248. 2-[6′-(*O-trans*-Cinnamoyl)-β-D-glucopyranosyloxy)]-3-methyl-4*H*-pyran-4-one, a New Acylated Pyrone Glucoside from *Silene vulgaris (Caryophyllaceae)*

Part 186. Papers on Organic Natural Products[1])

by **Robert P. Borris**[a])[2]), **Karlheinz Seifert**[b]), **Siegfried Johne**[b]) und **Manfred Hesse**[a])

[a]) Organisch-chemisches Institut der Universität Zürich, Winterthurerstrasse 190, CH-8057 Zürich

[b]) Institut für Biochemie der Pflanzen, Akademie der Wissenschaften der DDR, Weinberg 3, DDR-4010 Halle/Saale

(27.IX.82)

Summary

A previously unreported acylated 4-pyrone glycoside, 2-[6′-*O-trans*-cinnamoyl)-β-D-glucopyranosyloxy]-3-methyl-4*H*-pyran-4-one (**1**) has been isolated from the basic fraction of a chloroform/methanol extract of the shoots of *Silene vulgaris* (Moench) Garcke *(Caryophyllaceae)* and identified mainly on the basis of the spectral characteristics of this compound and its triacetate derivative as well as chemical degradation.

Several researchers have examined species of the genus *Silene*. In the course of these studies, a number of flavonoids and flavonoid glycosides [3], saponins [4], triterpene glycosides [5], 'cardiac glycosides' [6], anthocyanins [7], lipids [8], oligosaccharides [9], and phytoecdysones [10] have been detected or isolated and identified.

Although it has been reported that alkaloids have been detected in various species of the genus *Silene* [2], to date, no alkaloids have been isolated from this genus. However, the basic fraction of a chloroform/methanol extract of the shoots of the bladder campion, *Silene vulgaris* (Moench) Garcke *(Caryophyllaceae)*, gives intense positive reactions with several alkaloid-detecting reagents (*Mayer*'s, *Wagner*'s, *Dragendorf*'s, and potassium iodoplatinate), and was for this reason selected for examination.

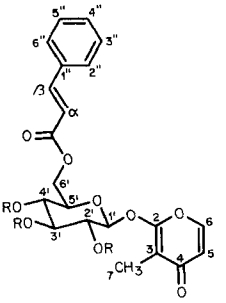
Chromatographic fractionation of this material, initially on silica gel, and then on controlled porosity glass powder afforded the main reactive component **1** as colorless crystals.

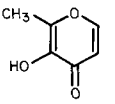
[1]) Part 185, see [1].

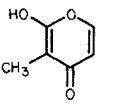
[2]) 1981-1982 Postdoctoral fellow, Organisch-chemisches Institut der Universität Zürich. Present address: Department of Pharmacognosy, College of Pharmacy, Rutgers, The State University of New Jersey, P.O. Box 789, Piscataway, New Jersey 08854, U.S.A.

0018-019X/82/8/2481-05$01.00/0

Electron impact mass spectrometry suggested a molecular weight of 293. This value was rejected as being too low, in light of information provided by the IR., ^{1}H-NMR. and ^{13}C-NMR. spectra. Field desorption mass spectrometry revealed a molecular ion at *m/z* 418. Microanalysis indicated the absence of nitrogen, and







1 R=H
2 R=COCH$_3$

3 **4**

in conjunction with the molecular weight suggested a molecular formula of $C_{21}H_{22}O_9$. The base peak at *m/z* 131 in the electron impact mass spectrum of the isolate, along with less abundant fragments at *m/z* 103 and 77 suggested the presence of a cinnamate ester in the molecule. This contention was supported by a strong IR. absorption at 1712 cm^{-1}, characteristic of an α,β-unsaturated ester, and by ^{13}C-NMR. resonances ($CDCl_3$) at 166.6 (*s*), 144.8 (*d*), 134.2 (*s*), 130.1 (*d*), 128.7 (*d*), 128.0 (*d*), and 116.6 (*d*) ppm. A five-proton *m* between 7.7–7.2 and two one-proton doublets at 6.50 and 7.67 ppm in the ^{1}H-NMR. spectrum confirmed the presence of this moiety. The large coupling constant (16 Hz) between the side-chain olefinic proton signals indicates that these protons are *trans*. Examination of the ^{13}C-NMR. spectra of **1** and its triacetate derivative **2**, prepared by treatment of **1** with acetic anhydride/pyridine, in the region of 60–105 ppm suggested the presence of a hexopyranoside in this isolate (see *Table*). The chemical shift of the anomeric C-atom (in **1** at 104.4 ppm) requires that the sugar is involved in a β-glycosidic linkage, and that the cinnamate ester is not located at C(1′) or C(2′) of the sugar. Comparison of the ^{1}H-NMR. spectra of **1** and **2** shows that on acetylation, the 2 H–C(6′) do not shift downfield, establishing C(6′) as the original site of acylation. Proton double-resonance experiments on the triacetate derivative **2** allowed definition of the sugar moiety. The H–C(5′) is observed as a $d \times t$ with a small coupling (4 Hz) to 2 H–C(6′) and a large coupling (10 Hz) to H–C(4′) indicating that both of these protons are axial. Irradiation of H–C(5′) causes the two proton *d* at 4.30 ppm (2 H–C(6′)) to collapse to a *s*, and the $d \times d$ at 5.20 ppm (H–C(4′)) to simplify to a *d* with a remaining coupling constant of about 9 Hz, indicating that H–C(3′) is also axial. As the anomeric proton is seen as a *d* with a large (8 Hz) coupling constant, H–C(2′) must also be axial. Therefore the sugar is glucose. The large negative optical rotation is consistent with the presence of a β-D-glucoside.

On the basis of ^{13}C-NMR. data, the remainder of the molecule, $C_6H_5O_2$, consists of two protonated sp^2 C-atoms, a carbonyl group, two fully substituted sp^2 C-atoms, an O-atom, and a methyl group not attached to the O-atom. In the ^{13}C-NMR. (SFORD, $CDCl_3$) spectrum of **1**, the *d* at 154.5 and 117.7 ppm indicate the presence of an α,β-unsaturated carbonyl group in which the C(β)-atom is attached to O-atom. This suggests that **1** is a 2,3-disubstituted pyran-4-one with the methyl group and the acylated glucoside as the two substituents. Comparison of the C-atom chemical shifts of the pyranone portion of **1** with those of maltol (**3**) [11] suggests that in **1**, the glycoside should be attached at C(2) and the methyl group at C(3). Methanolysis of **1** afforded, in addition to methyl cinnamate, the free pyranone aglycone which differed from **3** in the ^{13}C-NMR. chemical shifts of the two substituted ring C-atoms (see *Table*). Whereas C(2) in **3** resonates at 144.1 ppm, in the isolated aglycone this C-atom was found at 149.0, while C(3), at 142.8 ppm in maltol, appeared at 142.7 ppm in the hydrolysate. These differences are in agreement with structure **4** for the aglycone. Further the change in chemical shift of C(2) in going from **4** to **1** is about 12 ppm, consistent in magnitude with the shifts usually seen in going from a free hydroxyl to a glycoside [12]. If the aglycone were maltol, the observed change in chemical shift of 18 ppm at C(3) would substantially exceed the 5–10 ppm shift usually encountered. Thus, all of the presented data are in accord with structure **1** for the isolate.

Table. *^{13}C-NMR. data of compounds* **1**–**4** *and ethyl cinnamate*

C-Atom	**1** $(CD_3)_2SO$	**1** $CDCl_3$	**2** $CDCl_3$	**3** $(CD_3)_2SO$	Ethyl Cinnamate $(CD_3)_2SO$	**4** $(CD_3)_2SO$
2	161.1	162.9	161.2	144.1		149.0
3	141.4	142.3	141.0	142.8		142.7
4	173.9	175.3	173.4	172.1		172.2
5	116.0	117.7[a])	117.1[a])	113.2		113.3
6	155.4	154.5	153.5	154.4		154.3
7	15.0	15.6	15.3	13.8		13.9
1′	103.3	104.4	99.2	from [11]		
2′	76.0	76.4	72.6			
3′	74.1[a])	74.5[b])	71.9[b])			
4′	69.9	69.9	68.7			
5′	73.8[a])	73.6[b])	71.4[b])			
6′	63.3	63.5	61.7			
1″	133.8	134.2	134.0		134.4	
2″,6″	128.2	128.0	128.0		126.2	
3″,5″	128.8	128.7	128.8		128.9	
4″	130.3	130.1	130.4		130.2	
β	144.4	144.8	145.4		144.3	
α	117.7	116.6[a])	117.1[a])		118.4	
C=O	165.8	166.6	166.0		166.2	
CH_3CO			20.8, 20.6, 20.6			
CH_3CO			169.9, 169.3, 169.3			
CH_3CH_2					14.4, 60.2	

[a])[b]) Assignments within each footnoted group may be interchanged.

The aglycone **4** has been previously detected as a constituent of tobacco smoke [13], but to our knowledge, no previous isolation from a non-pyrolyzed plant sample of compounds containing a pyran-4-one moiety with this substitution pattern has so far been reported.

This work was supported by the *Schweizerischer Nationalfonds zur Förderung der wissenschaftlichen Forschung* and the *Legerlotz-Stiftung* at Organisch-chemisches Institut der Universität Zürich, both are thankfully acknowledged.

Experimental Part

General remarks. Column chromatography was carried out on silica gel (*Merck*, 70–230 mesh) or on controlled porosity glass powder, *CPG-10*, 120 Å pore size, 200–400 mesh *(Fluka)*. Thin layer chromatography (TLC.) was performed on 0.2 mm layers of *silica gel 60F$_{254}$ (Merck)*, and visualized with potassium iodoplatinate reagent. Solvents used were analytical reagent grade, and distilled prior to use. Optical rotations were determined in MeOH using a *Perkin* model *241* polarimeter. IR. spectra were determined with a *Perkin-Elmer* model *297* spectrometer in KBr, and absorption bands in wavenumber (cm^{-1}). UV. spectra were recorded with a *Perkin-Elmer* model *555* instrument in MeOH, absorptions in nm ($\log\varepsilon$). ^{1}H-NMR. spectra were determined at 200 MHz with a *Varian XL-200* superconducting spectrometer, while ^{13}C-NMR. spectra were obtained at 25.2 MHz with a *Varian XL-100* instrument. Chemical shifts (δ) are reported in parts per million (ppm), and coupling constants, J, are reported in Hz. Signal multiplicities are abbreviated as: s = singlet, d = doublet, t = triplet, m = multiplet, and br. = broad. Field desorption (FD.) and electron impact (EI.) mass spectra (MS.) were obtained with a *Varian MAT 711* instrument coupled to a *Varian-SS-100MS* data system, important signals in *m/z* (rel.-%). Abbreviations: SFORD = single frequency off-resonance decoupled.

Plant Material. Seeds of *Silene vulgaris* (Moench) Garcke were obtained from the botanical gardens of Geneva, Switzerland, and cultivated in the gardens of the Institut für Biochemie der Pflanzen, Halle, during the 1981 growing season. Shoots from these plants were collected in August 1981.

Isolation. The powdered, air-dried shoots of *S. vulgaris* (574 g) were exhaustively extracted with $CHCl_3/CH_3OH$ 7:3 in a *Soxhlet* apparatus. After removal of solvent under reduced pressure, the residue was suspended in 10% hydrochloric acid (1 liter) and extracted with an equal volume of ether. The acidic aqueous phase was then basified (K_2CO_3) and exhaustively extracted with $CHCl_3/CH_3OH$ 4:1. After removal of solvent under reduced pressure, the residue was dissolved in 10% hydrochloric acid (500 ml) and again partitioned against diethyl ether. Basification of the aqueous fraction followed by exhaustive extraction with $CHCl_3/CH_3OH$ 4:1 and evaporation of solvent under reduced pressure afforded 4.24 g of a crude mixture of plant 'bases'.

Chromatography of the crude basic fraction on silica gel (300 g) eluted with $CHCl_3/CH_3OH$ 19:1 followed by repetitive chromatography on glass powder (125 ml column volume) eluted with $CHCl_3$ afforded a white powder which was then crystallized from CH_3OH to yield 1.72 g (0.30% of dried plant) of colorless plates of **1**, m.p. 163.0–163.7°. $[\alpha]_D^{22} = -96.8°$ ($c = 1.367$). – IR.: 3380, 2900, 1712, 1645, 1618, 1450, 1258, 1207, 770. – UV.: max. 269 (4.34), 215 (4.34), 205 (4.34); min. 231 (3.79), 209 (4.33); 222 *S* (4.23) (unchanged in acid or base). – ^{1}H-NMR. ($CD_3OD + D_2O$): 7.83 (*d*, $J = 5.5$, H–C(6)); 7.67 (*d*, $J = 16$, cinnamate β-H); 7.7–7.2 (*m*, arom. H); 6.50 (*d*, $J = 16$, cinnamate α-H); 6.38 (*d*, $J = 5.5$, H–C(5)); 4.47–4.44 (*m*, 2 H–C(6′)); 3.7–3.3 (*m*, H–C(2′), H–C(3′), H–C(4′), H–C(5′)); 2.38 (*s*, H_3–C(7)). – ^{13}C-NMR.: see *Table*. – FD.-MS.: 418 (M^+). – EI.-MS.: 418 (0, M^+), 293 (11), 155 (9), 149 (5), 148 (8), 147 (8), 132 (11), 131 (100), 127 (37), 126 (52), 125 (9), 103 (26), 97 (6), 77 (14), 71 (14), 69 (6), 57 (5), 55 (11), 51 (6), 43 (18).

$C_{21}H_{22}O_9 \cdot 2\ CH_3OH$ (482.18) Calc. C 57.24 H 6.27% Found C 57.17 H 6.17%

Acetylation of **1**. To **1** (100 mg) in $CHCl_3$ (5 ml) and pyridine (5 ml) was added acetic anhydride (3 ml). The mixture was stirred at r.t. for 24 h, washed with 10% aqueous K_2CO_3-solution, dried (Na_2SO_4) and solvent was removed under reduced pressure to yield a white solid residue. Crystallization from CH_3OH afforded the triacetate **2** as colorless needles (109 mg, 91%): m.p.

186.3–187.9°, $[\alpha]_D^{22} = -81.5°$ ($c = 0.74$). – IR.: 1762, 1758, 1720, 1710, 1659, 1655, 1640, 1375, 1220, 1070, 908, 830. – UV.: max. 269 (4.18), 216 (4.14), 205 (4.12); min. 231 (3.55), 208 (4.11); 222 *S* (4.04). – ^{1}H-NMR. ($CDCl_3$): 7.66 (*d*, $J = 16$, cinnamate β-H); 7.49 (*d*, $J = 5.7$, H–C(6)); 7.6–7.3 (*m*, 5 arom. H); 6.39 (*d*, $J = 16$, cinnamate α-H); 6.28 (*d*, $J = 5.7$, H–C(5)); 5.39 (*d*, $J = 8$, H–C(1′)); 5.3–5.1 (*m*, H–C(2′), H–C(3′), therein at 5.20, $d \times d$, $J_1 = 10$, $J_2 \approx 9$, H–C(4′)); 4.30 (*d*, $J = 4$, 2 H–C(6′)); 3.75 ($d \times t$, $J_1 = 10$, $J_2 = 4$, H–C(5′)); 2.30 (*s*, H_3–C(7)); 2.13, 2.04 and 2.02 (3 *s*, 3 CH_3CO). – ^{13}C-NMR.: see *Table*. – FD.-MS.: 544 (M^+). – EI.-MS.: 419 (9), 257 (6), 211 (7), 210 (14), 132 (10), 131 (100), 127 (9), 126 (7), 109 (6), 103 (10), 91 (21), 77 (6), 57 (8), 55 (8), 44 (12), 43 (33), 41 (9).

$C_{27}H_{28}O_{12} \cdot CH_3OH$ (576.18) Calc. C 58.31 H 5.60% Found C 58.40 H 5.29%

Methanolysis of **1**. Anhydrous HCl gas was bubbled through a solution of **1** (70 mg) in dry CH_3OH (15 ml) for 6 h at 0°. The mixture was then stirred at r.t. overnight. After removal of solvent under reduced pressure, the residue was partitioned between $CHCl_3$ and water. The organic phase was dried (Na_2SO_4), concentrated and chromatographed on silica gel (20 g) eluted with $CHCl_3/CH_3OH$ 99:1 to afford methyl cinnamate as a colorless oil (19 mg, 81%). – EI.-MS.: 162 (100, M^+), 161 (10), 132 (5), 131 (54), 121 (10), 104 (7), 103 (42), 102 (9), 77 (31), 63 (7), 59 (6), 57 (13), 55 (8), 52 (10), 51 (64), 50 (22), 43 (11), 41 (8), and 2-hydroxy-3-methyl-4*H*-pyran-4-one (**4**) as an off-white solid (13 mg, 71%). – ^{1}H-NMR. ($(CD_3)_2SO$): 8.78 (br. *s*, OH); 8.01 (*d*, $J = 5.8$, H–C(6)); 6.32 (*d*, $J = 5.8$, H–C(5)); 2.25 (*s*, H_3–C(7)). – ^{13}C-NMR.: see *Table*. – EI.-MS.: 126 (100, M^+), 97 (18), 71 (61), 70 (6), 69 (12), 57 (5), 56 (10), 55 (43), 54 (9), 53 (11), 52 (10), 44 (8), 43 (65), 42 (17), 41 (10).

REFERENCES

[1] *K. Homberger & M. Hesse,* Helv. Chim. Acta *65*, 2548 (1982).

[2] *S.Y. Zolotnitskaya,* Izv. Akad. Nauk Armyan S.S.R., Biol. i Selkhoz. Nauki *7*, 27 (1954), Chem. Abstr. *48*, 11727e (1954).

[3] *V.I. Litvinenko & V.M. Darmograi,* Dopov. Akad. Nauk Ukr. R.S.R., Ser. B *30*, 639 (1968), Chem. Abstr. *71*, 13320v (1969); *V. Plouvier,* C.R. Hebd. Séances Acad. Sci., Ser. D *269*, 646 (1969); ibid. *265*, 516 (1967); *G.N. Zemtsova & S.F. Dzhumyrko,* Farmatsiya (Moscow) *25*, 26 (1976), Chem. Abstr. *85*, 90154k (1976); *V.M. Darmograi & V.I. Litvinenko,* Farm. Zh. (Kiev) *26*, 70 (1971), Chem. Abstr. *76*, 83549d (1972); *G.N. Zemtsova, V.Y. Glyzin & S.F. Dzhumyrko,* Khim. Prir. Soedin. *11*, 516 (1975); *M.J. Chopin, M.L. Bouillant, H. Wagner & K. Galle,* Phytochemistry *13*, 2583 (1974); *V.N. Darmograi,* Khim. Prir. Soedin. *12*, 540 (1976); ibid. *13*, 114 (1977); *V. Plouvier,* C.R. Hebd. Séances Acad. Sci., Ser. D *287*, 567 (1978).

[4] *E. Wagner,* Seifensieder Ztg. *68*, 35 (1941), Chem. Abstr. *35*, 3033 (1941); *A.M. Sokolskaya & A. Magzumov,* Uchenye Zapiski Kazakh. Univ. *44*, 99 (1958), Chem. Abstr. *55*, 12555c (1961).

[5] *E.T. Tegisbaev, T.K. Chumbalov & N.K. Abubakirov,* Rastit. Resur. *1*, 102 (1965), Chem. Abstr. *63*, 4573c (1965); *V.G. Bukharov & L.N. Karneeva,* Khim. Prir. Soedin. *7*, 205, 412 (1971); *D.K. Yukhananov & L.A. Sapunova,* Rastit. Resur. *12*, 244 (1976), Chem. Abstr. *85*, 30634g (1976).

[6] *T.M. Dalakishvili & E.P. Kemertelidze,* Tr. Inst. Farmakokhim., Akad. Nauk Gruz. S.S.R. *1969*, 89, Chem. Abstr. *74*, 10397b (1971).

[7] *J. Kamsteeg, J. van Brederode, F.J.E.M. Kuppers & G. van Nigtevecht,* Z. Naturforsch. C. Biosci. *33*, 475 (1978).

[8] *G.R. Jamieson & E.H. Reid,* Phytochemistry *10*, 1575 (1971).

[9] *J.E. Courtois & U. Ariyoshi,* Bull. Soc. Chim. Biol. *44*, 31 (1962); *J. Davy & J.E. Courtois,* C.R. Hebd. Séances Acad. Sci., Ser. D *261*, 3483 (1965); *iidem,* Medd. Norsk. Farm. Selsk. *28*, 197 (1966), Chem. Abstr. *67*, 91053s (1967); *J. Davy,* Ann. Pharm. Fr. *24*, 703 (1966).

[10] *N.K. Abubakirov,* in 'Front. Bioorg. Chem. Mol. Biol., Proc. Int. Symp. 1978', S.N. Ananchenko ed., pp. 257–259, Pergamon Press, Oxford 1980; *Z. Saatov, B.Z. Usmanov & N.K. Abubakirov,* Khim. Prir. Soedin. *15*, 793, 797 (1979).

[11] *C.A. Kingsbury, M. Cliffton & J.H. Looker,* J. Org. Chem. *41*, 2777 (1976).

[12] *K.R. Markham & B. Ternai,* Tetrahedron *32*, 2607 (1976); *K.R. Markham, B. Ternai, R. Stanley, H. Geiger & T.J. Mabry,* ibid. *34*, 1389 (1978).

[13] *J.N. Schumacher, C.R. Green, F.W. Best & M.P. Newell,* J. Agric. Food Chem. *25*, 310 (1977).