



9 ξ -O- β -D-GLUCOPYRANOSYLOXY-5-MEGASTIGMEN-4-ONE FROM *LAMIUM ALBUM*

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Abstract—9 ξ -O- β -D-Glucopyranosyloxy-5-megastigmen-4-one, a new C_{13} glucoside, has been isolated from the leaves of *Lanium album*. The structure was elucidated on the basis of LSIMS and NMR spectral analysis, notably ^{13}C PENDANT, COSY 45, HMBC and HMQC. © 1997 Elsevier Science Ltd. All rights reserved

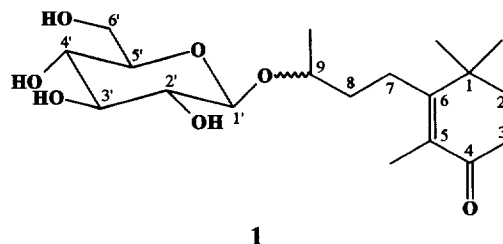
INTRODUCTION

Lanium album L., commonly known as 'white dead-nettle', is one of the *ca.* 40 species of annual or perennial herbs from the genus *Lanium* L. [1]. Previous phytochemical investigations have revealed the presence of a number of secondary metabolites of different classes from different parts of this plant: flavonoids, iridoid glucosides, terpenoids etc. [2–12]. We now wish to report on the isolation and characterisation of a new C_{13} glucoside from the leaves of this plant.

RESULTS AND DISCUSSION

Chromatography of a methanolic extract of the leaves of *L. album* yielded a glycoside which on the basis of extensive spectroscopic analysis, was identified as a novel natural product: 9 ξ -O- β -D-glucopyranosyloxy-5-megastigmen-4-one (1).

The LSIMS spectra of 1 revealed a prominent $[\text{M} + \text{H}]^+$ (positive ion mode) ion at m/z 373 and a $[\text{M} - \text{H}]^-$ (negative ion mode) ion at m/z 371 suggesting $M_r = 372$, solving for $\text{C}_{19}\text{H}_{32}\text{O}_7$. An UV absorption peak at 246 nm was in accordance with the presence of an α,β -unsaturated carbonyl chromophore. In addition to the signals (δ_{H} 3.16–4.35) resulting from the protons of a hexose moiety, the ^1H NMR spectrum (Table 1) revealed signals for four methylenes (δ_{H} 1.66, 1.84, 2.44, and 2.35/2.54), four methyls including a *gem*-dimethyl group (δ_{H} 1.76, 1.23 and 1.19), and an oxymethine (δ_{H} 3.97), all of which constituted the



aglycone part of the molecule. A ^1H doublet ($J = 7.8$ Hz) at δ 4.35 (δ_{C} 100.9) (Table 1) identified the hexose anomeric proton, from which an ^1H – ^1H COSY 45 spectrum enabled identification of H-2 to H-6 of the monosaccharide and confirmed it as β -D-glucose. The assignments of all other protons, especially the protons on the C_4 side-chain were also confirmed from ^1H – ^1H COSY 45 correlations. While the deshielded methyl at δ_{H} 1.76 indicated its attachment to an olefinic double bond, the absence of any signals for olefinic protons suggested that the two contributing carbons (C-5 and C-6) for this double bond were quaternaries (δ_{C} 130.3 and 167.5). The extent of deshielding on C-6 (δ_{C} 167.5) provided strong evidence for the presence of a β -carbonyl group. The attachment of the glucose moiety at C-9 was substantiated from the fact that the chemical shifts for H-9 and C-9, in the ^1H and ^{13}C NMR spectra, were more deshielded compared to that of an oxymethine in such a side-chain [13], and this was further evident from the presence of only one oxymethine (δ_{H} 3.97) in the aglycone moiety. In the HMBC spectrum, among the key correlations, the *gem*-dimethyl (δ_{H} 1.19; δ_{C} 25.8) showed 3J couplings, mutually to the carbons from each other, to C-6 (δ_{C} 167.5) and to C-2 (δ_{C} 36.8); 5-Me (δ_{H} 1.76)

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Table 1. ^1H (400 MHz), NMR and ^{13}C PENDANT NMR (100 MHz) data of **1**

C	$\delta^1\text{H}$ NMR	$\delta^{13}\text{C}$ NMR	Predicted $\delta^{13}\text{C}$ § NMR
1	—	†	36.3
2	1.84 <i>t</i> (6.8)	36.8	37.4
3	2.44 <i>t</i> (6.6)	34.0	34.2
4	—	†	198.6
5	—	130.3*	132.2
6	—	167.5*	163.2
7	2.54 <i>dt</i> (6, 12) 2.35 <i>dt</i> (6, 12)	25.9	25.3
8	1.66 <i>m</i>	36.0	33.3
9	3.97 <i>q</i> (6.2)	74.4	70.3
1'	4.35 <i>d</i> (7.8)	100.9	103.4
2'	3.16 <i>dd</i> (7.8, 9.0)	74.0	74.1
3'	3.36‡	76.8	76.9
4'	3.34‡	70.5	71.0
5'	3.26‡	76.5	77.3
6'	3.86 <i>dd</i> (11.8, 1.0) 3.68 <i>dd</i> (11.8, 6.1)	61.6	62.3
5-Me	1.76 <i>s</i>	†	11.1
1-Me ₂	1.19 <i>s</i>	25.8	26.8
9-Me	1.23 <i>d</i> (6.2)	18.4	21.8

Coupling constants (*J* in Hz) in parentheses.

* Obtained from HMBC correlation.

† Neither observed in the ^{13}C PENDANT, nor could be obtained from HMBC.

‡ Obtained from COSY 45.

§ Obtained through Specinfo Chemical Database Service, Daresbury, U.K.

showed 3J correlations to C-6 and 2J to C-5 (δ_{C} 130.3); 8-CH₂ (δ_{H} 1.66) displayed 3J correlation to C-6; 9-Me (δ_{H} 1.23) showed 3J connectivity to C-8 (δ_{C} 36.0) and 2J to C-9 (δ_{C} 74.4). Owing to the paucity of the sample a good ^{13}C PENDANT [14] spectrum could not be obtained, but with a combination of HMQC and HMBC experiments it was possible to conclude an unambiguous assignment of the structure of this compound as **1**. The ^{13}C data of **1** are generally in good agreement with the hypothetical spectral data generated from the Specinfo Chemical Database for this molecule (Table 1). However, there are some differences between the observed and predicted chemical shifts. The disparities between the observed and predicted signals around C-9 are probably a consequence of the prediction program not being able to take into account the effects of glucosylation at C-9 and the absolute configuration at C-9. Compound **1** belongs to the 'megastigmen' class of compounds which have been reported from a number of genera, notably, *Epimedium* [13], *Cydonia* [15], *Vinca* [16], *Pluchea* [17], *Podocarpus* [18], etc., of different plant families.

EXPERIMENTAL

UV: MeOH; NMR: CD₃OD on a Bruker AVANCE DRX400 instrument using standard Bruker microprograms. The chemical shifts are expressed in ppm;

LSIMS (+ve and -ve ion modes); glycerol matrix using a Cs⁺ primary ion beam on a VG Quattro triple-quadrupole mass spectrometer (VG Biotech, Altrincham, U.K.); EIMS: on the same instrument; CC: Silica gel 60G; HPLC: Gilson model 806 HPLC coupled with Gilson UV-Visible detector. RP and NP stand for reversed-phase prep. C₈ column and semi-prep. Apex II diol column. RP-seps were monitored at 242 nm.

Plant material. Leaves of *Lamium album* were collected locally and identified by James Kingdon, Dept Biological Sciences, University of Exeter.

Extraction. Freeze-dried leaves (22.6 g) were extracted with MeOH (350 ml) using a Soxhlet apparatus for 12 hr. The resulting extract was made to 70% aq. methanolic extract and defatted with *n*-hexane. The extract was then concd using a rotary evaporator at a maximum temp. of 45°.

Isolation of compounds. The concd extract was subjected to silica gel CC, eluted with mixts of CHCl₃-MeOH of increasing polarity. Frs eluted with 30% MeOH-CHCl₃ were combined together and sepd by RP-HPLC using an isocratic elution system (50% MeOH in H₂O, 5 ml min⁻¹). Forty frs of 5 ml each were collected. Compound **1** was purified from the combined RP-HPLC frs 20-35 by NP-HPLC (isocratic elution with 6% MeOH in CH₂Cl₂, 2 ml min⁻¹, *R_f* 12.8 min).

9ξ-O-β-D-Glucopyranosyloxy-5-megastigmen-4-one (**1**). (0.3 mg). Gum. UV λ_{max} nm (log ϵ): 246 (4.02); ^1H NMR: (Table 1); ^{13}C NMR: Table 1; LSIMS: *m/z* 373 [M+H]⁺ (positive ion mode) and 371 [M-H]⁻ (negative ion mode); EIMS *m/z* (rel. int.): 193 [M-179]⁺ (90), 177 (40), 165 (100), 152 (95), 138 (90), 122 (88), 110 (92), 98 (92), 82 (90), 74 (95), 70 (92), 61 (90), 46 (85), 44 (96).

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