*Phytochemistry*, Vol. 31, No. 3, pp. 1003–1007, 1992
Printed in Great Britain.

0031-9422/92 $5.00+0.00


# ACYLATED FLAVONOLS AND OTHER CONSTITUENTS FROM *GALEANA PRATENSIS**

EMMA MALDONADO,† RUBÉN A. TOSCANO, CLAUDIA MANCERA,‡ MA. TERESA TRIPP‡ and ALFREDO ORTEGA

Instituto de Química, Universidad Nacional Autónoma de México, Circuito Exterior, Ciudad Universitaria, Coyoacán 04510 México, D. F.; ‡Departamento de Química Orgánica, División de Estudios Superiores, Facultad de Ciencias Químicas e Industriales, Universidad Autónoma del Estado de Morelos, Ave. Universidad 1001, Cuernavaca, Morelos, México

(*Received in revised form* 23 *August* 1991)

**Key Word Index**—*Galeana pratensis*; Heliantheae; Compositae; acyl flavonols; sesquiterpene lactones; pratensins A and B.

**Abstract**—The chemical investigation of *Galeana pratensis* afforded, in addition to known sesquiterpene lactones, the new acyl flavonoids, pratensins A (5,7-dihydroxy-3,6,4′-trimethoxy-8-tigloyloxyflavone) and B [5,4′-dihydroxy-3,6,7-trimethoxy-8-(2-methylbutyroyloxy)flavone]. Structures of pratensins A and B were elucidated by spectroscopic methods, chemical transformations and X-ray analysis.

## INTRODUCTION

No chemical investigation has been reported on the small genus *Galeana* (three species), which belongs to the subtribe Bahiinae, tribe Heliantheae. We have now studied *Galeana pratensis* (H.B.K.) Rydberg which, in addition to some known compounds, contained two acylated flavonols. Compounds acylated at the flavonoid nucleus are rare in the Compositae family [1] except for the tribe Astereae [2–4]. This is the first report of their occurrence in the Heliantheae.

## RESULTS AND DISCUSSION

Several known compounds such as β-sitosterol, stigmasterol, ursolic acid, mexicanin I [5], 8-epihelenalintiglate [6] and 4-epipulchellin-2-*O*-tiglate [7] were isolated from *Galeana pratensis*. In addition, three flavonols were also isolated. One of them was identified as santin (**1**) (5,7-dihydroxy-3,6,4′-trimethoxyflavone) [8, 9]. The other two, pratensins A (**2**) and B (**10**), were shown to be two new acyl flavonols.

Pratensin A (**2**) exhibited UV absorptions in methanol (Table 1) indicative of its flavone or flavonol nature. The NaOAc induced shift of band II (+4 nm) suggested the presence of a OH-7 group. The IR spectrum of **2** showed absorptions for hydroxyl and γ-pyrone carbonyl groups, double bonds and an ester (1735 $cm^{-1}$) function. The ester was characterized as a tiglate by the $^1H$ NMR (Table 2) and mass spectra. The $^1H$ NMR spectrum also showed signals for an AA′BB′ system, indicating a 4′-substituted ring B, three methoxy and two phenolic hydroxy groups, one of them localized at C-5.

The presence of a 4′-methoxy group in **2** was established by the $[B_2]^+$ fragment in the mass spectrum, as well as from the C-1′–C-6′ $^{13}C$ NMR chemical shifts (Table 3). The $^{13}C$ NMR data also revealed that a second methoxyl should be placed at C-3, as the signals for C-2, C-4 and C-10 were shifted by *ca* 4, 2 and 1 ppm to lower field, respectively, compared to those induced by a hydroxyl [10].

The chemical shifts observed for C-5–C-10 in pratensin A are in agreement with structure **2**, if it is considered that the acylation of a phenolic hydroxyl induces downfield shifts of the *ortho* and *para* carbons, and the opposite shift for the *ipso* carbon [11]. Thus taking compound **3** as a model [10], the *ipso* C-8 in **2** was shielded by 6.3 ppm. The *ortho* and *para* carbons, C-7, C-9 and C-5, were shifted downfield by 3.6, 2.9 and 5 ppm, respectively.

Methylation of **2** provided further evidence of the proposed structure, as in the 7-methyl derivative **4**, the expected lowfield shifts of the carbons *ipso* (1.2 ppm, C-7), *ortho* (3.9 ppm, C-6; 4.1 ppm, C-8) and *para* (3.0 ppm, C-10) were observed.

Table 1. UV spectral data of compounds **2**–**11** (in MeOH) [$\lambda_{max}$ nm (log ε)]

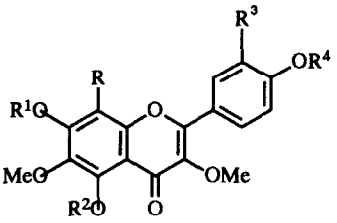
| Compound | Band I | Band II | Others |
|---|---|---|---|
| **2** | 333 (4.31) | 270 (4.35) | |
| **2*** | 368 (4.22) | 274 (4.52) | 296 sh (4.33) |
| **4** | 330 (4.29) | 272 (4.28) | |
| **5** | 328 (4.21) | 288 (4.30) | 215 (4.24); 383 sh (3.76) |
| **6** | 328 (4.21) | 272 (4.31) | 298 sh (4.19) |
| **7** | 328 (4.16) | 269 (4.12) | 230 (4.13) |
| **8** | 329 (4.12) | 254 (3.98) | 228 (4.07) |
| **9** | 308 (4.30) | 254 (4.37) | |
| **11** | 312 (4.39) | 242 (4.38) | 211 (4.40); 403 sh (3.66) |

*Determined in methanol–sodium acetate.

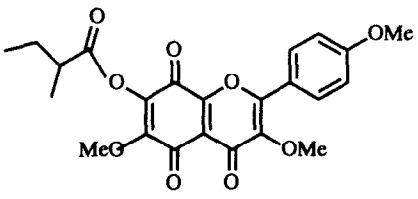
*Contribution No. 1081 of Instituto de Química, UNAM.
†Author to whom correspondence should be addressed.

Table 2. $^1$H NMR spectral data of compounds **2**–**11** (80 MHz, $CDCl_3$, TMS as internal standard)

| H | 2 | 4 | 5 | 6 | 7 | 8 | 9 | 11 |
|---|---|---|---|---|---|---|---|---|
| 2′,6′ *d* | 7.93 (9) | 7.94 (9) | 8.10 (9.5) | 7.94 (9.5) | 7.92 (9) | 7.88 (9) | 7.98 (9) | 8.02 (9) |
| 3′,5′ *d* | 6.92 (9) | 6.94 (9) | 6.98 (9.5) | 6.94 (9.5) | 6.95 (9) | 6.94 (9) | 7.20 (9) | 6.94 (9) |
| OCOR | 7.23 *qq* (7.5, 1.5) | 7.26 *qq* (7.5, 1.5) | | 2.73 *qn* (7) | 2.69 *qn* (7) | 2.72 *qn* (7) | 2.75 *br qn* (7) | 2.64 *qn* (7) |
| | 2.01 *br q* (1.5) | 2.01 *br q* (1.5) | | 1.78 *m* | 1.72 *br h* (7) | 1 72 *br h* (7) | 1.77 *br h* (7) | 1.69 *br h* (8) |
| | 1.92 *brd* (7.5) | 1.94 *brd* (7.5) | | 1.38 *d* (7) | 1.35 *d* (7) | 1.34 *d* (7) | 1.39 *d* (7) | 1.31 *d* (7) |
| | | | | 1.04 *t* (7.5) | 1.03 *t* (7) | 1.03 *t* (7) | 1.05 *t* (7) | 1.03 *t* (8) |
| OMe *s* | 4.04 | 4.01 | 4.10 | 4.03 | 3.94 | 3.86 | 3.99 | 4.16 |
| *s* | 3.85 | 3.94 | 3.95 | 3.86 | 3.87 | 3.86 | 3.88 | 3.94 |
| *s* | 3.82 | 3.85 | 3.88 | 3.81 | 3.83 | 3.75 | 3.78 | 3.84 |
| | | 3.82 *s* | 3.84 *s* | | | | | |
| OH or | 6.42 *br* | | 5.35 *s* | 6.38 *br* | 11.38 *br* | 2.50 *s* | 2.50 *s* | |
| OAc | | | 10.85 *s* | | 2.34 *s* | 2.34 *s* | 2.34 *s* | |



**1** R = $R^1$= $R^2$= $R^3$=H; $R^4$= Me
**2** R = OTig; $R^1$= $R^2$=$R^3$= H; $R^4$= Me
**3** R = $R^3$= OMe; $R^1$ = $R^2$= $R^4$= H
**4** R = OTig; $R^1$= $R^4$= Me; $R^2$= $R^3$= H
**5** R = OH; $R^1$= $R^4$= Me; $R^2$= $R^3$= H
**6** R = O-2-Mebu; $R^1$= $R^2$= $R^3$= H; $R^4$= Me
**7** R = O-2-Mebu; $R^1$= Ac; $R^2$= $R^3$= H; $R^4$ = Me
**8** R = O-2-Mebu; $R^1$= $R^2$= Ac; $R^3$= H; $R^4$ = Me
**9** R = O-2-Mebu; $R^1$= Me; $R^2$= $R^4$= Ac; $R^3$= H
**10** R = O-2-Mebu; $R^1$= Me; $R^2$= $R^3$= $R^4$= H



**11**

Alkaline hydrolysis of **4** gave the desacyl derivative **5**, proving that the acyl moeity was bonded to C-8 oxygen. As expected, the mass spectrum exhibited a $[M-1]^+$ fragment with a relative intensity >20% typical of flavonols with a free hydroxyl at C-8 [10]. In the $^{13}$C NMR spectrum of **5**, the expected downfield shifts for C-8 and the upfield shifts for C-5, C-7 and C-9 were observed as well as a bathochromic shift of band II (16 nm) in the UV spectrum. Compound **5** has been synthesized [12, 13] and the reported characteristics are identical with those of **5** obtained in our study.

When pratensin A (**2**) was hydrogenated (Pd/C 5%, EtOAc, 1 hr), the dihydro derivative **6** was obtained. Upon acetylation compound **6** gave the mono and diacetyl derivatives **7** and **8**.

A second hydrogenation of **2** under the above conditions also afforded **6**, but the reaction mixture was not worked-up immediately. After 12 hr, it had changed from yellow into orange-reddish. At this time compound **11** was obtained. The spectral data revealed its *p*-quininoid nature. Thus, the UV spectrum showed absorptions at $\lambda$ 242, 312 and 403 nm comparables to those exhibited by similar *p*-quinones [14] and the IR showed a carbonyl absorption at 1695 $cm^{-1}$, with shoulders at 1705, 1685 and 1676 $cm^{-1}$, as well as a strong band at 1608 $cm^{-1}$ attributed to the quinone double bonds. In the mass spectrum, the characteristic quininoid fragments $[M+2]^+$ and $[M-acyl+2]^+$ were present. Finally, four carbonyl signals were observed in the $^{13}$C NMR spectrum, two corresponding to the quinone, one to C-4 and

Table 3. $^{13}$C NMR spectral data of compounds **2**, **4**, **5**, **9** and **11**

| C | 2* | 4* | 5* | 9† | 9‡ | 11* |
|---|---|---|---|---|---|---|
| 2 | 154.8 | 155.5 | 155.7 | 153.6 | 152.9 | 154.0 |
| 3 | 138.0 | 138.2 | 137.7 | 139.6 | 138.5 | 133.7 |
| 4 | 178.3 | 178.4 | 178.8 | 173.7 | 173.2 | 177.2 |
| 5 | 149.5 | 150.1 | 144.1 | 145.3 | 144.2 | 173.3 |
| 6 | 131.7 | 135.6 | 135.8 | 141.2 | 140.1 | 149.2 |
| 7 | 150.4 | 151.6 | 147.8 | 150.4 | 149.7 | 146.9 |
| 8 | 118.9 | 123.0 | 130.5 | 130.3 | 130.0 | 170.6 |
| 9 | 143.7 | 143.1 | 140.6 | 143.0 | 142.1 | 153.2 |
| 10 | 103.6 | 106.7 | 106.9 | 113.7 | 112.6 | 116.9 |
| 1′ | 122.2 | 121.9 | 122.6 | 127.9 | 126.8 | 121.2 |
| 2′,6′ *d* | 129.6 | 129.7 | 130.2 | 129.7 | 129.1 | 129.7 |
| 3′,5′ *d* | 114.3 | 114.4 | 114.2 | 121.7 | 121.9 | 114.5 |
| 4′ | 161.6 | 161.8 | 161.4 | 152.3 | 151.9 | 161.5 |
| 1″ | 165.2 | 165.2 | | 173.0 | 171.5 | 173.0 |
| 2″ | 126.8 | 127.2 | | 41.1 | 39.7 *d* | 40.2 *d* |
| 3″ | 140.4 *d* | 141.2 *d* | | 26.8 *t* | 25.9 *t* | 26.1 *t* |
| 4″ | 12.1 *q* | 12.1 *q* | | 11.7 *q* | 11.0 *q* | 11.2 *q* |
| 5″ | 14.6 *q* | 14.4 *q* | | 16.9 *q* | 16.3 *q* | 16.3 *q* |
| 7-OMe | | 61.4 *q* | 61.1 *q* | 61.8 *q* | 61.4 *q* | |
| 6-OMe *q* | 60.4 | 60.7 | 60.4 | 61.4 | 61.2 | 61.1 |
| 3-OMe *q* | 59.8 | 59.8 | 59.6 | 60.2 | 59.6 | 59.5 |
| 4′-OMe | 55.5 *q* | 55.5 *q* | 55.4 *q* | | | 55.5 *q* |

*Determined at 20 MHz in DMSO-$d_6$. †Determined at 75 MHz in $CDCl_3$ (OAc $\delta$: 20.3, 21.2, 168.9, 169.2). ‡Determined at 75 MHz in DMSO-$d_6$ (OAc $\delta$: 20.3, 20.5, 168.5, 168.6).

the fourth to the ester carbonyl, whose presence was also supported by the IR, MS and $^{13}$C NMR spectra.

Production of the quinoid compound **11** from **6** must be by way of a transacylation process, in which the acyl group at the C-8 oxygen migrates to the C-7 oxygen. Thus, formation of the *para* quininoid system should be a highly favoured process, in which the presence of the catalyst is necessary for the reaction to occur, as when **6** was left in ethyl acetate (without catalyst) it was recovered unchanged after 24 hr. Also a free phenolic group at C-7 is required for the acyl group to leave the C-8 oxygen, seeing that **4** did not form the quinone when it was left in Pd/C–EtOAc for 48 hr.

Pratensin B (**10**) was isolated from the mother liquors of santin (**1**) as the diacetyl derivative **9**, whose spectral data showed that it possesses three methoxyl and three acyl groups (one 2-methylbutyrate and two acetates) linked to a flavonol with the same oxygenation pattern as **2**. In **9**, however, one acyl group is located at C-4′, as the signal for H-3′ and H-5′ appear at lower field than those for compounds **2**–**8**. The chemical shift of C-1′–C-6′ in the $^{13}$C NMR spectrum of **9** confirmed the above assumption. Nevertheless, the relative position at the different groups in **9** could not be established by spectral methods, consequently the X-ray diffraction molecular structure (Fig. 1) was obtained. It showed that **9** is the 5,4′-diacetoxy-3,6,7-trimethoxy-8-(2-methylbutyroyloxy) flavone. Therefore, the structure of the natural pratensin B should be represented as **10**.

In addition to the mentioned compounds, *G. pratensis* also possesses a complex mixture of acylglucopyranoses, which could not be separated. These uncommon type of compounds have been isolated from *Bahia* species [15] and their presence in *Galeana* supports its inclusion in the subtribe Bahiinae.

## EXPERIMENTAL

Ground, dried aerial parts of *G. pratensis* (H.B.K.) Rydberg (3.3 k) collected in Cuernavaca, Morelos, México (specimen deposited in the Herbarium of the Instituto de Biología, UNAM, AOH-118), were extracted with hexane, EtOAc and MeOH affording 83, 136 and 254 g of residue, respectively. The hexane

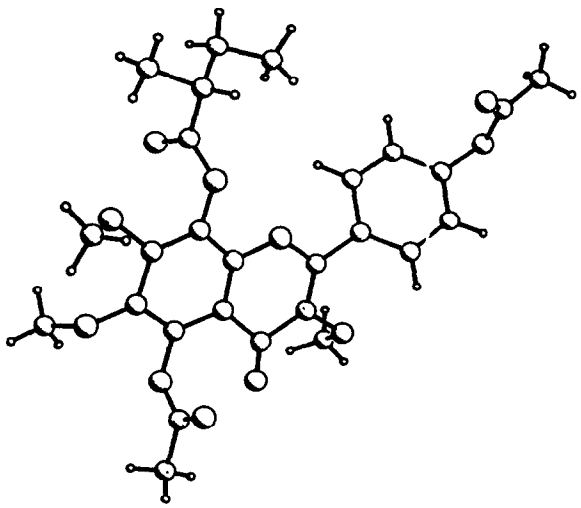


Fig. 1. Stereoscopic view of diacetylpartensin B.

extract was fractionated on a silica gel column eluted with a hexane–EtOAc gradient. Frs eluted with hexane–EtOAc (19:1) gave a mixture of β-sitosterol and stigmasterol (1.33 g). Frs eluted with hexane–EtOAc (7:3), (3:2) and (2:3) were combined with the EtOAc and MeOH extracts and decolourized with activated charcoal. After successive CC (silica gel Merck G) of the resulting residue, ursolic acid (15.8 mg), mexicanin I (2.66 g), 8-epihelenalintiglate (3.05 g), 2-*O*-acetyl-4-epipulchellin (0.77 g), santin (164 mg), pratensin A (2.76 g) and diacetyl pratensin B (73 mg) were obtained. Diacetylpratensin B was isolated after esterification of the mother liquors of santin. Known compounds were identified by comparing their physical and spectral data with those described in literature.

*Pratensin A* (**2**). Pale yellow needles, mp 184–186° (EtOAc–hexane); IR $\nu_{max}^{CHCl_3}$ $cm^{-1}$: 3493, 1735, 1655, 1627, 1580; UV, $^1H$ and $^{13}C$ NMR: see Tables 1–3; EI-MS *m/z* (rel. int.): 442 $[M]^+$ ($C_{23}H_{22}O_9$; 43); 427 $[M-Me]^+$ (1); 359 $[M-C_5H_7O]^+$ (82); 345 $[359-CH_2]^+$ (7); 316 $[359-Ac]^+$ (2); 301 $[316-Me]^+$ (4); 197 $[A_1-C_5H_7O]^+$ (3); 169 $[197-CO]^+$ (3); 161 $[B_1-H]^+$ (44); 135 $[B_2]^+$ (9); 83 $[C_5H_7O]^+$ (100); 55 $[C_4H_7]^+$ (52).

*Diacetylpratensin B* (**9**). Crystals, mp 164–165° (EtOAc–hexane). IR $\nu_{max}^{CHCl_3}$ $cm^{-1}$: 1767, 1717, 1651, 1627; UV, $^1H$ and $^{13}C$ NMR: see Tables 1–3; EI-MS *m/z* (rel. int.): 528 $[M]^+$ ($C_{27}H_{28}O_{11}$; 2); 486 $[M-C_2H_2O]^+$ (10); 402 $[486-C_5H_8O]^+$ (100); 387 $[402-Me]^+$ (7); 359 $[M-2C_2H_2O-C_5H_9O]^+$ (26); 345 $(359-CH_2]^+$ (12); 211 $[A_1-C_2H_2O-C_5H_9O]^+$ (3); 161 $[B_2-C_2H_2O]^+$ (13); 85 $[C_5H_9O]^+$ (10); 57 $[C_4H_9]^+$ (38); 43 $[Ac]^+$ (32).

7-O-*Methylpratensin A* (**4**). An ethereal soln of $CH_2N_2$ was added dropwise to a suspension of **2** (146 mg) in $Et_2O$ (10 ml) at 0°. The reaction mixture was left to stand overnight. The crude mixture was purified by CC (silica gel; hexane–EtOAc 4:1) to give 118.2 mg of **4** as yellow crystals, mp 136–138° from EtOAc–hexane. IR $\nu_{max}^{CHCl_3}$ $cm^{-1}$: 3360, 1733, 1650, 1600, 1560; UV, $^1H$ and $^{13}C$ NMR: see Tables 1–3; EI-MS *m/z* (rel. int.): 456 $[M]^+$ ($C_{24}H_{24}O_9$, 27); 441 $[M-Me]^+$ (1); 373 $[M-C_5H_7O]^+$ (100); 359 $[373-CH_2]^+$ (7); 315 $[373-Me-Ac]^+$ (3); 211 $[A_1-C_5H_7O]^+$ (3); 135 $[B_2]^+$ (8); 83 $[C_5H_7O]^+$ (42); 55 $[C_4H_7]^+$ (26).

8-O-*Detigloyl-7-O-methylpratensin A* (**5**). An ethereal soln of $CH_2N_2$ was added dropwise to a soln of **2** (111 mg) in $Et_2O$–MeOH (15 ml, 5:1). After completion of the reaction the solvent was removed, the residue was dissolved in MeOH and MeONa was added. The reaction mixture was refluxed by 15 min and neutralized with HOAc. The pptd NaOAc was filtered off and the filtrate concd and crystallized from $Me_2CO$–hexane to give 44.3 mg **5**, mp 214–216°; IR $\nu_{max}^{CHCl_3}$ $cm^{-1}$: 3554, 1655, 1599; UV, $^1H$ and $^{13}C$ NMR: see Tables 1–3; EI-MS *m/z* (rel. int.): 374 $[M]^+$ ($C_{19}H_{18}O_8$; 100); 373 $[M-1]^+$ (22); 359 $[M-Me]^+$ (75); 341 $[359-H_2O]^+$ (22); 316 $[359-Ac]^+$ (13); 313 $[M-H_2O-Ac]^+$ (7); 301 $[316-Me]^+$ (7); 298 $[341-Ac]^+$ (8); 197 $[A_1-Me]^+$ (18); 169 $[A_1-Ac]^+$ (13); 161 $[B_1-H]^+$ (16); 135 $[B_2]^+$ (26); 119 $[B_1-Ac]^+$ (24).

*Dihydropratensin A* (**6**). Compound **2** (71.8 mg) was hydrogenated in EtOAc (10 ml) over 5% Pd/C (54 mg) for 1 hr. Filtration and solvent evapn left a residue, which gave, after crystallization ($Me_2CO$–hexane), 64 mg **6**, mp 144–145°; IR $\nu_{max}^{CHCl_3}$ $cm^{-1}$: 3497, 1760, 1655, 1595, 1580; UV, $^1H$ and $^{13}C$ NMR: see Tables 1–3; EI-MS *m/z* (rel. int.): 444 $[M]^+$ ($C_{23}H_{24}O_9$; 11); 360 $[M-C_5H_8O]^+$ (100); 345 $[360-Me]^+$ (32); 342 $[360-H_2O]^+$ (6); 327 $[360-Me-H_2O]^+$ (6); 317 $[360-Ac]^+$ (4); 197 $[A_1-C_5H_9O]^+$ (3); 169 $[197-CO]^+$ (3); 161 $[B_1-H]^+$ (7); 135 $[B_2]^+$ (10); 85 $[C_5H_9O]^+$ (12); 57 $[C_4H_9]^+$ (39).

*Acetylation of dihydropratensin* (**6**). Compound **6** (60 mg) in pyridine (1 ml) and $Ac_2O$ (1 ml) was left to stand for 20 min and worked-up as usual to give, after CC (silica gel; hexane–EtOAc, 4:1), 12.5 mg **7** and 22.1 mg **8**. Compound **7**: yellow crystals, mp 157–158° ($Me_2CO$–hexane) IR $\nu_{max}^{CHCl_3}$ $cm^{-1}$: 3500, 1778, 1650, 1600, 1580; UV, $^1H$ and $^{13}C$ NMR: see Tables 1–3; EI-MS *m/z* (rel. int.): 486 $[M]^+$ ($C_{25}H_{26}O_{10}$; 12); 402 $[M-C_5H_8O]^+$ (43); 360 $[M-C_2H_2O-C_5H_8O]^+$ (92); 345 $[360-Me]^+$ (28); 342 $[360-H_2O]^+$ (9); 327 $[360-Me-H_2O]^+$ (9); 197 $[A_1-C_5H_9O-Ac]^+$ (3); 169 $[197-CO]^+$ (6); 161 $[B_1-H]^+$ (8); 135 $[B_2]^+$ (13); 85 $[C_5H_9O]^+$ (16); 57 $[C_4H_9]^+$ (100); 43 $[Ac]^+$ (39). Compound **8**: crystals, mp 154–156° (EtOAc–hexane): IR $\nu_{max}^{CHCl_3}$ $cm^{-1}$: 1780, 1642, 1620, 1608. UV, $^1H$ and $^{13}C$ NMR: see Tables 1–3; EIMS *m/z* (rel. int.): 528 $[M]^+$ ($C_{27}H_{28}O_{11}$; 6); 486 $[M-C_2H_2O]^+$ (26); 444 $[M-2C_2H_2O]^+$ (1); 443 $[M-C_2H_2O-Ac]^+$ (26); 402 $[486-C_5H_8O]^+$ (71); 360 $[M-2C_2H_2O-C_5H_8O]^+$ (100); 345 $[360-Me]^+$ (20); 342 $[360-H_2O]^+$ (7); 327 $[360-Me-H_2O]^+$ (3); 197 $[A_1-C_5H_9O-Ac]^+$ (3); 169 $[197-CO]^+$ (5); 161 $[B_1-H]^+$ (6); 135 $[B_2]^+$ (9); 85 $[C_5H_9O]^+$ (13); 57 $[C_4H_9]^+$ (89); 43 $[Ac]^+$ (57).

*Synthesis of compound* **11**. Compound **2** (91.3 mg) in EtOAc (10 ml) was hydrogenated with Pd/C (63 mg, 5%) as catalyst for 3 hr. Compound **6** was the only product from the reaction (comparative TLC). After 12 hr the yellow reaction mixture had turned orange-reddish. TLC showed that **6** had been transformed in the more polar compound **11**, which was isolated after the usual work-up and crystallization from $Me_2CO$–hexane, as orange-reddish needles (84.5 mg), mp 193–195°; IR $\nu_{max}^{CHCl_3}$ $cm^{-1}$: 1766, 1705 sh, 1695, 1685 sh, 1676 sh, 1630, 1608; UV, $^1H$ and $^{13}C$ NMR: see Tables 1–3; EIMS *m/z* (rel. int.): 444 $[M+2]^+$ (1); 443 $[M+1]^+$ (<1); 442 $[M]^+$ ($C_{23}H_{22}O_9$; 3); 360 $[M+2-C_5H_8O]^+$ (9); 358 $[M-C_5H_8O]^+$ (40); 343 $[358-Me]^+$ (6); 340 $[358-H_2O]^+$ (8); 315 $[358-Ac]^+$ (7); 147 $[B_1-Me]^+$ (12); 135 $[B_2]^+$ (16); 85 $[C_5H_9O]^+$ (31); 57 $[C_4H_9]^+$ (100). Compound **6** (4 mg) was also obtained.

*X-Ray structure determination of diacetylpratensin D* (**9**). Crystals of **9** ($C_{27}H_{28}O_{11}$) were monoclinic, with $a=8.343$ (2), $b=24.906$ (5) and $c=12.731$ (3) Å; $\beta=94.33°$ (2), $P_{21}$, $Z=4$, $D_c=1.33\ g\,cm^{-3}$ and $\mu=8.36\ cm^{-1}$. The intensity data were collected from a single crystal on a Nicolet P3/F difractometer with the $2\theta:\theta$ scan mode at 293 K ($2\theta_{max}=105°$) by used Ni-filtered Cu$K_\alpha$ radiation (1.54178 Å). The data were corrected for Lorentz and polarization factors, but not for absorption. Of 3412 unique reflections collected, 2995 reflections greater than $3\sigma(F)$ were considered observed.

The structure was solved by direct methods using SHELXTL [16] and refined by blocked-cascade least-square calculations. All non-hydrogen atoms except C-20 and C-50 were refined anisotropically. The hydrogen atoms were added at idealized positions with fixed isotropic $U=0.06\ Å^2$. The final discrepance factors were $R=0.065$ and $R_w=0.085$; the final difference map had no peaks greater than $0.26\ eÅ^{-3}$.

*Acknowledgements*—We are very grateful to Messrs J. Cárdenas, R. Gaviño, L. Velasco and M. Torres for technical assistance. We also thank J. L. Villaseñor (Botany Department, Instituto de Biología, U.N.A.M.) for botanical classification of the plant material and to R. Villena for collection of the X-ray data.

## REFERENCES

1. Wollenweber, E. (1985) *Phytochemistry* **24**, 1493.
2. Bohlmann, F., Jakupovik, J. and Lonitz, M. (1977) *Chem. Ber.* **110**, 301.
3. Bohlmann, F., Zdero, C., Grenz, M., Dhar, A. K., Robinson, H. and King, R. M. (1981) *Phytochemistry* **20**, 281.
4. Bohlmann, F., Grenz, M., Dhar, A. K. and Goodmann, M. (1981) *Phytochemistry* **20**, 105.

5. Domínguez, E. and Romo, J. (1963) *Tetrahedron* **19**, 1415.
6. Bohlmann, F., Mixra, L. N. and Jakupovic, J. (1985) *Phytochemistry* **24**, 1021.
7. Bohlmann, F., Zdero, C. and Ahmed, M. (1982) *Phytochemistry* **21**, 1679.
8. Payne, T. G. and Jefferies, P. R. (1973) *Tetrahedron* **29**, 2575.
9. Rodríguez, J., Tello, H., Quijano, L., Calderòn, J., Gómez, F., Romo, J. and Ríos, T. (1974) *Rev. Latinoamer. Quím.* **5**, 41.
10. Roitman, J. N. and James, L. F. (1985) *Phytochemistry* **24**, 835.
11. Markham, K. R., Chari, V. M. and Mabry, T. J. (1982) in *The Flavonoids: Advances in Research* (Harborne, J. B. and Mabry, T. J., eds), p. 23. Chapman & Hall, London.
12. Rajagopalan, S., Visweswara Rao, K. and Seshadri, T. R., F.A.Sc. (1947) *Proc. Indian Acad. Sci. A.* **26**, 18.
13. Herz, W., Anderson, G. D., Wagner, H., Maurer, G., Flores, G. and Farkas, L. (1973) *Tetrahedron Letters* 2571.
14. Lee, H. H. and Tan, C. H. (1967) *J. Chem. Soc. (C)* 1583.
15. Pérez, A. L. and Romo de Vivar, A. (1989) *Phytochemistry* **28**, 1639.
16. Sheldrick, G. M. (1983) An Integrated System for Solving, Refining and Displaying Crystal Structures from Diffraction Data (4th Revision). University of Göttingen.