PERGAMON

Phytochemistry 50 (1999) 463–465

PHYTOCHEMISTRY

Triterpenes from *Rubus cochinchinensis*

Trinh Phuong Lien [b], Christine Kamperdick [a], Tran Van Sung [b], Günter Adam [a,*]

[a] *Institute of Plant Biochemistry, Weinberg 3, D-06120 Halle/Saale, Germany*
[b] *Institute of Chemistry, National Centre for Natural Science and Technology of Vietnam, Nghia Do, Tu Liem, Hanoi, Vietnam*

Revised 4 August 1998

Abstract

The investigation of leaves of *Rubus cochinchinensis* (Rosaceae) afforded five triterpenes including the new compound 2-O-acetylsuavissimoside F1. © 1998 Elsevier Science Ltd. All rights reserved.

Keywords: *Rubus cochinchinensis*; Rosaceae; Triterpenes; Suavissimoside F1; 2-O-acetylsuavissimoside F1

1. Introduction

Rubus cochinchinensis Tratt. is an endemic Vietnamese medicinal plant, the decoction of which is used to treat constipation and hepatitis (Do Tat Loi, 1991). An infusion of the dried leaves is utilized as an aperient at child birth (Perry, 1989). No phytochemical studies have been available until now. In continuation of our search for new bioactive compounds from Vietnamese medical plants (Kamperdick, Phuong, Sung & Adam, 1998), we now report on the constituents of this species.

2. Results and discussion

The EtOAc and the *n*-BuOH extract of the leaves afforded besides epicatechin five triterpenes of ursolic acid type: ursolic acid, 2-oxo-pomolic acid (**1**), which is a rare triterpene isolated until now only from infected *Malus pumila* (Kemp, Holloway & Burden, 1985) and from *Sanguisorba alpina* (Jia, Liu & Liu, 1993), tormentic acid (**2**), suavissimoside F1 (**3**) and the new compound **4**. Like **3**, which is known from *Rubus suavissimus* (Gao, Chen, Tanaka, Kasai, Seto & Tanaka, 1985), **4** is a glucoside. Its molecular weight of 722 obtained from the ESI MS comprises 42 mass units more than **3** suggesting **4** as an acetyl derivative of **3**. This was confirmed in the NMR spectra by an 3H singlet at δ_H 2.07 and two additional carbon signals at δ_C 170.8 and 21.3. Comparison of the carbon shifts of **3** and **4** shows differences only in ring A, which thus must carry the acetyl group in position 2 or 3. Acetylation of a secondary hydroxyl group generally causes a deshielding of the α-carbon of 3–4 ppm and a shielding of the β-carbons of 2–3 ppm (Breitmaier & Voelter, 1990). This information locates the acetyl group in position 2, resulting in a shift of +4.8 ppm for the α-carbon C-2 and of −3.4 and −3.8 ppm for the β-carbons C-1 and C-3, respectively, compared with **3**. The new compound **4** thus can be regarded as 2-O-acetylsuavissomoside F1.

3. Experimental

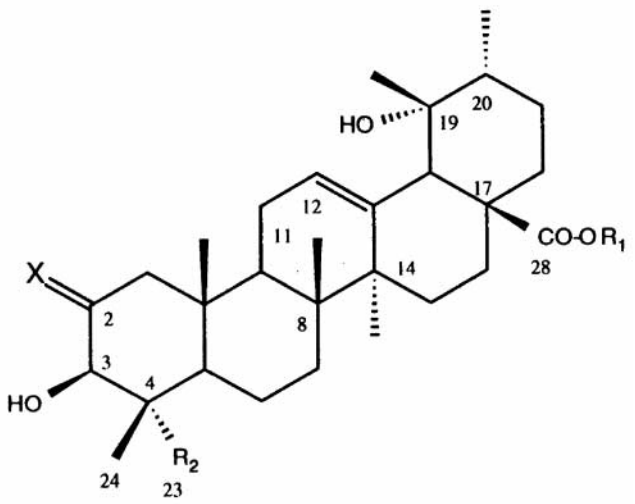
EIMS: AMD 402, 70 eV. ESI MS: Finnigan TSQ 700. NMR: Varian Gemini 300, Unity 500. CC: silica gel 60, 40–63 nm (Merck).

3.1. Plant material

Leaves of *Rubus cochinchinensis* Tratt. were collected in Cuc Phuong, Province Ninh Binh, North Vietnam, in March 1996 and identified by Dr Tran Dinh Dai. A voucher specimen is deposited at the Institute of Ecology of the National Centre for Natural Science and Technology, Hanoi, Vietnam.

* Corresponding author. Tel.: +0049-345-5582-216; Fax: +0049-345-5582-102. E-mail: gadam@ibp.uni-halle.de.

0031-9422/98 $ - see front matter
PII: S0031-9422(98)00598-6



	X	R_1	R_2
1	O	H	CH_3
2	α-OH,H	H	CH_3
3	α-OH,H	glc	COOH
4	α-OAc,H	glc	COOH

3.2. Extraction and isolation.

Dried and powdered leaves (1.5 kg) were extracted 3× with 95% aq. MeOH at room temp. The aq. solution, which remained after evaporation of the organic solvent under red. pres., was extracted successively with *n*-hexane, EtOAc and *n*-BuOH (each 3×), to yield 9 g *n*-hexane extract, 80 g EtOAc extract and 100 g *n*-BuOH extract. 40 g of the EtOAc extract were fractionated on silica gel with solvents of increasing polarity (0–30% MeOH in $CHCl_3$ followed by $CHCl_3$–MeOH–H_2O, 65:35:5, 13 frs). Frs 2–3 (5 g, eluted with *n*-hexane–$CHCl_3$, 3:7) were separated into ten further frs on silica gel with *n*-hexane–acetone (6:4). Fr 5 yielded 20 mg of ursolic acid (CC with *n*-hexane–acetone 8:2) and 30 mg of **1** (CC with *n*-hexane–acetone, 7:3). Fr 8 (2 g, eluted with 100% $CHCl_3$) afforded 200 mg of epicatechin after CC on silica gel using $CHCl_3$–MeOH–H_2O (98:16:1). 35 g of the *n*-BuOH extract were separated on silica gel using $CHCl_3$–MeOH–H_2O (65:35:5, 10 frs). Frs 2–3 (1.5 g) were fractionated on silica gel using $CHCl_3$–MeOH (95:5) followed by reversed phase chromatography on RP8 with MeOH–H_2O (85:15) giving 30 mg of **2**. Fr 6 (1.6 g) afforded 20 mg of **4** after chromatography on silica gel using EtOAc–MeOH–H_2O (25:2:1). Purification of frs 7–8 (3 g) over silica gel using EtOAc–MeOH–H_2O (50:8:5) yielded 300 mg of **3**.

*3.3. 2-Oxo-pomolic acid (**1**)*

Amorphous. $[\alpha]_D^{23}$ +67.1° (MeOH, *c* 0.20). ^{13}C NMR data correspond to those of the β-D-glucopyranosyl ester (Jia et al., 1993) apart from the missing sugar moiety.

*3.4. Suavissimoside F1 (**3**)*

Crystals from $CHCl_3$/MeOH), mp 253–256°. $[\alpha]_D^{25}$ +15.6° (MeOH, *c* 0.22). NMR data are in agreement with ref. Gao et al., 1985, except δ_C of C-17 (ref. Gao et al., 1985: 42.4, compound **3**: 48.2), which is supposed to result from a misprint.

*3.5. 2-O-Acetylsuavissimoside F1 (**4**)*

Amorphous. $[\alpha]_D^{28}$ −11.9° (MeOH, *c* 0.25). Positive ESI MS *m/z* (rel. int.): 745 (100) $[M + Na]^+$. EIMS *m/z* (rel. int.): 560 [aglycone]$^+$ (8), 542 (12), 514 (100), 498 (25), 454 (14), 442 (70), 246 (16), 233 (29), 218 (16), 201 (27), 187 (24), 146 (61), 119 (22). IR_{max}^{KBr} (cm^{-1}): 3431 (br), 2969, 2933, 2879, 1714, 1691, 1458, 1393, 1154, 1117, 1057, 1033, 933. ^1H NMR (pyridine-d_5, 300 MHz): δ 1.08 (3H, *d*, *J* = 6.3 Hz, H_3-30), 1.19 (6H, *s*, 2 Me), 1.42, 1.66 and 1.73 (each 3H, *s*, Me), 2.07 (3H, *s*, Ac), 4.09 (1H, *m*, glc), 4.23 (1H, *dd*, *J* = 8.4 and 8.4 Hz, glc), 4.31–4.53 (4H, overlapping, glc), 4.78 (1H, *d*, *J* = 10.2 Hz, H-3), 5.25 (<1H, *s*, OH), 5.55 (1H, *br s*, H-12), 5.61 (1H, *ddd*, *J* = 10.6, 10.6 and 4.0 Hz, H-2), 6.32 (1H, *d*, *J* = 8.0 Hz, H-1′). ^{13}C NMR (pyridine-d_5, 125 MHz): δ 44.8 (C-1), 73.3 (C-2), 77.1 (C-3), 55.0 (C-4), 52.0 (C-5), 21.2 (C-6), 33.1 (C-7), 40.6 (C-8), 47.9 (C-9), 38.5 (C-10), 24.1 (C-11), 127.9 (C-12), 139.3 (C-13), 42.08 (C-14), 29.1 (C-15), 26.0[a] (C-16), 48.5 (C-17), 54.4 (C-18), 72.6 (C-19), 42.06 (C-20), 26.6[a] (C-21), 37.6 (C-22), 13.3 (C-24), 17.1[b] (C-25), 17.2[b] (C-26), 24.5 (C-27), 176.9 (C-28), 26.8 (C-29), 16.7[b] (C-30), 170.8 (CH_3-CO), 21.3 (CH_3-CO), 95.8 (C-1′), 74.0 (C-2′), 79.3[c] (C-3′), 71.1 (C-4′), 78.9[c] (C-5′), 62.2 (C-6′); [a,b,c] assignments interchangeable, C-23 not detected.

Acknowledgements

We thank the Bundesministerium für Bildung, Wissenschaft, Forschung und Technologie, Bonn, for financial support, Dr J. Schmidt, Halle, for mass spec-

tra, as well as Dr T. D. Dai, Hanoi, for the identification of the plant material. One of us (T. P. L.) is indebted to the DAAD, for a grant.

References

Breitmaier, E., & Voelter, W. (1990). *Carbon-13 NMR spectroscopy* (p. 337). Weinheim: VCH.

Do Tat Loi (1991). *Nhung cay thuoc va vi thuoc Viet Nam* (Glossary of Vietnamese Medicinal plants) (p. 450). Hanoi, Vietnam: Nha xuat ban Khoa hoc va Ky thuat (Science and Technics Publication).

Gao, F., Chen, F.-H., Tanaka, T., Kasai, R., Seto, T., & Tanaka, O. (1985). *Chemical Pharmaceutical Bulletin, 33,* 37.

Jia, Z.-J., Liu, X.-Q., & Liu, Z.-M. (1993). *Phytochemistry, 32,* 155.

Kamperdick, C., Phuong, N. M., Sung, T. V., & Adam, G. (1998). *Phytochemistry, 48,* 577.

Kemp, M. S., Holloway, P. J., & Burden, R. S. (1985). *Journal of Chemical Research Miniprint, 00,* 1848.

Perry, L. M. (1989). *Medicinal plants of East and Southeast Asia* (p. 346). Cambridge: MIT Press.