

SESQUITERPENE LACTONES AND OTHER CONSTITUENTS OF *EUPATORIUM LANCIFOLIUM* AND *E. SEMISERRATUM**

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Key Word Index—*Eupatorium lancifolium*; *Eupatorium semiserratum*; *Eupatorium pilosum*; Eupatorieae; Compositae; heliangolides; germacradienolides; sesquiterpene lactones; flavones; obliquin hydrate; antileukaemic activity; cytotoxicity.

Abstract—The isolation of the cytotoxic and/or antileukaemic heliangolides eupacunolin, eupacunin, desacetyleupacunin, a new germacradienolide and a coumarin from *Eupatorium lancifolium* is reported. Structure elucidation of the previously known heliangolides was completed. *E. semiserratum* gave the antileukaemic germacradienolide desacetyleupaserrin and the flavones eupatorin, pectolinarigenin, hispidulin and salvigenin. Diploid *E. pilosum* was identical chemically with a previously studied polyploid.

INTRODUCTION

As part of a continuing study of *Eupatorium* species which elaborate sesquiterpene lactones with cytotoxic or antitumor activity [1†–8], we have investigated *Eupatorium lancifolium* (T. & G.) Small (*E. semiserratum* DC. var. *lancifolium* T. & G.), a diploid whose distribution is restricted to limited areas of southern Arkansas, northern Louisiana and eastern Texas [9–11]. Differentiation between *E. lancifolium* and *E. semiserratum* on the basis of morphology [9, 11] has caused some difficulties; we find that the two species can be distinguished fairly easily by the yellow secretion formed when the stems of *E. semiserratum* are broken. The chloroform extracts are now found to be chemically distinct as well, each giving rise to different sesquiterpene lactones or flavones.

RESULTS AND DISCUSSION

E. lancifolium furnished the cytotoxic lactone eupacunolin (**1a**), the antileukaemic lactone eupacunin (**1e**), desacetyleupacunin (**1f**), a new germacradienolide **2a** and a coumarin **3**. Compounds **1a** and **1e** were described earlier [12, 13] as constituents of *E. cuneifolium* Willd, whereas **1f**, available also by hydrolysis of **1a** [12, 13], has not been reported previously as a natural product. Acetylation of **1f** gave a diacetate identical with acetyleupacunin (**1g**).

The location of the primary hydroxyl group of eupacunolin was left uncertain in the earlier report

[12, 13]. We have now shown it to be at C-15 by comparing the ¹H NMR spectra of **1a** and its oxidation product **1d**, the H-5 signal of **1d** having experienced a significant paramagnetic shift due to the newly introduced conjugation (Table 1). As for its stereochemistry, the shift of the aldehydic proton demonstrated that the 4,5-double bond was *cis*; that the 9,10-double bond was *cis* also, as in the case of eupacunin, could be shown independently by an NOE experiment involving H-9 and H-14. Acetylation of **1a** gave the diacetyl derivative **1b**; partial hydrolysis of **1a** and **1c**, with H-3 moving to higher field and H-6 to lower due to the deshielding influence of the β -orientated 3-hydroxyl as in **1f**, confirmed the previously postulated allocation of the angeloyl ester group to C-8. The CD curves of **1a**, **1e** and **1f** were analogous to those of eurencurvin (**1h**) and its deoxy analog **1i**, thus confirming our suggestion [14] that the absolute configurations of eupacunin and its congeners are as shown. As the result of an arithmetical error, the magnitudes of $[\theta]$ for **1h** and **1i** as well as the compound numbered **4a** in ref. [14] were reported incorrectly [14]. The correct values are: for **1h**, $[\theta]_{266} - 160$, $[\theta]_{228} + 3500$ (last reading); for **1i**, $[\theta]_{260} - 600$, $[\theta]_{228} + 3700$ (last reading); for **4a** of ref. [14], $[\theta]_{260} + 760$, $[\theta]_{228} - 9100$ (last reading). We are indebted to Professor G. Snatzke, University of Bochum, for discovering the error.

The new substance **2a**, C₂₂H₂₈O₈, an α -methylene- α,β -unsaturated lactone (IR, ¹H NMR and ¹³C NMR spectra see Tables 1 and 2), had hydroxyl groups (broad IR band at 3460 cm⁻¹) and a *trans*-acetylsarracenoyl ester moiety (Tables 1 and 2). The number of C—O multiplets in the ¹³C NMR spectrum and the chemical shifts of H-2 and H-3 in the ¹H NMR spectrum were in agreement with the presence of hydroxyl groups on C-2 and C-3, the germacradienolide skeleton being deduced by the usual spin decoupling experiments which will not be detailed. H-8 at δ 5.84 was at lower field than H-6 at δ 5.15; hence the ester was attached to C-8. The small value for $J_{7,8}$ (< 1 Hz) indicated that H-8 was α .

The stereochemistry of the hydroxyl groups was deduced by comparing coupling constants with model

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†Compounds **1a**, **1e** and **1f** described in this report exhibited presumptive activity against P-388 lymphocytic leukemia in the mouse in tests carried out under auspices of the National Cancer Institute. Eucannabinolide [2] was a confirmed active.

Table 1. ^1H NMR spectra (270 MHz in CDCl_3 unless specified otherwise with TMS as internal standard)*

Assignment	1a†	1b†	1c	1d†	1e	1f	1g	2a
H-1	5.84 <i>d</i> (<i>br</i>) (11, 2)	6.56 <i>dd</i> (11, 2)	5.37 <i>d</i> (<i>br</i>) (10.5)	5.76 <i>m</i>	5.33 <i>m</i> ‡	5.41 <i>dd</i> (11, 1.5)	6.16 <i>m</i> ‡	5.10 <i>d</i> (<i>br</i>) (9.5)
H-2a	2.67 <i>ddd</i> (15, 11, 2.5)	2.34 <i>m</i>	2.09 <i>m</i>	2.50 <i>m</i>	2.20 <i>m</i>	2.18 <i>ddd</i> (15, 11, 2.5)	‡	4.45 <i>dd</i> (9.5, 8)
H-2b	2.46 <i>ddd</i> (15, 4.5, 2)	2.34 <i>m</i>		2.50 <i>m</i>		2.0‡	‡	
H-3	5.81 <i>dd</i> (4.5, 2.5)	5.74 <i>dd</i> (5, 2.5)	4.68 <i>m</i>	6.37 <i>t</i> (4)	5.33 <i>m</i> ‡	4.51 <i>m</i>	5.37 <i>dd</i> (5, 2.5)	4.12 <i>d</i> (8)
H-5	6.06 <i>d</i> (<i>br</i>) (11)	5.92 <i>d</i> (<i>br</i>) (11)	5.52 <i>d</i> (<i>br</i>) (10)	6.77 <i>d</i> (<i>br</i>) (11)	5.26 <i>d</i> (<i>br</i>) (10, 1)	5.31 <i>d</i> (<i>br</i>) (10.5)	5.31 <i>d</i> (<i>br</i>) (11)	5.04 <i>d</i> (<i>br</i>) (10)
H-6	6.44 <i>dd</i> (11, 8.5)	6.47 <i>dd</i> (11, 9)	6.49 <i>t</i> (10)	6.45 <i>m</i>	5.98 <i>dd</i> (10, 9)	6.51 <i>t</i> (10.5)	6.16 <i>m</i> ‡	5.13 <i>dd</i> (10, 9)
H-7	3.32 <i>m</i>	2.94 <i>m</i>	3.36 <i>m</i>	3.59 <i>m</i>	2.88 <i>m</i>	2.88 <i>m</i>	2.87 <i>m</i>	2.96 <i>m</i>
H-8	6.32 <i>m</i>	6.22 <i>dd</i> (6, 2.5)	5.89 <i>d</i> (<i>br</i>) (6)	6.41 <i>m</i>	5.89 <i>d</i> (<i>br</i>) (6)	5.89 <i>d</i> (<i>br</i>) (5.5)	5.86 <i>d</i> (<i>br</i>) (5.5)	5.86 <i>dd</i> (<i>br</i>) (5, 2, <1)
H-9a	5.50 <i>dd</i> (6, 1)	5.62 <i>dd</i> (6, 1.5)	5.34 <i>d</i> (<i>br</i>) (2)	5.48 <i>d</i> (<i>br</i>) (5.5)	5.41 <i>d</i> (<i>br</i>) (6)	5.37 <i>dd</i> (5.5, 1)	5.46 <i>dd</i> (5.5, 1)	2.87 <i>dd</i> (15, 5) 2.39 <i>dd</i> (15, 2)
H-13a	6.44 <i>d</i> (3.5)	6.47 (3)	6.30 <i>d</i> (3)	6.50 <i>d</i> (3)	6.33 <i>d</i> (3.5)	6.30 <i>d</i> (3.5)	6.32 <i>d</i> (3.5)	6.34 <i>d</i> (3.5)
H-13b	5.71 <i>d</i> (3)	5.71 <i>d</i> (3)	5.58 <i>d</i> (<i>br</i>)	5.82 (<i>br</i>)	5.58 <i>d</i> (3)	5.56 <i>d</i> (3)	5.53 <i>d</i> (3)	5.64 <i>d</i> (3)
H-14	1.92 (<i>br</i>)	1.85 (<i>br</i>)	1.68 (<i>br</i>)	1.73 (<i>br</i>)	1.73 (<i>br</i>)	1.77 (<i>br</i>)	1.78 (<i>br</i>)	1.60 (<i>br</i>)
H-15	4.48 (<i>br</i>)§	4.76§	3.98 (<i>br</i>)§	9.66	1.84 (<i>br</i>)¶	1.79 (<i>br</i>)¶	1.83 (<i>br</i>)¶	1.87 (<i>br</i>)¶
H-3'	5.94 <i>q</i> (7, 1.5)	6.13 <i>q</i> (<i>br</i>) (7)	6.18 <i>q</i> (<i>br</i>) (7)	5.98 <i>q</i> (<i>br</i>) (7)	6.23 <i>q</i> (<i>br</i>) (7)	6.18 <i>qq</i> (1, 1)	6.16 <i>m</i> ‡ (7)	6.56 <i>q</i> (<i>br</i>) (7)
H-4¶	1.96 <i>dd</i> (7, 1.5)	2.11 <i>d</i> (<i>br</i>) (7)	2.01 <i>d</i> (<i>br</i>) (7)	1.98 <i>dq</i> (7, 1.5)	2.03 <i>dq</i> (7, 1)	2.04 <i>dd</i> (7, 1)	2.01 <i>dq</i> (7, 1)	2.15 <i>d</i> (7)
H-5¶	2.01 (<i>br</i>)	2.07 (<i>br</i>)	1.94 (<i>br</i>)	2.03 (<i>br</i>)	1.96 (<i>br</i>)	1.99 (<i>br</i>)	1.91 (<i>br</i>)	4.69§
Ac¶	2.13	2.10, 2.09 1.92		2.14	2.11		2.17 1.91	2.01

*Unmarked signals are singlet. Figures in parentheses are coupling constants in hertz.

†Run in pyridine- d_5 .

‡Superimposed or obscured signals.

§Two proton intensity, centre of AB system.

||At δ 9.46 in CDCl_3 solution.

¶Three proton intensity.

compounds. Thus, the values of $J_{2,3}$ in 3β -hydroxy-8-acyloxy-*trans*-1(10),4,5-germacradiene-*trans*-6,12-olides are *ca* 5 and 9 Hz, whereas in the 3α -hydroxy analogs they are approximately 3 Hz each. As $J_{2,3}$ in **2** was 8 Hz the hydroxyl on C-3 was β . The magnitudes of $J_{1,2a}$ and $J_{1,2b}$ (9.5 Hz each) are consistent with α -orientation of the C-2 hydroxyl since in eupaserrin (**2b**), with 2-OH α , $J_{1,2}$ and $J_{2,3}$ are *ca* 9.6 and 9 Hz, respectively. The proposed stereochemistry at C-2 and C-3 is thus the same as that of eupafolitin from *E. perfoliatum* L. where the stereochemistry of the glycol moiety was deduced by application of the benzoate chirality rule and arguments based on the observed coupling constants [15]. The absolute stereochemistry is as shown in the formula because of the strongly negative n, n^* lactone Cotton effect.

A minor constituent of *E. lancifolium* was the coumarin **3** or less likely **4**, mp 168°, $[\alpha]_D - 213^\circ$. Racemic **3** and **4** have been synthesized [16] but the spectral differences between the two isomers are minimal. Optically active **3**, mp 168°, $[\alpha]_D + 177^\circ$, has been prepared by hydration of (+)-obloquin and by resolution of synthetic **3** [16]. The

coincidence of mp and the sign of rotation suggested that we were dealing with the enantiomer of (+)-obloquin hydrate whose absolute configuration was unknown. However, the relatively large difference in absolute value of $[\alpha]$ did not entirely exclude the possibility that the compound was **4**.

A previous study of the biologically active fractions from *E. semiserratum* by Kupchan and coworkers [17] afforded the antileukaemic lactones eupaserrin (**2b**) and desacetyleupaserrin (**2c**), as well as the cytotoxic flavone eupatorin (**5d**). Examination of the entire extract of a *E. semiserratum* collection from near Tallahassee has now yielded **2c** and relatively large quantities of eupatorin, pectolarigenin (**5a**), hispidulin (**5c**) and salvigenin (**5b**). The sizeable amount of flavonoid material undoubtedly accounts for the yellow secretion when the stems are broken and the deep yellow color of the extract; the difference in sesquiterpene lactone and flavone content of *E. lancifolium* and *E. semiserratum* appears to support their segregation into two distinct taxa.

E. pilosum Walt. [9, 10] is as of this date one of only two *Eupatorium* species *sensu stricto* [18] which do not

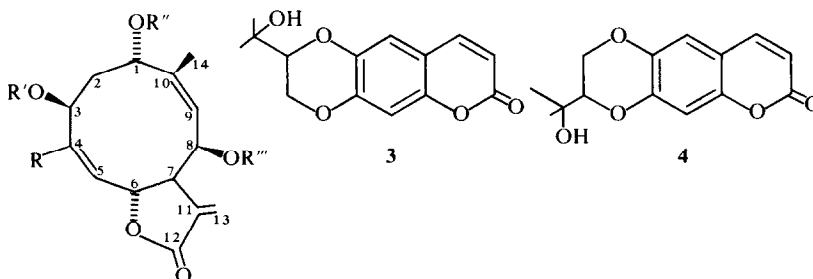
Carbon†	1a	1e	1f‡	2a
1	65.06 <i>d</i>	65.22 <i>d</i>	62.93 <i>d</i>	126.16 <i>d</i>
2	33.68 <i>t</i>	32.39 <i>t</i>	35.82 <i>t</i>	74.79 <i>d</i> §
3	70.78 <i>d</i>	73.23 <i>d</i>	71.41 <i>d</i>	83.61 <i>d</i>
4	142.31	139.18	141.64	144.23
5	126.63 <i>d</i>	126.11 <i>d</i>	125.08 <i>d</i>	132.19 <i>d</i>
6	74.34 <i>d</i>	74.34 <i>d</i>	74.24 <i>d</i>	75.03 <i>d</i> §
7	47.57 <i>d</i>	47.51 <i>d</i>	46.89 <i>d</i>	52.64 <i>d</i>
8	67.78 <i>d</i>	67.78 <i>d</i>	67.24 <i>d</i>	71.82 <i>d</i>
9	123.30 <i>d</i>	123.32 <i>d</i>	121.34 <i>d</i>	44.06 <i>t</i>
10	141.58	141.58	142.43	136.62
11	134.44	134.55	135.90	135.69
12	169.04§	168.93§	168.71	170.52§
13	122.28 <i>t</i>	121.88 <i>t</i>	120.00 <i>t</i>	121.57 <i>t</i>
14	17.38 <i>q</i>	17.46 <i>q</i>	17.63 <i>q</i>	13.49 <i>q</i>
15	65.06 <i>t</i>	23.35 <i>q</i>	23.00 <i>q</i>	15.94 <i>q</i>
1'	166.07	166.02	165.61	164.44
2'	126.63	126.43	127.31	127.06
3'	140.87	141.20	137.66 <i>d</i>	146.89 <i>d</i>
4'	20.38 <i>q</i>	20.40 <i>q</i>	20.06 <i>q</i>	19.92 <i>q</i>
5'	154.97 <i>q</i>	15.99 <i>q</i>	15.37 <i>q</i>	65.41 <i>t</i>
Ac	169.86§	169.23§		169.56§
	21.06 <i>q</i>	21.06 <i>q</i>		20.68 <i>q</i>

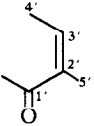
*Unmarked signals are singlets.

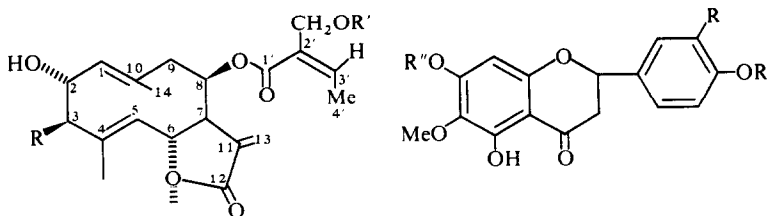
†Assignments tentative and not verified by SFRD.

‡In $\text{DMSO}-d_6$.

§Assignments interchangeable.



- 1a R = CH_2OH , R' = Ac, R'' = H, R''' =  (Ang)
 1b R = CH_2OAc , R', R'' = Ac, R''' = Ang
 1c R = CH_2OH , R', R'' = H, R''' = Ang
 1d R = CHO, R' = Ac, R'' = H, R''' = Ang
 1e R = Me, R' = Ac, R'' = H, R''' = Ang
 1f R = Me, R', R'' = H, R''' = Ang
 1g R = Me, R', R'' = Ac, R''' = Ang
 1h R = CH_2OH , R' = Ac, R'' = H, R''' = MeBu
 1i R = Me, R' = Ac, R'' = H, R''' = MeBu



- 2a R = OH, R' = Ac
 2b R = H, R' = Ac
 2c R, R' = H
 5a R, R'' = H, R' = Me
 5b R = H, R', R'' = Me
 5c R, R', R'' = H
 5d R = OH, R', R'' = Me

elaborate sesquiterpene lactones in detectable amounts; this distinguishes it chemically from *E. rotundifolium* (and hybrids of the latter) to which it has been assigned by some authors. In our earlier study of this taxon [19] we utilized a collection of polyploids (probably mainly tetraploid) from the Florida panhandle which gave slavigenin, cirsimaritin and two unusual δ -lactones of polyhydroxy-C₂₆ acids. Examination of the diploid variety, whose range is limited to St. Johns County, Florida, has now given essentially the same results.

EXPERIMENTAL

Extraction of *E. lancifolium*. Above-ground material of *E. lancifolium* (T. & G.) Small (1.1 kg), collected by Dr. R. K. Godfrey and Mr. D. Gage on 2 September, 1979, 10 miles west of Chidchester, Arkansas (RKG 77 203 on deposit in herbarium of Florida State University), was extracted with CHCl₃ and worked up in the usual manner [20]. The crude gum (100 g) was preadsorbed on 150 g of silicic acid (Mallinkrodt 100 mesh) and chromatographed over 1.2 kg of silicic acid packed in CHCl₃-toluene (1:1). Fractions were collected as follows: fractions 1–3 (CHCl₃-toluene, 1:1, 2l. each), 4–7 (CHCl₃, 2l. each), 8 (CHCl₃-MeOH, 49:1, 1.5l.), 9–14 (CHCl₃-MeOH, 49:1, 0.5l. each), 15–17 (CHCl₃-MeOH, 49:1, 2l. each), 18–25 (CHCl₃-MeOH, 19:1, 1.5l. each) and 26 (CHCl₃-MeOH, 9:1, 3l.).

Purification of fraction 11 by TLC (CHCl₃-MeOH, 9:1) gave two bands each of which was repurified twice by TLC in the same solvent system. This permitted separation of 36 mg of eupacuin (1e) and 20 mg of the coumarin 3. TLC of fraction 18 with EtOAc-hexane (1:1) and rechromatography of the first band using CHCl₃-MeOH (9:1) gave 80 mg of desacetylupacunolin (1f). TLC of fraction 21 (CHCl₃-MeOH, 9:1) gave 0.5 g of eupacunolin (1a), while repeated TLC of fraction 22 (CHCl₃-MeOH, 9:1) gave 100 mg 2a.

1e remained a brittle gum, $[\alpha]_D + 55.4^\circ$ (Me₂CO, *c* 0.0138), reported [12, 13] + 55°. Direct comparison with an authentic sample established the identity of 1e. CD curve (MeOH) $[\alpha]_{270} - 260$, $[\alpha]_{238} + 4080$, $[\alpha]_{223} 0$ (last reading). Detailed high resolution ¹H NMR and ¹³C NMR spectra which have not been reported previously are listed in Tables 1 and 2. [Calc. for C₂₂H₂₈O₇: MW, 404.1835. Found: MW (MS), 404.1835].

1f, remained a gum, $[\alpha]_D + 95.8^\circ$ (Me₂CO, *c* 0.0263), reported [12, 13] + 114; UV λ_{\max} 215 nm (ϵ 13 500); CD curve (MeOH): $[\theta]_{271} - 385$, $[\theta]_{237} + 4780$, $[\theta]_{226} + 1360$ (last reading); ¹H NMR and ¹³C NMR spectra listed in Tables 1 and 2. [Calc. for C₂₀H₂₆O₉: MW, 362.1728. Found: MW, 362.1696]. Other significant peaks in the high resolution MS were at *m/z* (composition, rel. int.): 345 (C₂₀H₂₅O₅, 16.6), 262 (C₁₅H₁₈O₄, 10), 244 (C₁₅H₁₆O₃, 2.1), 100 (C₅H₈O₂, 19.1), 96 (C₆H₈O, 18.9), 95 (C₆H₇O, 26.3) and 83 (C₅H₇O, 100). Acetylation of 20 mg of 1f (0.2 ml pyridine, 0.1 ml Ac₂O) followed by the usual work-up and TLC (CHCl₃-MeOH, 25:1) gave 12 mg of gummy 1b identical with material prepared by acetylation of 1e. [Calc. for C₂₄H₃₀O₈: MW, 446.1941. Found: MW (MS), 446.1933]. The ¹H NMR spectrum is listed in Table 1.

1a, although reported crystalline [12, 13], could not be induced to crystallize as is often the case in this series. $[\alpha]_D + 53.5^\circ$ (Me₂CO, *c* 0.056), reported [12, 13] $[\alpha]_D + 46^\circ$; CD curve: $[\theta]_{281} - 69$, $[\theta]_{238} + 4800$, $[\theta]_{222} 0$ (last reading); ¹H NMR and ¹³C NMR spectra listed in Tables 1 and 2. [Calc. for C₂₂H₂₈O₈: MW, 420.1784. Found: MW (MS), 420.1789]. Acetylation of 28 mg of 1a in the usual manner furnished gummy 1b whose ¹H NMR spectrum is listed in Table 1. [Calc. for C₂₆H₃₂O₁₀: MW, 504.1995. Found: MS (MS), 504.1995]. Hydrolysis of 40 mg

of 1a by stirring with 4 ml 1% KOH in H₂O at room temp. for 20 hr acidification with HOAc, extraction with CHCl₃ and TLC of the washed and dried organic layer (CHCl₃-MeOH, 22:3) gave 18 mg of non-crystalline 1c, slightly impure by NMR criteria. The ¹H NMR spectrum is listed in Table 1. A soln of 72 mg of 1a in 10 mg CHCl₃ was stirred with 680 mg active MnO₂ at room temp. for 5 days. The soln was filtered and concd *in vacuo*. TLC (CHCl₃-MeOH, 93:7) afforded 53 mg of 1d as a gum. IR ν_{\max} cm⁻¹: 3580, 1780, 1750, 1695 and 1685; MS *m/z*: 418 (M⁺), 400, 399, 388, 374, 358, 317, 275, 274, 257, 240, 230, 228, 212; ¹H NMR spectrum listed in Table 1.

2a could not be induced to crystallize. IR $\nu_{\max}^{\text{CHCl}_3}$ cm⁻¹: 3450, 1770, 1725 and 1250, $[\alpha]_D + 92.8^\circ$ (Me₂CO, *c* 0.0468); CD curve $[\theta]_{262} - 4530$, $[\theta]_{236} + 8240$ (last reading); ¹H NMR and ¹³C NMR spectra are listed in Tables 1 and 2. [Calc. for C₂₂H₂₈O₈: MW, 420.1784. Found: MW (MS), 420.1785]. Other significant peaks in the MS were at *m/z* (composition, rel. int.): 262 (C₁₅H₁₈O₃, 3.5), 245 (C₁₅H₁₇O₃, 4.6), 234 (C₁₄H₁₈O₃, 5.9), 218 (C₁₃H₁₄O₃, 9.8), 216 (C₁₄H₁₆O₂, 8.5), 161 (C₇H₁₃O₄, 11.0%) 141 (C₇H₉O₃, 70.1) and 81 (C₅H₅O, 100).

3 was crystallized from EtOAc-hexane, mp 168°. $[\alpha]_D - 213^\circ$ (CHCl₃, *c* 0.00235), IR $\nu_{\max}^{\text{CHCl}_3}$ cm⁻¹: 3450, 1730, 1630, 1570, 1300 and 1160; ¹H NMR (270 MHz, CDCl₃) δ : 6.27 (*J* = 9.5 Hz, H-3), 7.42 (*d*, *J* = 9.5 Hz, H-4), 7.00 (H-5), 6.87 (H-8), 4.5 (*dd*, *J* = 11.5, 2.5 Hz) and 4.11 (*dd*, *J* = 11.5, 9.5 Hz, methylene protons), 3.96 (*dd*, *J* = 9.5, 2.5 Hz, methinyl proton), 1.42 and 1.37 (Me \times 2); ¹³C NMR: δ : 161.28 (*s*, C-2), 114.39 (*d*, C-3), 114.10 (*d*, C-5), 143.03 (*d*, C-4), 140.85 (*s*, C-6), 146.84 (*s*, C-7), 104.81 (*s*, C-9), 112.84 (*s*, C-10), 65.38 (*t*, CH₂O), 78.71 (*d*, -CHO), 70.59 (*s*, C-O), 25.86 *q* and 25.42 *q* (Me); MS *m/z*: 262 (M⁺), 204, 175 and 59 (base peak).

Extraction of *E. semiserratum*. Above-ground material of *E. semiserratum* DC (3.6 kg), collected by Dr. R. K. Godfrey on 7 August 1968, along state route 71, 8 miles south of Blountstown, Calhoun Co., Florida, was extracted with CHCl₃ and worked up in the usual manner. The deeply yellow, crude gum (20 g) was adsorbed on 40 g of silicic acid and chromatographed over 300 g silicic acid in 250 ml fractions as follows: fractions 1–8 (CHCl₃-toluene, 1:1), 9–14 (CHCl₃), 15–20 (CHCl₃-MeOH, 99:1), 21–25 (CHCl₃-MeOH, 97:3), 26–30 (CHCl₃-MeOH, 19:1), 31–36 (CHCl₃-MeOH, 9:1).

Fractions 9–14, which showed one major spot, were combined and recrystallized from EtOAc-hexane to give 150 mg of 2c, mp 134–136°, identical with an authentic specimen. Fractions 15–20 were combined. After purification by prep. TLC the major constituent crystallized from C₆H₆-MeOH as yellow needles, mp 188–189°, and was identified as salvigenin (5b) by IR, NMR and comparison with an authentic sample. Fractions 21–25 were combined and purified by prep. TLC (CHCl₃-MeOH, 19:1). Recrystallization from CHCl₃-MeOH afforded yellow needles of pectolarigenin (5a), mp 216–218°, yield 200 mg, identified by IR, NMR and comparison with an authentic sample. Fractions 26–30 were mixtures of two flavonoids which were separated by prep. TLC. The upper band was crystallized from CHCl₃-MeOH, mp 195–197°, yield 100 mg, and was identified as eupatorin by IR, NMR and comparison with an authentic sample. The lower band, after crystallization from CHCl₃-MeOH, had mp 290–291°, yield 150 mg, and was identified as hispidulin by IR, NMR and direct comparison with an authentic sample.

Extraction of *E. pilosum*. Above-ground parts of diploid *E. pilosum* Walt (8.5 kg), collected by Dr. R. K. Godfrey and Mr. D. Gage on 12 July 1978 along route 16, 1 mile west of I-10, St. Johns County, Florida (RKG 76609 on deposit in herbarium of Florida State University), were extracted with CHCl₃ and worked up in the usual fashion. Chromatography as described earlier [19]

gave 0.35 g of salvigenin, 0.3 g of cirsimaritin and 0.2 g of the δ -lactone of Z-5,7,9,11,15-pentahydroxyhexacos-2-enoic acid.

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NOTE ADDED IN PROOF

A compound, mp 167°, $[\alpha]_D + 171^\circ$, presumed to be identical with obloquin hydrate, has recently been isolated from *Conocliniopsis prasiifolia* [Bohlmann, F., Zdero, C., King, R. M. and Robinson, H. (1980) *Phytochemistry* **19**, 1547] and *Acratopappus hagei* [Bohlmann, F., Zdero, C., Gupta, R. K., King, R. M. and Robinson, H. (1980) *Phytochemistry* **19**, 2695].