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FLAVONES, SESQUITERPENE LACTONES AND GLYCOSIDES ISOLATED FROM *CENTAUREA ASPERA* VAR. *STENOPHYLLA*

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Key Word Index—*Centaurea aspera*; Compositae; stenophyllolide; melitensin; dehydromelitensin; flavones; glycosides; sesquiterpene lactones; methoxyluteoline; glucuronides.

Abstract—From the alcoholic extract of *Centaurea aspera* var. *stenophylla* benzoic acid, *p*-hydroxybenzoic acid, apigenin, 6-methoxyluteolin, 11,13-dehydromelitensin, melitensin, stenophyllolide, ethyl-7-*O*-apigenin-glucuronate and the glucosides of sitosterol and stigmasterol were isolated and characterized. Stenophyllolide was shown to be 9,15-dihydroxygermacra-1(10),4,11-trien-12,6-olide.

INTRODUCTION

From the flowers of *Centaurea aspera* var. *stenophylla* a new sesquiterpene lactone, named stenophyllolide, has been isolated by Viguera and his collaborators [1, 2]. We have examined an extract of the flowers of *C. aspera* var. *stenophylla* and now report the identification of the flavones, glycosides and sesquiterpene lactones isolated from this alcoholic extract. We have also elucidated the structure of stenophyllolide.

RESULTS AND DISCUSSION

The well known plant constituents isolated and identified were benzoic acid, *p*-hydroxybenzoic acid, apigenin and sitosterol and stigmasterol glucosides. In addition the following components, which have only been isolated a few times or not previously isolated from other plants, were characterized.

6-Methoxyluteolin or 6-methoxy-3′,4′,5,7-tetrahydroxyflavone

^{1}H NMR, IR, UV maxima and their shifts with the usual reagents require the compound described in the experimental to be either an 8-methoxy-3′,4′,5,7-tetrahydroxyflavone or a 6-methoxy-3′,4′,5,7-tetrahydroxyflavone. However, its mass spectrum supported the later structure, since its $[M]^+$ peak at m/z 316 was the base peak and $[M-18]^+$ at m/z 298, showed an abundance of 81.5%, while the former structure would give $[M-15]^+$ as the base peak, and $[M-18]^+$ should not show an abundance higher than 10% [3]. All the properties of the compound (mp, IR, UV, ^{1}H NMR) coincide with those of the eupafolin, isolated from *Eupatorium cuneifolium* by Kupchan *et al.* [4], with the exception of the mp of its tetraacetate (184–185°, our tetraacetate 195°). The same compound was isolated from *Digitalis schischkinii* by Imre *et al.* [5], and from *Nepeta hindostana* and given the name nepetin by Krishnaswamy *et al.* [6].

Melitensin and 11,13-dehydromelitensin

Both compounds have been isolated previously from natural sources, melitensin from *Centaurea melitensis* [7] and 11,13-dehydromelitensin from *Centaurea pullata* [8]. The mp, IR and ^{1}H NMR spectra of our melitensin coincided fully with those reported, and in addition were the same as a synthetic sample, obtained by us from artemisin [9]. 11,13-Dehydromelitensin was identified by its reduction by $NaBH_4$ to melitensin.

Ethyl-7-O-apigenin-glucuronate

This glucuronide has been isolated by Zapesochnaya and Banlkovskii [10] from *Achillea cartilaginae*; our experimental data are coincident with what they call apigenin 7-*O*-β-D-ethyl glucuronide.

Stenophyllolide

This is the main sesquiterpene lactone isolated from this plant (574 mg). Although neither our purified compound nor its diacetate melted when heated to over 200°, it did slowly decompose, and Viguera [1] reported the mp 186–188° for stenophyllolide. We believe that the isolated compound is stenophyllolide since it showed the same IR spectrum, molecular formula, about the same rotation and it was extracted as the main sesquiterpene lactone from the same plant collected in the same place as in the previous report [1].

The IR spectrum of stenophyllolide showed bands of a γ-lactone (1755 cm^{-1}), hydroxyl group (3290 cm^{-1}), and double bond (1655 and 965 cm^{-1}). The mass spectrum, together with elemental analysis data, suggested the molecular formula $C_{15}H_{20}O_4$. Of the four oxygens, two formed a lactone ring and the other two were hydroxyl groups, as was confirmed by its diacetate derivative, for which elemental analysis and the mass spectrum afforded the molecular formula $C_{19}H_{24}O_6$. The exocyclic methylene, conjugated with a carbonyl lactone was easily recognizable in the two one-proton-doublets at δ6.05 and 5.65 (J = 3, 2 Hz). These doublets were coupled with H-7

Table 1. ^{1}H NMR spectra of stenophyllolide and its diacetate at 200 MHz*

Protón	Stenophyllolide	Stenophyllolide diacetate
H-1	4.99 (1H, *br dd*) (J = 12, 5, 1.2 Hz)	5.32–5.16 (1H, *m* overlapped with H-9)
H-2	2.2–1.8 (2H, *m*)	2.22 (2H, *m*)
H-3	2.2–1.8 (2H, *m* overlapped)	2.58 (2H, *m*)
H-5	4.68 (1H, *d*, J = 10 Hz)	4.85 (1H, *d*, J = 8.8 Hz)
H-6	4.83 (1H, *dd*, J = 10; 9 Hz)	4 58 (1H, hidden *t*, J = 8 8 Hz) (overlapped H-15)
H-7	2.68 (1H, *m*)	2.73 (1H, *m*)
H-8	1 8–1 6 (2H, *m*)	1.94 (2H, *m*)
H-9	4.05 (1H, *m*)	5.32–5.16 (1H, *m* overlapped with H-1)
H-13$_a$	6.05 (1H, *d*, J = 3.2 Hz)	6.32 (1H, *d*, J = 3.2 Hz)
H-13$_b$	5.65 (1H, *d*, J = 3.2 Hz)	5 58 (1H, *d*, J = 3.2 Hz)
H-14	1.30 (3H, *d*, J = 1.2 Hz)	1.40 (3H, *d*, J = 1.2 Hz)
H-15$_a$	4.06 (1H, *dd*, J = 12; 5 Hz)	
H-15$_b$	3.80 (1H, *dd*, J = 12, 5 Hz)	4.58 (2H, *m* overlapped H-6)
AcO		2.10 and 2.05 (6H, 2*s*)
OH	4.06–3.8	

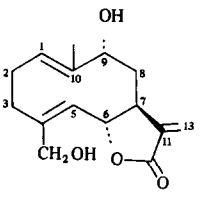
*Run in CD_3SOCD_3 for stenophyllolide and in $CDCl_3$ for its diacetate

(δ2.68), since when irradiated with the frequency 2.68, they collapsed into two singlets. The vinylic proton H-1 was observed in the broadened doublet of doublets (J = 12, 5 and 1, 2 Hz) at δ4.99; irradiation at the H-14 frequency (1.30) sharpened this doublet of doublets. The signal of H-14 appeared at 1.30 as a three-proton broad doublet which sharpened when irradiated at the H-1 frequency. The other vinylic proton at H-5 was seen as a doublet (J = 10 Hz) at 4.68. The signal of the lactonic proton at H-6 was a deformed triplet (*dd* under amplification) at 4.83, which when irradiated at H-7, collapsed to a doublet (J = 10 Hz). There are two hydroxyl groups, one on a primary carbon and the other on a secondary carbon; the signal for $\underline{CH_2}OH$ appeared as two doublet of doublets (eight peaks) and resulted from the coupling between themselves and the hydroxyl group; they were centred at 4.06 and 3.80 (J = 12 and 5 Hz), respectively. These signals are partially overlapped by a signal for the proton (H-9) geminal to a secondary hydroxyl group, which appeared as a hidden multiplet at 4.05 and should be located at C-9, because this signal was affected only by irradiation of the frequency of H-8 (1.80–1.60). If the hydroxyl group was located at C-8, the signal of its geminal proton should be affected by irradiation of H-7, and this was not the case. The signal of H-7 was a complex multiplet, centred at 2.68, and suffered alteration by irradiation of H-13, H-8 and H-6. The signals of H-2 and H-3 were a complex multiplet at 2.20–1.80 (4H) and they were not affected by irradiation of H-7. The signal of H-8 was a complex multiplet (1.80–1.60) which suffered alteration by irradiation of H-7 and H-9. Finally, the hydroxyl groups appeared as a broad signal at 4.74.

We now turn to the stereochemistry of this molecule. If it is assumed that H-7 is α as in other compounds whose absolute stereochemistry has been established by X-ray analysis, the large value of $J_{5,6}$ (10 Hz) and $J_{6,7}$ (9 Hz) requires the H-6 to be β and H-5 to be α. That the lactone ring is *trans*-fused is also supported by the magnitude of $J_{7,13a}$ and $J_{7,13b}$ (> 3 Hz) [11].

This structure and stereochemistry was confirmed by the ^{1}H NMR spectrum of stenophyllolide diacetate. The signal of H-9, geminal to the hydroxyl group, was moved about 1 ppm to lower field by acetylation and appeared now at δ5.32–5.16, overlapped with H-1; therefore this signal suffers alteration by irradiation at H-2, H-14 and H-8. In a similar way the signal of H-15, geminal to the other hydroxyl group moved to 4.58, overlapped with H-6, and therefore it suffered alteration when irradiated at H-5 and H-7. The magnitude of the coupling constant between H-13 and H-7 was the same in the acetate and the free alcohol.

Finally all these points of chemistry and stereochemistry were confirmed by the X-ray study of crystalline stenophyllolide (to be published elsewhere). In addition this X-ray study has shown that the hydroxyl group at C-9 is situated equatorially and that the solid compound adopts the conformation of cross-structure for the decadien-ring with the groups $C_{14}H_3$ and $C_{15}H_2OH$ situated parallel on the same side (see structure **2**).



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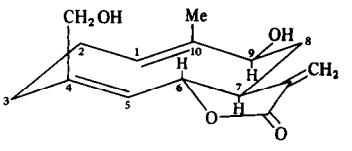

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EXPERIMENTAL

Extraction and separation. Dried flowers of *Centaurea aspera* var. *stenophylla*, collected at Saler (Valencia), during June–July and authenticated by Dr. J. Mansanet, Professor of Botany at Valencia University (6.6 kg) were extracted exhaustively first with hexane and afterwards with EtOH. The alcoholic extract reduced *in vacuo* to about 1 l. was treated with H_2O (2 l.) and an excess of lead acetate (20 g). After 48 hr the extract was filtered and the EtOH was removed again *in vacuo* and the remaining aq. soln was continuously re-extracted with Et_2O. The Et_2O left a bitter syrup of 67.39 g (1.02% of dry plant).

This syrup was chromatographed on a silica gel (300 g) column, from which C_6H_6–EtOAc eluted successively eight crystalline compounds. Giving in the first parenthesis the proportions of C_6H_6–EtOAc and in the later parenthesis the amount of compound separated, they were: Fr. 1 (100:0) benzoic acid (45 mg), Fr. 2 (85:15) *p*-hydroxybenzoic acid (23 mg), Fr. 3 (80:20) apigenine (520 mg), Fr. 4 (80:20) 6-methoxyluteoline (360 mg), Fr. 5 (75·25) melitensin + 11,13-dehydromelitensin (105 mg), Fr. 6 (60:40) stenophyllolide (574 mg), Fr. 7 (25:75) (ethyl 7-*O*-apigeninglucuronate (63 mg), Fr. 8 (25·75) sitosterol and stigmasterol glucosides (140 mg).

Apigenin (4′,5,7-*trihydroxyflavone*). Mp 347° (EtOH), IR ν_{max} cm^{-1}: 3500–2300 (OH), 1640 (CO flavone) 1600, 1550, 1500, 1450, 1350, 1240, 1180, 825, ^1H NMR (DMSO-d_6, 60 MHz). δ12.9 (*s*, 1H, OH-5), 7 9 (*d*, 2H, J = 9 Hz, H-2′, H-6′), 6.95 (*d*, 2H, J = 9 Hz, H-3′, H-5′), 6.7 (*s*, 1H, H-3), 6.4 (*d*, 1H, J = 2 Hz, H-8), 6.2 (*d*, 1H, J = 2 Hz, H-6); UV λ_{max}^{MeOH} nm: 268, 297 sh, 326 with shifts expected by adding NaOMe, NaOAc, $AlCl_3$ [12] Apigenin triacetate, mp 184–185° (lit 182° [13]) with expected IR, ^1H NMR and UV properties.

6-*Methoxyluteolin* (6-*methoxy*-3′,4′,5,7-*tetrahydroxyflavone*) Yellow crystals from MeOH–CH_2Cl_2, mp 272°, IR ν_{max} cm^{-1}: 3500–2500 (OH), 1650 (CO flavone), 1610, 1570, 1500, 1460, 1380, 1280, 1200, 1160 and 840; ^1H NMR (DMSO-d_6, 60 MHz)· δ13 1 (*s*, 1H, HO-5), 7.4 (*m*, 2H, H-2′, H-6′), 6.9 (*d*, 1H, J = 8.6 Hz, H-5′), 6.66 (*s*, 1H, H-8), 6.58 (*s*, 1H, H-3), 3.78 (*s*, 3H, OMe), UV λ_{max} nm: 225, 273, 347 with expected UV shifts by adding NaOMe, NaOAc, H_3BO_3, $AlCl_3$; MS *m/z* (rel. int): 316 $[M]^+$ (100), 301 $[M-15]^+$ (81.5), 298 $[M-H_2O]^+$ (56.9). 6-Methoxy-3′,4′,5,7-tetraacetoxyflavone, mp 195–198°, which showed an IR band of a phenolic acetate (1775 cm^{-1}), but not of an OH group and in addition it gave the expected signals in ^1H NMR spectrum.

11,13-*Dehydromelitensin* (8,15-*dihydroxyelema*-1,3,11-*trien*-12,6-*olide*). This was eluted from the column in fraction 5, mixed with melitensin, from which was separated by re-chromatography on silica gel, eluted with hexane–Et_2O (4:6), mp 137° (hexane–Et_2O), $[\alpha]_D^{20}$ +85 7° ($CHCl_3$); IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3590 (free OH), 3400 (asoc. OH), 3080, 1765 (CO of γ-lactone), 1640, 1400, 1380, 1250, 1130, 1050, 970 and 920; ^1H NMR (CD_3COCD_3, 60 MHz): δ5.97 (2*d*, 2H, H-13), partially overlapped with 5.9 (*dd*, 1H, J = 9.3 and 18.7 Hz, H-1); 5.4 and 4.9 (2*d*, 2H, J = 2 Hz, H-3); 5.06 and 4.85 (2 *dd*, 2H, H-2), 4.30 (*t*, 1H, J = 11.3 Hz, H-6), 3.98 (*br s*, 2H, H-15), partially overlapped with 4.1–3.6 (*m*, 1H, H-8), 3.85 and 3.05 (2 *br s*, 2OH), 2.63 (*tt*, 1H, J = 2.7 and 11.3 Hz, H-7), 2.45 (*d*, 1H, J = 11.3 Hz, H-5), 1.73 (*m*, 2H, H-9), 1.13 (*s*, 3H, H-14); MS *m/z* (rel. int.): 264 $[M]^+$ (0.18), 249 $[M-15]^+$ (0.42), 246 $[M-H_2O]$ (0.4). 11,13-Dehydromelitensin diacetate, mp 121–122°, $[\alpha]_D^{20}$ +95.45° ($CHCl_3$); IR ν_{max} cm^{-1}. 3110, 2980–2860, 1775 (CO of γ-lactone), 1730 (CO acetate), 1650, 1385, 1230, 1260, 965, 925; ^1H NMR ($CDCl_3$, 60 MHz): δ6.20 and 5.63 (2*d*, 2H, J = 2.7 Hz, H-13), 5.86 (*dd*, 1H, J = 7.3 and 2.0 Hz, H-1), 5.48 and 4.88 (2*d*, 2H, J = 1.5 Hz, H-3), partially overlapped with 5.4–4.9 (*m*, 2H, H-2), 4.57 (*br s*, 2H, H-15), partially overlapped with 4 5–4.3 (*h.m*, 1H, H-8), 4.26 (*t*, 1H, J = 11.3 Hz, H-6), 2.87 (*tt*, 1H, J = 11.3 and 2.7 Hz, H-7), 2.46 (*d*, 1H, J = 11.3 Hz, H-5), 2.12 and 2.10 (2*s*, 6H, 2 × OAc), 2.0–1.2 (*m*, 2H, H-9), 1.15 (*s*, 3H, H-14).

Melitensin (8,15-*dihydroxyelema*-1,3-*dien*-12,6-*olide*). Crystalline product, mp 162–163° (hexane–Et_2O); $[\alpha]_D^{20}$ +62.2° (EtOH); IR ν_{max} cm^{-1}: 3320 (OH), 3100, 2990–2830, 1765 (CO of γ-lactone), 1665, 1640, 1450, 1375, 1230, 1040, 915, 890; ^1H NMR (CD_3COCD_3, 60 MHz): δ6.08 (*dd*, 1H, J = 10.6 and 18.6 Hz, H-1), 5.51 and 5.01 (2 *dd*, 2H, J = 2 Hz, H-3), 5.20 and 4.95 (*m*, 2H, H-2), 4.47 (*dd*, 1H, J = 12 and 10 Hz, H-6), 4.05 (*br s*, 2H, H-15), 3.84 (*m*, 1H, H-8), 2.98–2.72 (*m*, 1H, H-11), 2.63 (*q*, J = 7.3 Hz, H-7), 2.35 (*d*, 1H, J = 12 Hz, H-5), 1.70 (*m*, 2H, H-9), 1.31 (*d*, 1H, J = 7.3 Hz, H-13), 1.15 (*s*, 3H, H-14); MS *m/z* (rel. int.): 266 $[M]^+$ (0.07), 251 $[M-15]^+$ (0.31), 248 $[M-18]^+$ (0.43). Melitensin-diacetate, mp 139–140° (hexane–CH_2Cl_2), $[\alpha]_D^{20}$ +62.3° (EtOH), IR ν_{max} cm^{-1}: 3110, 2980–2880, 1780 (CO of γ-lactone), 1730 (CO acetate), 1650, 1385, 1240, 970 and 925.

Stenophyllolide (9,15-*dihydroxygermacra*-1(10),4,11-*trien*-12,6-*olide* (**1**). Decomposes slowly when heated over 200°, $[\alpha]_D^{20}$ +72.4° (MeOH). Found by high resolution MS 246.1262, calculated for $C_{15}H_{18}O_3$ $[M-H_2O]^+$ = 246.1256, IR ν_{max} cm^{-1}: 3300 (OH), 2990–2860, 1755 (CO of γ-lactone), 1655, 1435, 1250, 1135, 1030 and 960; MS *m/z* (rel. int.): 264 $[M]^+$ (0.6), 246 $[M-H_2O]^+$ (4.5), 233 $[M-CH_2OH]^+$ (4.45); ^1H NMR values at 200 MHz are shown in Table 1 and they were all confirmed by double resonance studies. Diacetate of stenophyllolide, crystals, which decomposed slowly when heated over 200°, $[\alpha]_D^{20}$ +80.7° (EtOH). Found by high resolution MS 289.1441, calculated for $C_{17}H_{21}O_4$ $[M-59]^+$ 289.144; IR ν_{max} cm^{-1}: 3050, 2990–2860, 1760 (CO of γ-lactone), 1735 (CO acetate), 1375, 1250, 1235 and 970; ^1H NMR values, confirmed by double resonance studies at 200 MHz (Table 1).

Ethyl 7-O-*apigenin-glucuronate.* Mp 220–223° ($CHCl_3$–EtOH); IR ν_{max} cm^{-1}: 3350 (OH), 1740 (CO ester), 1660 (CO flavone), 1610, 1500, 1450, 1350, 1180, 1100, 1010, 840; ^1H NMR (DMSO-d_6, 60 MHz)· δ8.0 (*d*, 2H, J = 8.6 Hz, H-2′, H-6′), 7.2–6.8 (*m*, 4H, H-3, H-3′, H-5′, H-8), 6.53 (*d*, 1H, J = 2 Hz, H-6), 5 8–5.2 (*br s*, 4H, CHOH of sugar), 4.18 (*q*, 2H, J = 6 6 Hz, $CO_2CH_2CH_3$), 1.24 (*t*, 3H, J = 6.6 Hz, $CO_2CH_2C\underline{H}_3$). Pentaacetate derivative, mp 202–204°, IR ν_{max} cm^{-1} 1750 (CO acetate), 1650 (CO flavone), 1610, 1500, 1450, 1370, 1210, 1170, 840; ^1H NMR ($CDCl_3$, 60 MHz): δ7.9 (*d*, 2H, J = 8.6 Hz, H-2′, H-6′), 7.25 (*d*, 2H, J = 8.6 Hz, H-3′, H-5′), 7 05 (*d*, 1H, J = 1 8 Hz, H-8), 6.74 (*d*, 1H, J = 1.8 Hz, H-6), 6.6 (*s*, 1H, H-3), 5 4 (*br s*, 5H, CHOAc of sugar), 4.23 (*q*, 2H, J = 7.3, $CO_2C\underline{H}_2CH_3$), 2.48, 2.38 and 2.1 (3*s*, 15H, 5Ac–), 1.28 (*t*, 3H, J = 7.3 Hz, $CO_2CH_2C\underline{H}_3$). Glucuronate hydrolysis was carried out on 75 mg of compound heated to reflux with 2 N HCl and a few drops of EtOH for 4 hr; the aglycone was extracted with $CHCl_3$ and identified as apigenine by IR, UV and mp. The sugar in the aq. hydrolysate was neutralized by filtration through Lewatit M5OOG2 (an ion exchange resin), dried by evaporation *in vacuo*, transformed into the trimethylsilylether and identified by gas chromatography as glucuronic acid, using an authentic sample of this acid for comparison under the same conditions The linkage between the sugar and the aglycone was made through 7-O, since the glycoside takes 180 min for a full hydrolysis (60 min would be enough for a 3-O-linkage of glucuronic acid) [14]. The same linkage was shown by the UV λ_{max}^{MeOH} at 269 and 337 nm of glucuronate, since the later band suffers the following bathochromic shifts: 53 nm in NaOAc or NaOMe (OH-4′ free), 48 nm with $AlCl_3$ (free OH-3); but the former band is unchanged with NaOAc (bound O-7).

Sitosterol glucoside and stigmasterol glucoside. Crystals of mp 282–284° (EtOH), IR ν_{max} cm^{-1}: 3400 (OH), 2960–2850, 1640, 1465, 1380, 1070, 1020, 800; acetate derivative mp 167–169°, $[\alpha]_D^{20}$

$-30°$ ($CHCl_3$); IR ν_{max} cm^{-1}: 2960–2870, 1750 (CO acetate), 1460, 1440, 1385, 1375, 1220, 1070 and 850. Hydrolysis of glycosides with 2 N HCl gave an aglycone identified as a mixture of sitosterol and stigmasterol and a sugar, identified as glucose by GC of the TMS derivative.

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REFERENCES

1. Sánchez Parareda, I., Sánchez Parareda, J. and Viguera, J. M. (1968) *Anal. Fís. Quím.* **64**, 633.
2. Fisher, N. H., Oliver, E J., and Fisher, H. D. (1979) *Prog. Chem. Org. Nat. Prod.* **38**, 286.
3 Goudard, M., Favre-Bonvin, J., Lebreton, P. and Chopin, J. (1978) *Phytochemistry* **17**, 145.
4. Kupchan, S. M., Siegel, C. W., Hemingway, R. J. and Knox, J. R. (1969) *Tetrahedron* **25**, 1603.
5. Imre, S., Oztünc, A. and Wagner, H. (1977) *Phytochemistry* **16**, 799.
6. Krishnaswamy, N. R., Seshadri, T. R. and Tahir, P. J. (1968) *Ind. J. Chem.* **6**, 676.
7. González, A. G., Arteaga, J. M., Bermejo, J. and Bretón, J. L. (1971) *An. Quím.* **67**, 1243.
8. Gonzalez, A. G., Bermejo, J., Cabrera, I. and Massanet, G. M. (1974) *An. Quím.* **70**, 74.
9. Arnó, M., Garcia, B., Pedro, J. R. and Seoane, E. (1983) *Tetrahedron Letters* **24**, 1741.
10. Zapesochnaya, G. G. and Banlkovskii, A. I. (1976) *Khim. Prir. Soedin* **6**, 814. *Chem. Abst.* **86**, 117590j.
11. Samek, Z. (1970) *Tetrahedron Letters* 671.
12. Thomas, M. B. and Mabry, T. J. (1968) *Phytochemistry* **7**, 787.
13. Schmid, L. and Waschkan, A. (1928) *Monatshefte* **49**, 83.
14. Harborne, J. B. (1965) *Phytochemistry* **4**, 107.