Pergamon

0031-9422(94)00737-3

Phytochemistry, Vol. 38, No. 3, pp. 699–701, 1995
Elsevier Science Ltd
Printed in Great Britain
0031-9422/95 $9.50 + 0.00

(20*R*)-*O*-(3)-α-L-ARABINOPYRANOSYL-PREGN-5-EN-3β,20-DIOL FROM *BRUCEA JAVANICA*

CHRISTINE KAMPERDICK, TRAN VAN SUNG,* TRINH THI THUY,* MAI VAN TRI* and GÜNTER ADAM†

Institute for Plant Biochemistry, P.O.B. 250, D-06018 Halle/Saale, Germany; *Institute of Chemistry, National Centre for Scientific Research of Vietnam, Nghia Do, Tu Liem, Hanoi, Vietnam

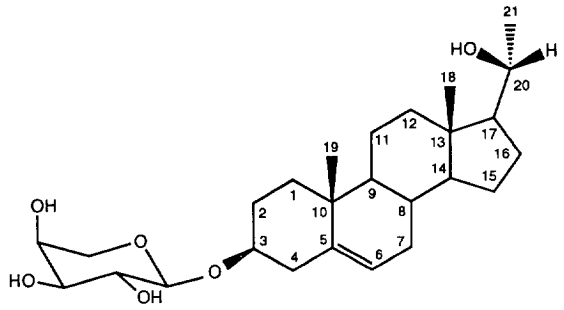
(*Received* 4 *May* 1994)

Key Word Index—*Brucea javanica*; Simaroubaceae; steroid glycoside; (20*R*)-*O*-(3)-α-L-arabinopyranosyl-pregn-5-en-3β,20-diol.

Abstract—The new glycoside, (20*R*)-*O*-(3)-α-L-arabinopyranosyl-pregn-5-en-3β,20-diol, was isolated from leaves of *Brucea javanica* Merr. The structure was determined by ^{1}H and ^{13}C NMR spectroscopy.

INTRODUCTION

Brucea javanica Merr. grows in the tropical areas of Asia, Indonesia and Australia. All parts of the plant taste bitter, especially the seeds [1]. Seeds and roots of this plant contain predominantly quassinoids [2], while the components of the leaves have not yet been studied. The leaves are used in folk medicine for poultices on boils, ringworm, scurf, centipede bites and enlarged spleens [1]. In this report we describe the isolation and structure elucidation of a new pregnane glycoside, (20*R*)-*O*-(3)-α-L-arabinopyranosyl-pregn-5-en-3β,20-diol (**1**), from the chloroform extract of the leaves of *Brucea javanica*.



1

RESULTS AND DISCUSSION

Compound **1** was isolated from the chloroform extract of the dried leaves of *Brucea javanica* Merr. by chromatography on silica gel. The IR spectrum exhibited absorption at 3400 cm^{-1} (OH). The FAB-mass spectrum of **1** showed the $[M + H]^+$ peak at *m/z* 451. The base peak at *m/z* 301 was from the loss of the arabinosyl residue and H_2O. Its HR-EI-mass spectrum, which contained no molecular ion peak, showed the important peaks at *m/z* 318.25766 ($C_{21}H_{34}O_2$, [aglycone]$^+$) and 300.24411 (base peak, $C_{21}H_{32}O$, [aglycone $- H_2O$]$^+$).

The ^{1}H NMR spectrum of **1** (Table 1) contained six arabinosyl signals and two methyl singlets at δ0.72 and 0.93 and one methyl doublet at δ1.42 (J = 6.6 Hz), suggesting a pregnane skeleton. This was supported by the ^{13}C NMR spectrum (three CH_3, eight CH_2, seven CH and three C for the aglycone, see Table 1). The chemical shift of the two olefinic carbons (a quaternary carbon at δ141.1 and a tertiary one at δ121.9) were in good agreement with the recently published data for pregn-5-en-3-O-glycosides [3]. The proton signal at δ3.89 showed the typical multiplicity of H-3 of 3β-hydroxysteroids (*tt*, J = 11.2 and 4.5 Hz). The downfield shift of the corresponding carbon to δ78.1 indicated the glycosylation of the 3β-hydroxy group with the arabinose molecule. A cross peak in the ^{1}H–^{1}H-COSY between the methyl doublet at δ1.42 and the proton signal at δ3.92 confirmed, that the second oxygenated carbon of the aglyone (δ_C69.0/δ_H3.92) should be located in the side chain at position C-20. The carbon signals of the steroidal skeleton were assigned by comparison with those of androstane by consideration of the substituent effects [4]. The assignment of the signals of the attached protons was obtained by the analysis of the ^{13}C–^{1}H-COSY. Considering the different substitution at C-3, their chemical shifts were consistent with those of 3β-acetoxy-pregn-5-en-20β-ol [5]. The interpretation of the ROESY-diagram confirmed the all-*trans* configuration of the steroidal skeleton and the location of the arabinosyl residue at C-3 (cross-peaks of H-8/H_3-18, H-8/H_3-19, H_{ax}-4/H_3-19, H-1′/H-3 and H-1′/H_{eq}-4; correlation between H_3-18/H_3-19 hidden under the T_1 noise). The configuration of **1** at C-20 was determined by comparison with the methyl proton shifts of the 20-epimers of pregn-5-en-3β,20-diol [6], whereas a

†Author to whom correspondence should be addressed.

Table 1. NMR spectral data of compound **1** (500/125 MHz in pyridine-d_5)

	δ_C (ppm)	CH_n*	δ_H (ppm), J (Hz)
1	37.7	CH_2	1.06/1.76
2	30.5	CH_2	1.75/2.14
3	78.1	CH	3.89 *tt* (11.2; 4.5)
4	39.4	CH_2	2.42 *tm*† (12.0)/2.67 *ddd* (13.3; 4.7; 2.0)
5	141.1	C	—
6	121.9	CH	5.35 *dt* (5.3; 2.5)
7	32.3	CH_2	1.51/1.89
8	31.9	CH	1.38
9	50.6	CH	0.90
10	37.1	C	—
11	21.2	CH_2	1.42
12	39.2	CH_2	1.08/1.88
13	41.7	C	—
14	56.9	CH	0.95
15	24.6	CH_2	1.12/1.61
16	26.7	CH_2	1.90/2.19
17	59.4	CH	1.51
18	12.7	CH_3	0.72 *s*
19	19.5	CH_3	0.93 *s*
20	69.0	CH	3.92
21	24.9	CH_3	1.42 *d* (6.6)
1′	103.2	CH	4.84 *d* (6.7)
2′	72.6	CH	4.43 *t*† (8.0)
3′	74.7	CH	4.19 *dd* (9.0; 3.3)
4′	69.6	CH	4.32‡
5′	67.0	CH_2	3.79 *dd*‡ (13.0; 2.0)/4.31 *dd*‡ (13.0; 2.0)

The proton shifts of overlapping signals were determined from the ^{13}C–^1H-COSY.
*From DEPT spectrum.
†The expected *dd* was not resolved and appeared as *t* with an averaged coupling constant.
‡Muliplet of higher order.

good correspondence was found for the values of the (20*R*)-configuration.

The signals of the sugar residue were assigned by the ^1H–^1H-COSY analysis. The coupling constants $J_{1',2'}$ (6.7 Hz) and $J_{2',3'}$ (9.0 Hz) indicated an interplanar angle of nearly 180° and equatorial hydroxyl groups, while the smaller value of $J_{3',4'}$ (3.3 Hz) belonged to a smaller angle (30–60°) and an axial hydroxyl group at C-4′. This characterized an α-arabinopyranosyl residue. The corresponding ^{13}C shifts were consistent with reported data for methyl-α-L-arabinopyranoside [7]. From these data the structure of the new glycoside was determined as (20*R*)-*O*-(3)-α-L-arabinopyranosyl-pregn-5-en-3β,20-diol (**1**).

EXPERIMENTAL

General. Mps: uncorr. EI-MS: AMD 402, 70 eV; FAB-MS: AMD 402, glycerine as matrix. NMR spectra: 500/100 MHz (Varian Unity 500).

Plant material. Brucea javanica (L.) Merr. was collected from Quang Nam-Da Nang Province in March 1991 and identified by Dr. Tran Dinh Dai. A voucher specimen is deposited at the Herbarium of the Institute of Ecology and Natural Resources of Vietnam, Nghia Do, Tu Liem, Hanoi.

Isolation of steroid. Dried ground leaves of *B. javanica* (800 g) were defatted with *n*-hexane at room temp. for 8 hr, followed by successive extraction with $CHCl_3$ and EtOH (each 1500 ml, 8 hr) in a Soxhlet apparatus. The residue of the $CHCl_3$ extract (31 g) was chromatographed over silica gel (Merck G 60) eluting with $CHCl_3$–MeOH mixtures. Evaporation of the $CHCl_3$–MeOH (9:1) fractions and recrystallization from MeOH yielded 33 mg of **1** as white needles. Mp 258–260°. $[\alpha]_{578}^{25}$ − 21° (MeOH; *c* 0.48). IR ν_{max}^{KBr} cm^{-1}: 3400 (br), 2933, 1457, 1378, 1088, 1000. FAB-MS *m/z* (rel. int.): 543 $[M + H + glycerine]^+$ (9), 451 $[M + H]^+$ (4), 301 $[M + H - C_5H_9O_4 - OH]^+$ (100), 283 $[M + H - C_5H_9O_4 - OH - H_2O]^+$ (54). EI-MS *m/z* (rel. int.): 318.25766 $C_{21}H_{34}O_2$ calc. 318.25784 $[aglycone]^+$ (2.4), 300.24411 $C_{21}H_{32}O$ calc. 300.24399 $[aglycone - H_2O]^+$ (100), 283.24591 $C_{21}H_{31}$ calc. 283.24625 $[aglycone - H_2O - OH]^+$ (62), 267.21301 $C_{20}H_{27}$ calc. 267.21318 (13), 257.22714 $C_{19}H_{29}$ calc. 257.22716 (13), 161.13330 $C_{12}H_{17}$ calc. 161.13333 (23), 147.11596 $C_{11}H_{15}$ calc. 147.11582 (18), 135.11689 $C_{10}H_{15}$ calc. 135.11684 (15). Further elution with $CHCl_3$–MeOH (17:3) afforded a mixture of phytosterol glycosides (main component stigmasterol glucoside, minor components sitosterol and campesterol glucoside), determined by MS and ^{13}C NMR spectroscopy.

Acknowledgements—We thank Dr J. Schmidt (Institute of Plant Biochemistry, Halle/S., Germany) for the mass spectra and the Ministry of Research and Technology, Bonn, Germany, for financial support. Two of us (M.V.T. and T.T.T.) are indebted to the International Foundation for Science (I.F.S.), Stockholm, Sweden, for financial support.

REFERENCES

1. Perry, L. M. (1989) *Medicinal Plants of East and Southeast Asia*, p. 389. The MIT Press, Cambridge.
2. Hegenauer, R. and Hegenauer, M. (1990) *Chemotaxonomie der Pflanzen* Bd. IX, p. 561. Birkhäuser, Basel.
3. Srivastar, S., Deepak, D. and Khare, A. (1994) *Tetrahedron* **50**, 789.
4. Blunt, J. W. and Stothers, J. B. (1977) *Org. Magn. Reson.* **9**, 439.
5. Kirk, D. N., Toms, H. C., Douglas, C. and White, K. A. (1990) *J. Chem. Soc. Perkin Trans. II* 1567.
6. Cooley, G., Kirk, D. N., Morgan, R. E. and Sáe Melo, M. L. (1977) *J. Chem. Soc. Perkin Trans. I* 1390.
7. Seo, S., Tomita, Y., Tori, K. and Yoshimura, Y. (1978) *J. Am. Chem. Soc.* **100**, 3331.