Journal of Natural Products
Vol. 51, No. 3, pp. 614-616, May-Jun 1988

10-*TRANS*-CINNAMOYLMELITTOSIDE AND OTHER IRIDOIDS FROM *CASTILLEJA WIGHTII*[1]

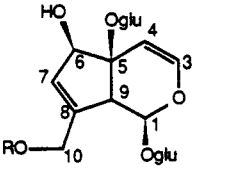
GILBERT N. BELOFSKY and FRANK R. STERMITZ*

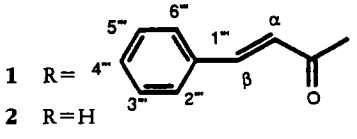
Department of Chemistry, Colorado State University, Fort Collins, Colorado 80523

We have been interested in the chemistry of *Castilleja* (Scrophulariaceae) species that are host to *Platyptilia* or *Amblyptilia* plume moths (1) or to *Euphydryas* checkerspot butterflies (2,3). A *Castilleja* native of the north central California coast, *Castilleja wightii* Elmer, is host to *Platyptilia pica* near Half Moon Bay in San Mateo County (4) and to *Euphydryas editha* in southwestern Mendocino County (M. Singer, personal communication, 1987). We report here on the iridoid glycoside content of a *C. wightii* population near Half Moon Bay.

The two major iridoids of *C. wightii* were found to be mussaenoside and 10-*trans*-cinnamoylmelittoside [**1**], a new iridoid whose structure was determined as described below. Bartsioside was identified as a major iridoid, and a small amount of the bitter phenylpropanoid glycoside verbascoside was also isolated.

The molecular formula of **1** was established as $C_{30}H_{38}O_{16}$ by high resolution mass spectrometry (hrfabms). Acetylation yielded a peracetate containing nine acetyl groups ($C_{48}H_{56}O_{25}$ by hrfabms).





The ^{1}H-nmr spectrum of **1** showed resonances typical for a *trans*-cinnamoyl group, and this was confirmed by the ^{13}C-nmr spectrum (Table 1). In the ^{13}C spectrum, 28 different carbons were seen, and, because two equivalent carbon pairs are present in the phenyl ring of the cinnamoyl moiety, all of the required 30 carbons were accounted for. Twelve carbon resonances (two groups of six closely matched pairs) were in the 62–100 ppm region of the ^{13}C-nmr spectrum. This fact suggested the presence of two sugar units as β-D-glucopyranoses, which was confirmed by the ^{13}C-nmr chemical shift values and two anomeric proton doublet resonances at 4.64 and 4.69 ppm in the ^{1}H-nmr spectrum. The chemical shifts of the ^{13}C resonances were typical of those for two independent glucopyranoses. Disaccharides linked at C-1, C-4, or C-6 show chemical shifts for these carbons which are 4–8 ppm removed from those observed for **1** (5,6). Two glucopyranose moieties would account for eight of the acetyl groups in the peracetate, so one additional OH must be present in **1**.

Nearly all ^{1}H- and ^{13}C-nmr resonances were quite close to those reported for melittoside [**2**] (7), with the exception of those expected to be changed upon placement of a *trans*-cinnamoyl group at C-10. The particular differences, besides those due to the cinnamoyl group proper, were the C-10 ^{1}H resonance at 4.87 ppm in **1**, in contrast to 4.30 ppm in **2**, and the C-10 ^{13}C resonance at 63.1 ppm for **1**, in comparison to 60.1 ppm in **2**. The structure was confirmed by INEPT and ^{1}H-^{13}C COSY nmr spectra, the data from which are included in Table 1.

[1]Paper 12 in the series "Chemistry of the Scrophulariaceae." For Paper 11, see Gardner and Stermitz (3).

TABLE 1. ^{13}C-^{1}H-Nmr COSY and ^{13}C-Nmr INEPT Spectral Data for 10-*trans*-Cinnamoylmelittoside [**1**] (Solvent CD_3OD, ppm).

^{13}C-nmr peak	INEPT (carbon type)	Correlation to ^{1}H-nmr peak	Carbon[a]
52.63	CH	3.3	C-9
62.43	CH_2	3.64–3.89	C-6″ }
63.00	CH_2	3.64–3.89	C-6′ }
63.14	CH_2	4.87	C-10
71.18	CH	|	C-4″ }
71.91	CH	|	C-4′ }
75.11	CH	3.27	C-2″ }
75.36	CH	to	C-2′ }
77.55	CH	3.43	C-3″ }
78.28	CH	|	C-3′ }
78.43	CH	|	C-5″ }
78.63	CH	|	C-5′ }
80.44	CH	4.43	C-6
80.51	C	—	C-5
94.88	CH	5.63	C-1
98.76	CH	4.64	C-1″ }
99.98	CH	4.69	C-1′ }
105.54	CH	5.15	C-4
118.72	CH	6.58	α
129.51	CH	7.62	C-3‴,C-5‴
130.24	CH	7.41	C-2‴,C-6‴
131.59	CH	5.91	C-7
131.81	CH	7.41	C-4‴
135.91	C	—	C-1‴
142.06	C	—	C-8
143.80	CH	6.42	C-3
147.08	CH	7.74	β
168.18	C	—	C=O

[a]Bracketed values are interchangeable.

EXPERIMENTAL

Plant material was collected at Bean Hollow, near Half Moon Bay, San Mateo County, California, by L.R. Heckard in 1985 and by J.R. Stermitz in 1987. It was identified as *C. wightii* by L.R. Heckard, Department of Botany, University of California at Berkeley, and a voucher specimen was deposited in the Jepson Herbarium at Berkeley. The Heckard collection was air dried and stored at room temperature for 1 year before analysis, while the Stermitz collection was analyzed 10 days after collection without special drying. Isolation results were essentially the same on both collections.

Dried and ground whole aboveground plants (Heckard collection), 16.5 g, were stirred in MeOH overnight, filtered, and the solution evaporated to yield a gummy solid which was dissolved in H_2O and extracted three times with Et_2O, three times with $CHCl_3$, and three times with *n*-BuOH. The *n*-BuOH layer was evaporated to dryness to yield 2.4 g of residue. Of this residue, 1.0 g was subjected to flash chromatography (Si gel; $CHCl_3$-MeOH, 70:30 increasing to 50:50). Combination of the first 11 fractions yielded 477 mg which was subjected to medium pressure liquid chromatography (mplc) (Si gel; CH_2Cl_2-EtOH, 80:20 increasing to 50:50). The first fraction yielded 35 mg of bartsioside and the second 122 mg of mussaenoside. Fractions 19–53 from the flash chromatography were combined to yield 356 mg of residue. This was subjected to mplc as above to yield 13 mg of verbascoside from the first fraction and 34 mg of **1** from the second fraction. Two subsequent fractions yielded 48 and 60 mg of **1**, contaminated with small amounts of unidentified compounds.

Bartsioside had ^{1}H- and ^{13}C-nmr spectra and tlc data identical with those of a previous isolate (8) and an authentic sample. Mussaenoside and verbascoside had tlc data and high field ^{1}H-nmr spectra identical with previous isolates (9).

10-*TRANS*-CINNAMOYLMELITTOSIDE [**1**].—Amorphous solid; $[\alpha]^{25}D$ −8.3 (c = 1.2, MeOH); hrfabms $[M+Na]^+$ 677.2025, calcd for $C_{30}H_{38}O_{16}Na$, 677.2046; nonaacetate de-

rivative hrfabms found: $[M+Li]^+$ 1039.3298, calcd for $C_{48}H_{56}O_{25}Li$, 1039.3271. Uv λ max (MeOH) 276 nm. Ftir (neat, NaCl) cm^{-1} 3338 vs, 2921 m, 1709 m, 1656 m, 1636 m, 1165 s, 1068 vs, 1014 vs, 947 m, 863 m, 769 m; ^1H nmr (CD_3OD, 500 MHz, Bruker 500) δ 3.27–3.43 (9H, m, H-9, H-2′,3′,4′,5′,2″,3″,4″,5″), 3.64–3.89 (4H, m, H-6′,6″), 4.43 (1H, br s, H-6), 4.64 (1H, d, 7.7, H-1″), 4.69 (1H, d, 7.7, H-1′), 4.87 (2H, H-10), 5.15 (1H, d, 6.4, H-4), 5.63 (1H, d, 4.4, H-1), 5.91 (1H, br s, H-7), 6.42 (1H, d, 6.4, H-3), 6.58 (1H, d, 16.1, α), 7.41 (3H, m, H-2‴,4‴,6‴), 7.62 (2H, m, H-3‴,5‴), 7.74 (1H, d, 16.1, β); ^{13}C nmr (CD_3OD, 67 MHz, Bruker 270) see Table 1; ^1H-^{13}C COSY nmr (CD_3OD, General Electric Q300) see Table 1.

ACKNOWLEDGMENTS

This work was supported by grant CHE-8521382 from the National Science Foundation. The 500 MHz ^1H-nmr spectrum was obtained at the Colorado State University Regional NMR Center funded by NSF Grant CHE-8208821. We are especially grateful to Dr. Dale R. Gardner and the Procter and Gamble Company for obtaining the ^1H-^{13}C-COSY nmr spectrum. We also thank L.R. Heckard for the original plant collection and identification and J.R. Stermitz for the second plant collection.

LITERATURE CITED

1. F.R. Stermitz, G.H. Harris, and W. Jing, *Biochem. Syst. Ecol.*, **14**, 499 (1986).
2. F.R. Stermitz, D.R. Gardner, F.J. Odendaal, and P.R. Ehrlich, *J. Chem. Ecol.*, **12**, 1459 (1986).
3. D.R. Gardner and F.R. Stermitz, *J. Chem. Ecol.*, **14** (in press).
4. H. Lange, *Hilgardia*, **19**, 561 (1950).
5. E. Breitmaier and W. Voelter, "^{13}C NMR Spectroscopy," 2nd ed., Verlag-Chemie, Weinheim New York, 1978.
6. G.N. Belofsky, M.S. Thesis, Colorado State University, Fort Collins, CO, 1987.
7. A. Bianco, P. Passacantilli, G. Righi, M. Nicoletti, and M. Serafini, *Phytochemistry*, **25**, 1981 (1986).
8. G. Harris, Ph.D. Dissertation, Colorado State University, Fort Collins, CO, 1987.
9. D.R. Gardner, J. Narum, D. Zook, and F.R. Stermitz, *J. Nat. Prod.*, **50**, 485 (1987).

Received 10 December 1987