product.

λ	589	578	546	436
α	277 0	290 7	337 2	639.8

($CHCl_3$; *c* 1 01%) IR ν_{max} cm^{-1}. 3100, 2220, 2120, 1750, 1650, 1630, 1580, 1230, 1030, 970, 915, 850, 800. ^1H NMR and ^{13}C NMR, see Tables 2 and 3

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KANSHONES D AND E, SESQUITERPENOIDS OF *NARDOSTACHYS CHINENSIS* ROOTS*

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Key Word Index—*Nardostachys chinensis*; Valerianaceae; sesquiterpenoid; kanshones D and E.

Abstract—Two new minor sesquiterpenoids, kanshones D and E, were isolated from *Nardostachys chinensis* along with isonardosinone, nardosinone diol, and nardofuran. The structures of kanshones D and E were elucidated by spectral means together with chemical transformation.

The crude drug, 'kanshoko' (Chinese spikenard), prepared from the rhizomes and roots of the valerianaceous plant *Nardostachys chinensis* Batalin (Valerianaceae), is used in the Oriental system of medicine for stomachic and sedative purposes, and is known to elaborate a number of sesquiterpenoids [1–11]. During the phytochemical investigation of this plant, we have so far isolated an iridoid, nardochin, and sesquiterpenoids, kanshones A, B and C, together with its known constituent nardosinone [12–14]. Further reinvestigation of the methylene chloride extract of this plant material has resulted in the isolation of two minor sesquiterpenoids kanshones D (**1**) and E (**2**) in addition to isonardosinone (**3**), nardosinone diol (**4**) and nardofuran (**5**), the latter two of which were previously reported as reaction products of nardosinone

*Part 63 in 'Sesquiterpenoids'. Also Part 130 in 'The Validity of the Oriental Medicines'.

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