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GUAIANOLIDES FROM *SAUSSUREA LAPPA*

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Key Word Index—*Saussurea lappa*; Compositae; sesquiterpenoids; guaianolides; α-methylene-τ-lactone; biological activity; dehydrocostus lactone.

Abstract—Two new sesquiterpene lactones with an α,β-unsaturated aldehyde group have been isolated from the roots of *Saussurea lappa*. Their stereostructures have been established by the combined use of spectroscopic as well as chemical methods via their synthesis from dehydrocostus lactone.

INTRODUCTION

In the course of our investigations of higher plants for biological activity as plant growth regulators, the hexane extract from the powdered costus roots (*Saussurea lappa*) was found to cause adventitious root formation in the stem cuttings of *Vigna radiata*. Costus roots are a rich source of two crystalline, sesquiterpene lactones, dehydrocostus lactone (**1**) and costunolide (**2**). Although biological activity of the terpenoids from this species is mainly attributed to the presence of the α-methylene-τ-lactone moiety [1–3], more potent compounds have been isolated from the same source. One of them is saussureal (**3**) [4], which displayed high growth regulatory activity and has a modified eudesmane skeleton with ring A being five-membered. 13-Methoxy-dihydrodehydrocostus lactone (**4**) [5] and 4-β-methoxydehydrocostus lactone (**5**) [6] have also been isolated and have shown fairly good biological activity as plant growth regulators. Our continued bioassay directed fractionation of the hexane extract has resulted in the isolation of two new sesquiterpene lactones, neither of which was responsible for the activity. We wish to report here the isolation and chemical characterization of the two new sesquiterpene lactones (**6** and **7**).

RESULTS AND DISCUSSION

A hexane extract of powdered costus roots was cooled when a major portion of the oil solidified. The solid portion contained mainly costunolide and dehydrocostus lactone. From the mother liquor, extensive column chromatography monitored by IR led to the isolation of two aldehydic sesquiterpene lactones **6** and **7**. Compound **6**, mp 178° has the molecular formula $C_{15}H_{16}O_3$–244.2876 [high resolution mass spectrometry (HRMS)]. The IR spectrum suggested the presence of α-methylene-τ-lactone and α,β-unsaturated aldehyde moieties. The ^1H NMR spectrum exhibited signals at δ6.1 (1H, *d*, $J = 2.5$ Hz) and 5.3 (1H, *d*, $J = 2.5$ Hz) for the exomethylene group of τ-lactone, and δ3.6 (1H, *t*, $J = 8$ Hz) for a proton germinal to the oxygen of the lactone ring. The signals at δ5.6 (1H, *m*) and 1.95 (3H, *bs*) could be attributed to a vinyl hydrogen and methyl, respectively. The ^{13}C NMR spectrum exhibited signals assignable to two carbonyl groups (one that of a conjugated aldehyde and the other that of a conjugated lactone), one secondary carbon bearing oxygen, one methyl, three methylenes and two methines. Of the four sp^2 carbons other than the exomethylene double bond, three were quaternary and one was secondary.

These data pointed to the close structural relationship of this guaianolide with dehydrocostus lactone. Of the two possible structures (**6** and **8**), **8** was eliminated due to the multiplicity of the H-6 signal, which was a triplet rather than a doublet. Further proof in favour of structure **6** was achieved by its synthesis from isodehydrocostus lactone (**9**) [7]. The latter on catalytic oxidation with selenium dioxide in the presence of *tert*-butyl hydroperoxide in methylene chloride for 24 hr yielded a tertiary allylic alcohol (**10**), represented without stereochemistry, as the major compound. Further oxidation with pyridinium chlorochromate, which brings about a 1,3-transposition in the case of allylic alcohols followed by oxidation to an α,β-unsaturated carbonyl [8], yielded a compound of mp 178° whose IR and NMR spectra were identical to those of **6**.

The other compound (**7**) was isolated as a liquid of molecular formula $C_{15}H_{16}O_3$—244.3086 (HRMS). Its IR, ^1H NMR and ^{13}C NMR spectra were quite similar to those of compound **6**, indicating the presence of an α-methylene-τ-lactone (1755 and 890 cm^{-1}) and an α,β-unsaturated aldehyde (1700 and 2710 cm^{-1}), the major

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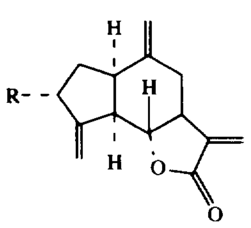

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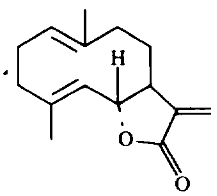

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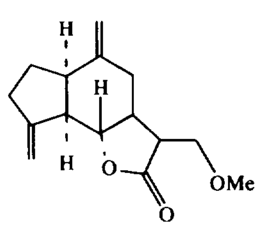

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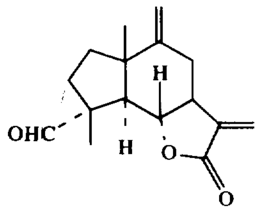

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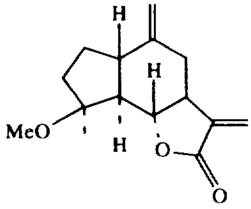

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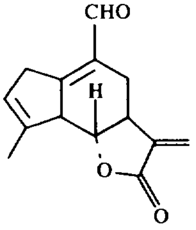

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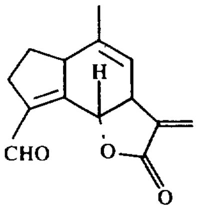

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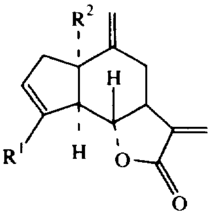
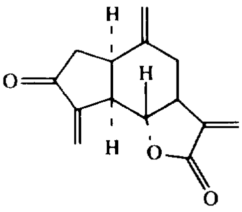


7: R_1 = CHO, R_2 = H
9: R_1 = Me, R_2 = H
10: R_1 = Me, R_2 = OH



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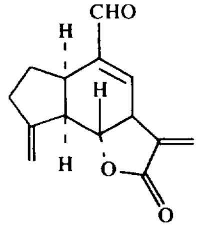

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differences being the presence of a second exomethylene group (δ5.0 and 5.4, both *br s*) rather than a vinyl methyl and a low field proton δ5.6 (1H, *bs*) attributable to the β-hydrogen of an α,β-unsaturated aldehyde. These spectral features were also supported by ^{1}H and ^{13}C NMR spectra (Tables 1 and 2), suggesting structure **7** for this compound. Structure **7** rather than the alternative structure **11** was established by oxidation of isozaluzanin-C (**12**) with pyridinium chlorochromate, which yielded a two-component mixture. Column chromatography of the mixture on silica gel gave the α,β unsaturated ketone (**13**) followed by a liquid, which had all spectra features identical with those of the natural product (**7**).

Table 1. ^{1}H NMR spectral data of compounds **6** and **7**

	6	**7**
C_3-H	5.6, 1H, *m*	6.5, 1H, *bs*
C_6-H	3.6, 1H, *t* (J = 8 Hz)	4.1, 1H, *t* (J = 8 Hz)
C_{13}-H	5.3 and 6.1, 1H each *d* (J = 2.5 Hz)	5.5 and 6.2, 1H, each, *bs*
C_{14}-H	9.0, 1H, *s*	5.0 and 5.4, 1H each, *bs*
C_{15}-H	1.95, 3H, *m*	9.8, 1H, *s*

EXPERIMENTAL

All compounds gave satisfactory HRMS, IR: Nujol, ^{1}H NMR and ^{13}C, TMS int. standard. CC sepns were performed on silica gel. Evapn of solvents from extracts and CC frs was carried out below 60°.

Isolation of guaianolides (**6** *and* **7**). The hexane extract (30 g) was kept overnight at 4° when a solid mixture (20 g) was obtained. It was subjected to CC on silica gel (1 kg), the column being eluted with solvents of increasing polarity. Elution with hexane gave a trace mixture (0.3 g) followed by a mixture of two compounds (1.3 g) eluted with hexane–Et_2O (20:1, 1 l). Further elution of the column with the same solvent gave a crude solid (2.5 g) identified as **2**, which was followed by a solid mixture (1.2 g) of **2** and **1** (TLC). This was followed by pure **1** (12.35 g) identified by spectral methods (IR and NMR spectra identical to those of an authentic sample). Elution of the column with hexane–Et_2O (10:1, 0.5 l)

Table 2. ^{13}C NMR spectral data of compounds **6** and **7**

C	**6**	**7**
1	134.4 (*s*)	25.3 (*d*)
2	52.3 (*t*)	22.7 (*t*)
3	135.4 (*d*)	134.3 (*d*)
4	168.5 (*s*)	144.0 (*s*)
5	53.3 (*d*)	52.3 (*d*)
6	79.4 (*d*)	78.8 (*d*)
7	56.1 (*d*)	54.4 (*d*)
8	22.5 (*t*)	20.9 (*t*)
9	40.8 (*t*)	38.6 (*t*)
10	148.4 (*s*)	122.5 (*s*)
11	131.8 (*s*)	130.4 (*s*)
12	169.3 (*s*)	168.8 (*s*)
13	120.6 (*t*)	120.5 (*t*)
14	198.7 (*d*)	124.2 (*t*)
15	20.6 (*q*)	200.1 (*d*)

The assignment of various carbon shifts was done by measuring the spectra under INEPT conditions.

gave a mixture of compounds (0.2 g). The Et_2O fr. (1.5 g) constituted a mixture of at least two major compounds. This fr. (1.5 g) was further chromatographed on silica gel/$AgNO_3$ (150 g). Elution of the column with hexane–Et_2O (9:1, 2 l) gave a pure solid compound, mp 178°, $C_{15}H_{16}O_3$, 244.2876 (HRMS). IR bands 3080, 2720, 1760, 1720, 1660 and 890 cm^{-1}. Further elution of the column with hexane–Et_2O (20:3, 1 l) yielded a mixture (0.1 g) followed by another pure liquid compound (**7**). $C_{15}H_{16}O_3$, 244.3086 (HRMS), IR bands at 3076, 2710, 1755, 1700, 1680 and 890 cm^{-1}.

Oxidation of compound **10** *with pyridinium chlorochromate.* Compound **10** was prepd by the known lit. method [8]. To a soln of alcohol **10** (0.5 g) in CH_2Cl_2 (15 ml) was added, with continuous stirring, a suspension of pyridinium chlorochromate (1.0 g) in CH_2Cl_2 (20 ml). The reaction mixture was worked up as usual after 2 hr, and evapn of the solvent gave a two-component mixture, which, on CC, yielded the starting material (0.1 g) and the compound, mp 178° 0.35 g, identical in all respects to the natural product.

Oxidation of isozaluzanin-C (**12**) *with pyridinium chlorochromate.* Compound **12** was prepd by the procedure already reported [7], 0.7 g of the product was oxidized as above and usual work up yielded a two-component mixture, CC sepn of which yielded a solid α,β unsaturated ketone (0.3 g) IR ν_{max} cm^{-1}: 3075, 1760, 1725, 1623 and 825, ^1H NMR: δ4.10 (*t*, 1 H, $J = 9$ Hz), 4.63 and 5.00 (*s*, 1H each), 5.13 and 6.40 (*d*, 1H each, $J = 3$ Hz) 5.90 and 6.25 (*d*, 1H, each $J = 2$ Hz). The other liquid compound (0.32 g) showed all the spectral features superimposable with those of the natural product (**7**).

REFERENCES

1. Paul, A. A., Bawdekar, A. S., Joshi, R. S., Kulkarni, G. H., Rao, A. S., Kelkar, G. R. and Bhattacharyya, S. C. (1960) *Perf. Ess. Oil Rec.* **51**, 115.
2. Kalsi, P. S., Chhabra, B. R., Chhabra, A. and Wadia, M. S. (1979) *Tetrahedron* **35**, 1993.
3. Kalsi, P. S., Kaur, P. and Chhabra, B. R. (1979) *Phytochemistry* **18**, 1877.
4. Talwar, K. K., Singh, I. P. and Kalsi, P. S. (1992) *Phytochemistry* **31**, 336.
5. Dhillon, R. S., Kalsi, P. S., Singh, W. P., Gautam, V. K. and Chhabra, B. R. (1987) *Phytochemistry* **26**, 1209.
6. Singh, I. P., Talwar, K. K., Arora, J. K., Chhabra, B. R. and Kalsi, P. S. (1992) *Phytochemistry* **31**, 2529.
7. Kalsi, P. S., Sharma, S. and Kaur, G. (1983) *Phytochemistry* **22**, 1993.
8. Sundararaman, P. and Herz, W. (1977) *J. Org. Chem.* **42**, 813.
9. Kalsi, P. S., Kaur, G., Sharma, S. and Talwar, K. K. (1984) *Phytochemistry* **23**, 2855.