Revision of the Absolute Configuration of Plumericin and Isoplumericin from *Plumeria rubra*

by **Brigitta Elsässer**[a]), **Karsten Krohn***[a]), **M. Nadeem Akhtar**[a]), **Ulrich Flörke**[a]), **Simeon F. Kouam**[b]), **Merlin Guy Kuigoua**[b]), **Bonaventure T. Ngadjui***[c]), **Berhanu M. Abegaz**[d]), **Sándor Antus**[e]), and **Tibor Kurtán**[e])

[a]) Department of Chemistry, University of Paderborn, Warburger Strasse 100, D-33098 Paderborn (e-mail: karsten.krohn@uni-paderborn.de)

[b]) Department of Chemistry, Teachers' Training College, University of Yaounde 1, BP 47, Yaoundé, Cameroon

[c]) Department of Organic Chemistry, Faculty of Science, University of Yaounde 1, BP 812, Yaoundé, Cameroon

[d]) Department of Chemistry, Faculty of Science, University of Botswana, Private Bag, 0022, Gaborone, Botswana

[e]) Department of Organic Chemistry, University of Debrecen, P. O. Box 20, H-4010 Debrecen, Hungary

Dedicated to Professor *András Lipták* on the occasion of his 70th birthday

The absolute configurations of plumericin (**1**) and isoplumericin (**2**), isolated from *Plumeria rubra*, were re-assigned based on a combination of X-ray crystal-structure determination and quantum-mechanical calculations of their circular dichroism (CD) spectra. The experimental CD spectra showed an excellent match with those calculated for the (1*S*,5*R*,8*R*,9*R*,10*R*) absolute configuration (corresponding to *ent*-**1** and *ent*-**2**, resp.), opposite to that generally accepted and published in the literature. Since the (false) plumericin configuration has been often used to derive the absolute configuration of related iridoids by chemical correlation, their absolute configurations also have to be reconsidered.

1. Introduction. – More than 200 iridoids have been found to occur in nature, and the majority of them differ only in the degree and type of substituents of the basic cyclopentanopyran skeleton [1][2]. Plumericin (**1**) and isoplumericin (**2**) are important tetracyclic members of this family, occurring mainly in the genera of *Plumeria* [3–8], *Allamanda* [7][9–12], *Himantanus* [13], and *Nerium* [14]. Species of the genus *Plumeria* are widely used in traditional medicine [15]. Compounds **1** and **2** exhibit remarkable anticancer [7][11][16–19], algicidal and barnicidal [20], and antifungal [21] activities. The structural modifications of plumieride (**3**), isolated from *Plumeria bicolor*, and the effect of these modifications on *in vitro* anticancer activity, have been studied recently [19].

In the present study, we investigated the constituents of *P. rubra*, and identified plumericin (**1**) and isoplumericin (**2**), as well as their analogues plumieride (**3**) and fulvoplumierin (**4**) (*Fig. 1*). In the course of this study, we succeeded to obtain suitable crystals for single-crystal X-ray-analysis of both compounds **1** and **2** (see *Fig. 2* below). The Cartesian coordinates of these very rigid structures were ideally suited to perform quantum-chemical circular-dichroism (CD) calculations, and to compare the calculated CD spectra with the experimental ones to confirm the absolute configurations of these important natural products.

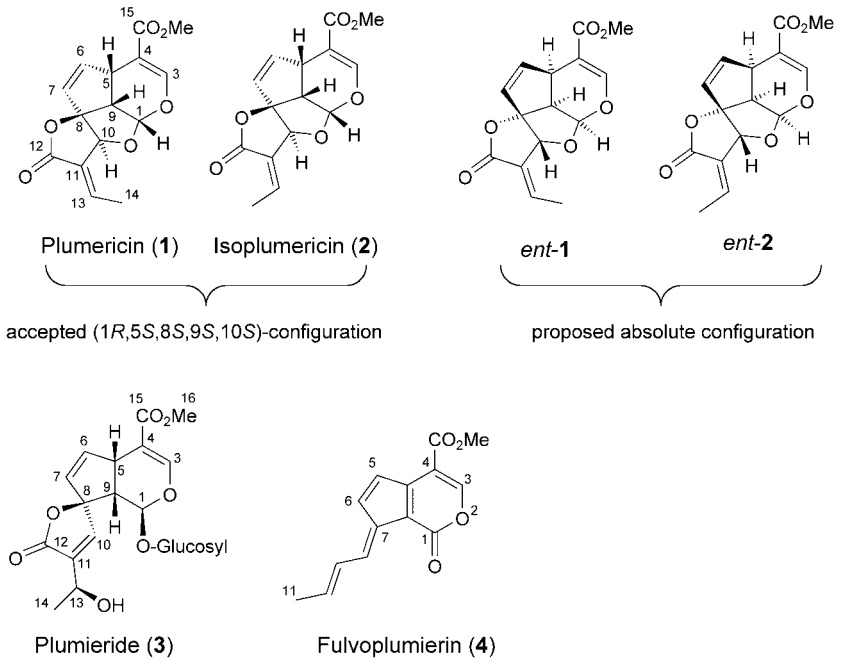


Fig. 1. *Structures of compounds isolated from* P. rubra. The accepted absolute configurations of **1** and **2** are wrong (see text), the correct structures are represented by *ent*-**1** and ent-**2**, resp.

Quantum-chemical CD calculations have been successfully applied for the determination of the absolute configurations of many natural products [22–24] (for a review, see [25]). To our great surprise, our experiments applied to putative **1** and **2** were not in agreement with the generally accepted absolute (1*R*,5*S*,8*S*,9*S*,10*S*)-configuration[1]), but suggested the *opposite* configurations, *i.e.*, those of the corresponding enantiomers *ent*-**1** and *ent*-**2** (*Fig. 1*).

2. Results and Discussion. – 2.1. *Analytical Data*. From the MeOH extract of the stem bark of *P. rubra*, plumericin (**1**) [5][14], isoplumericin (**2**) [5], fulvoplumierin (**4**) [15][26], and plumieride (**3**) [3][4] [27] were isolated, and purified by column chromatography. Their spectroscopic data were in good agreement with those reported, notably the NMR spectra [8][28][29]. The connectivities and relative configurations of **1** and **2** were further confirmed by single-crystal X-ray crystallography (*Fig. 2*). The optical rotations of plumericin (**1**) and isoplumericin (**2**) were of the same sign and order of magnitude as those reported in the literature: $[\alpha]_D^{20} = +179$ ($c = 14.6$) for **1** (literature: $[\alpha]_D^{20} = +173$ ($c = 1.10$, $CHCl_3$) [9]); and $[\alpha]_D^{20} = +194$ ($c = 0.64$, $CHCl_3$) for **2** (literature: $[\alpha]_D^{20} = +189$ ($c = 0.61$, $CHCl_3$) [9]). This, thus, confirmed identical absolute configurations of our samples with those isolated previously [5][9]. In papers dealing with these compounds, their absolute configurations are represented as shown in *Fig. 1* [5][9][12][14][16][30]. The assignment of the absolute configuration of

[1]) Arbitrary atom numbering (see *Fig. 1*). For systematic names, see *Exper. Part*.

plumieride (**3**) relied mainly on the formation of (+)-ethylsuccinic acid upon oxidation of a 13-deoxy reduced derivative of **3** [4]. In another paper, plumericin (**1**) was chemically connected by common degradation products with plumieride (**3**) [5]. However, neither **1** nor **3** possess a stereogenic center at C(11), which is ultimately correlated to (+)-ethylsuccinic acid; moreover, the literature assignments were sophisticated, but lengthy and very indirect.

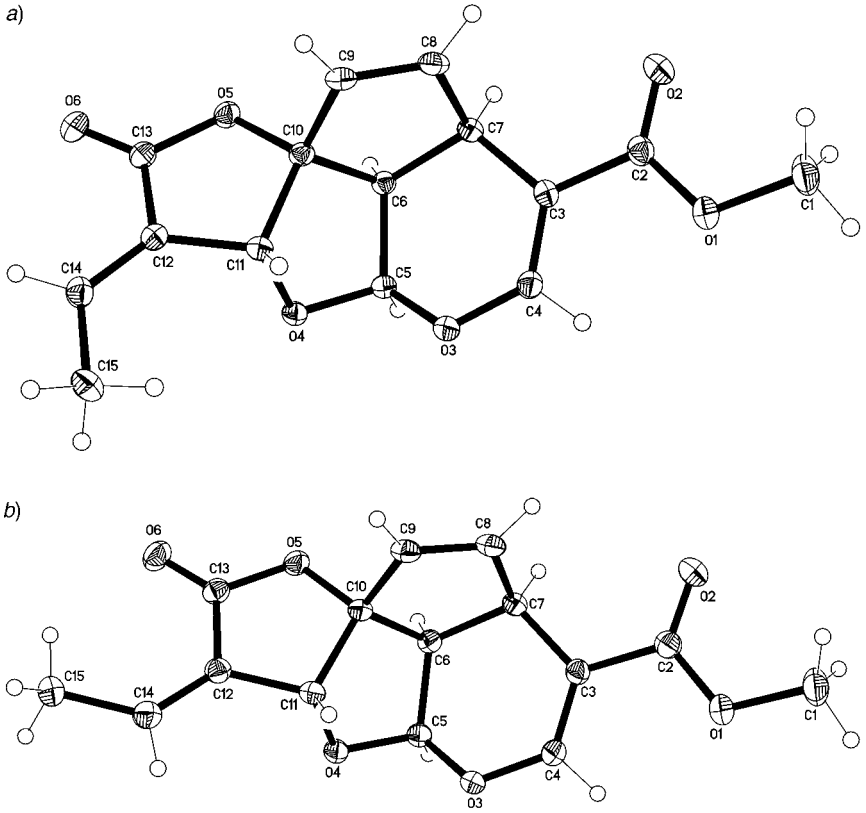
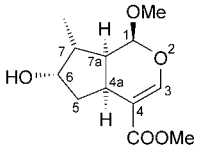


Fig. 2. *X-Ray crystal structures of* a) *plumericin* (*ent*-**1**) *and* b) *isoplumericin* (*ent*-**2**)

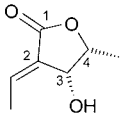
2.2. *Computational Approach.* Both plumericin (**1**) and isoplumericin (**2**) contain α,β-unsaturated ester and α,β-unsaturated γ-lactone chromophores, whose overall contribution determines their CD characteristics. The X-ray data of **1** and **2** revealed that the orientations of the two endocyclic C=C bonds relative to the corresponding C=O groups are almost planar in each chromophore, with dihedral angles C(13)–C(11)–C(12)–O and C(3)–C(4)–C(15)–O of -4.9° and -179.0° in the case of **1**, and of -7.9° and -178.2° in that of **2**, respectively. This implies that the unsaturated ester and γ-lactone chromophores can be viewed as inherently nonchiral chromophores, and, hence, their CD transitions derive from the chiral perturbation of the *neighboring* stereogenic centers. Although there are empirical rules for chirally perturbed planar enone chromophores [31], they cannot be used safely for complex

molecules such as **1** or **2**. This may be the reason why CD spectroscopy has not been exploited for the configurational assignment and characterization of these compounds. However, quantum-mechanical calculations of their CD spectra, and comparison with the experimental ones, offers a possibility to determine their absolute configurations by CD spectroscopy, especially in combination with X-ray crystallography.

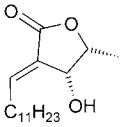
Recently, we determined the absolute configurations of several natural products by means of quantum-mechanical calculations of CD spectra [22–24]. In order to probe the method on plumericin (**1**), and to test whether the program would give reliable results for these complicated chromophores, we calculated the CD spectra of the two plumericin fragments **5** and **6**, which contain α,β-unsaturated ester and γ-lactone moieties, respectively. These fragments were selected because the CD data of the loganin derivative **5** [32], and that of 3-epilitsenolide D_2 (**7**) [33], are available in the literature.



5



6



3-Epilitsenolide D_2 (**7**)

The CD spectrum of a given compound is the sum of the CD spectra of all individual conformers. The CD spectra of the individual conformers can be calculated quantum-mechanically with, *e.g.*, the software package DZDO Program (Version 4.23) [34]. To get an overall CD spectrum that resembles the measured one, all major conformers of a given compound have to be considered according to their relative abundances. Therefore, first, a conformational analysis of fragment **5** was performed with Spartan SGI (Version 5.3). A Monte Carlo search, with the MMFF 94 force field, was applied to obtain low-energy conformers [35], whose heat of formation (ΔH_f) was subsequently computed by the semi-empirical AM1 method, which is incorporated in the Spartan program package [32]. All conformers of **5**, up to a given energy cut-off of 10–12 kJ/mol (*i.e.*, $>1\%$ abundance at 298 K) were taken into consideration. Next, the CD spectra of each conformer was computed [34], and then added up, the contribution of each conformer being determined by the *Boltzmann* statistics as:

$$\frac{N_i}{N_j} = e^{-\frac{E_i - E_j}{kT}} \quad (1)$$

In the case of the relatively rigid and conformationally restricted fragment **5**, the conformational analysis resulted in only one low-energy conformer, whose ΔH_f value and CD spectrum (*Fig. 3*) were computed. The calculated CD curve of **5** exhibited a strong positive *Cotton* effect at 238 nm, which agreed very well with the literature data (*Table 1*) [32].

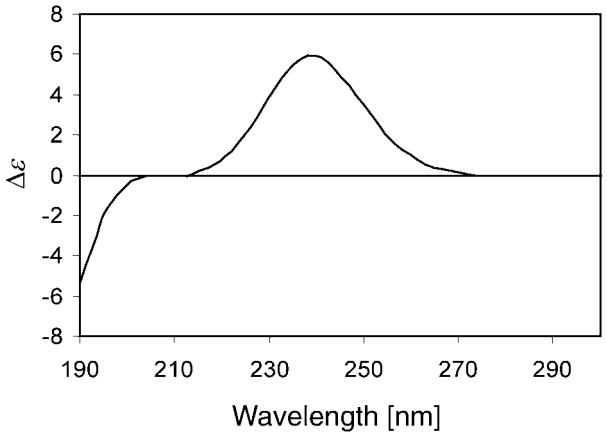


Fig. 3. *Calculated CD spectrum of the loganin derivative* **5**

Table 1. *Calculated Circular-Dichroism Data of the Fragments* **5** *and* **6**, *and Comparison with Experimental Data*. Positive and negative Cotton effects (CE) are indicated (+/−).

Fragment	Calculated		Experimental		
	λ [nm]	CE	λ [nm]	Δε	Ref.
5	238	+	236	+5.90	[32]
6[a])	219	−	226	−9.74	[33]

[a]) The experimental values refer to the analogue **7**.

A similar conformational analysis of fragment **6** resulted in four major conformers. Their individual CD spectra thus obtained were added up by the *Boltzmann* statistics using the appropriate heats of formation, to give the calculated overall CD spectrum of **6** (*Fig. 4*). The calculated CD spectrum exhibited a negative couplet at 219 nm, again in accordance with the literature data (226 nm; *Table 1*) [33]. The results of these two calculations demonstrate the accuracy of the applied calculations and also confirmed the absolute configuration of the loganin derivative **5**, which is homochiral with the corresponding substructure of the more-condensed plumericins (**1** and **2**; hitherto accepted configuration) regarding the stereogenic centers C(1), C(4a), and C(7a) of **5**. Moreover, the C(3) stereogenic center of **6** is homochiral with the corresponding C(10)[1]) stereogenic center of *ent*-**1** (proposed configuration).

Next, the coordinates of the X-ray structure were used for the calculation of the CD spectra of plumericin (**1**) and isoplumericin (**2**). Since both compounds are very rigid, the number of low-energy conformers is limited, which facilitated the quantum-mechanical calculations. For comparison, semi-empirical structure optimization and conformational analysis of plumericin (**1**) and isoplumericin (**2**) were performed. As expected, due to the very rigid tetracyclic structure, this optimization resulted in a single low-energy conformer for both compounds. Also, these conformers were virtually identical with the corresponding X-ray structures. Therefore, the X-ray structures were taken for further CD calculations without any restriction.

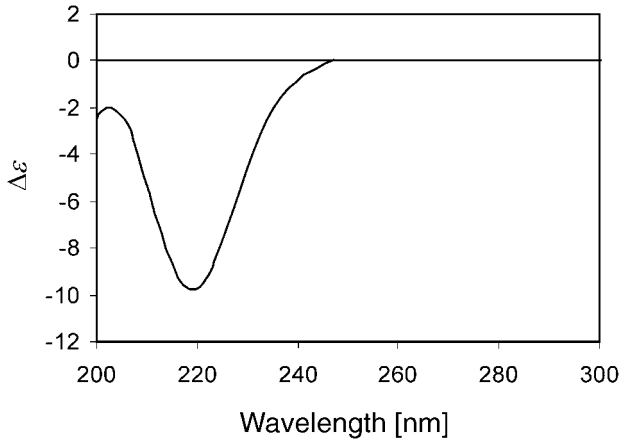


Fig. 4. *Calculated CD spectrum of fragment* **6**

The results of the calculated CD spectra for the arbitrarily selected *ent*-**1** and *ent*-**2**, and those measured for our plumericin and isoplumericin samples, are shown in *Fig. 5*. The matches were exceptionally good in both cases, probably because the X-ray structures indeed reflect the true conformations. Both the calculated and experimental curves showed a positive *Cotton* effect at *ca.* 191 nm ($n \rightarrow \sigma^*$ transition of the enone), and a negative and positive effect at 214 and 237 nm, respectively ($\pi \rightarrow \pi^*$ transition of the enone).

The excellent agreement of the data leaves no doubt that the absolute configuration of both plumericin (**1**) and isoplumericin (**2**) actually corresponds to (1*S*,5*R*,8*R*,9*R*,10*R*), and that the true structures of these compounds are those termed *ent*-**1** and *ent*-**2** (see *Fig. 1*), *i.e.*, the arbitrarily selected configuration for the calculation. It is also noteworthy that the measured CD spectra of plumericin and isoplumericin also contain a weak negative *Cotton* effect at 269 and 267 nm, respectively, which, most likely, stems from the $n \rightarrow \pi^*$ transition of the α,β-unsaturated γ-lactone chromophore. For plumieride (**3**), this transition has a weak positive *Cotton* effect, and the second transition, the $\pi \rightarrow \pi^*$ transition, is also inverted at 226 nm, which can be formally attributed to the 'switch' to an s-*trans* α,β-unsaturated γ-lactone from the s-*cis* form of **1** [31]. Since plumieride (**3**) [5][10][12] was chemically correlated with plumericin (**1**) [36] regarding C(5), C(8), and C(9), the two must be homochiral, and the absolute configuration of **3**, thus, also has to be revised.

Our results leave no choice but to revise the absolute configurations not only of the iridoids above, but probably also of most of the related, more-condensed iridoids such as allamcin [15], allamandicin [11][15][18], plumieride coumarate [20], oruwacin [20][37], and all of their respective glucosides. The question of absolute configuration could unambiguously be settled by X-ray structure analysis of one of the D-glucosides. Unfortunately, despite many attempts, we were not able to get suitable crystals of plumieride (**3**) or its derivatives.

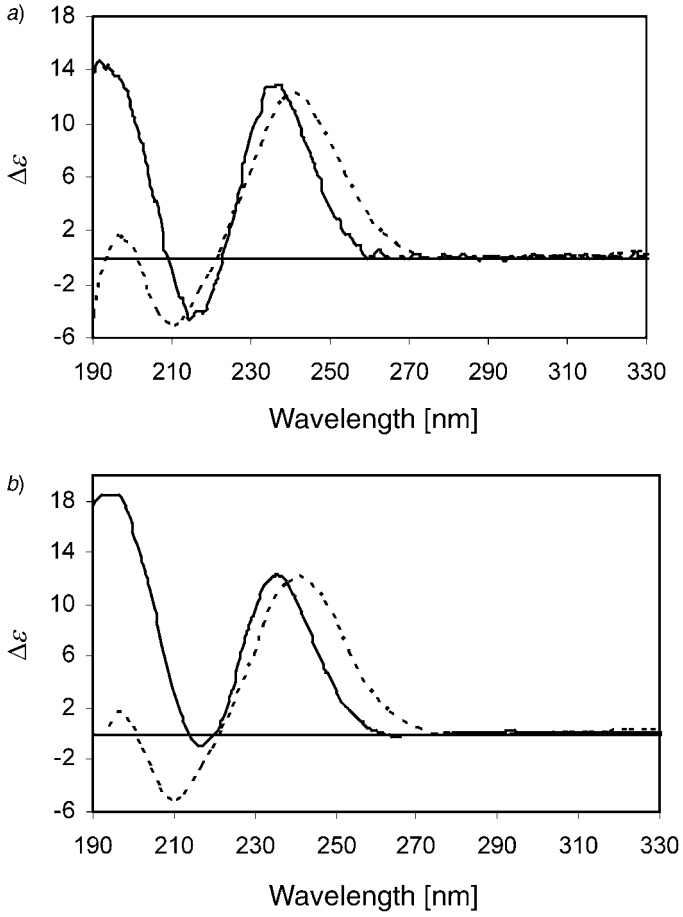


Fig. 5. *Comparison of calculated* vs. *exerimental CD spectra of* a) plumericin (*ent*-**1**) *and* b) *isoplumericin* (*ent*-**2**). Calculated spectra are represented by dashed lines (- - -), experimental ones by solid lines (——).

3. Conclusions. – Comparison of the quantum-mechanically calculated CD spectrum of plumericin and isoplumericin with the experimental spectra unequivocally settled their absolute configurations as (1*S*,5*R*,8*R*,9*R*,10*R*), which corresponds to the structures *ent*-**1** and *ent*-**2**, respectively, shown in *Fig. 1*. This absolute configuration is opposite to that of the generally assumed structures **1** and **2**. In addition, the configurations of all biogenetically linked or chemically correlated natural products should also be revised.

Experimental Part

General. Column chromatography (CC) was carried out on silica gel (70–270 or 230–400 mesh; *Merck*). Thin-layer chromatography (TLC) was performed on *Merck* precoated silica gel *60* F_{254} plates on aluminum foil, the eluent being hexane/acetone 60:40. Melting points (m.p.) were measured on a *Büchi SMP-20* melting-point apparatus; uncorrected. UV Spectra: *UV-210 PC* scanning spectrophotometer; λ_{max} (ε) in nm. Optical rotations

were measured on a *Perkin-Elmer* polarimeter in a 1-ml cell tube. CD Spectra: *JASCO J-810* polarimeter; at ambient temperature; λ ($\Delta\varepsilon$) in nm. IR Spectra: *Nicolet 510-P FT-IR* spectrometer; in cm^{-1}. NMR Spectra: *Bruker Avance-500* NMR spectrometer (500/125 MHz); chemical shifts δ in ppm rel. to residual solvent peaks (δ(H) 7.25 for $CDCl_3$, and 3.33 and 4.88 for CD_3OD), coupling constants *J* in Hz. Mass spectra: *Finnigan MAT-8230* double-focusing mass spectrometer; in *m/z* (rel. %).

Plant Material. The bark of *P. rubra* was collected in Yaounde, Cameroon, near the Higher Teachers' Training College, in December 2003. A voucher specimen (PM N° 8588/SRF/Cam.) was authenticated by *Mr. Nana*, botanist at the Cameroon National Herbarium, Yaounde, Cameroon, where it was deposited.

Extraction and Isolation. The air-dried, powdered bark of *P. rubra* (3.5 kg) was macerated in a mixture of CH_2Cl_2/MeOH 1:1 and then neat MeOH for 48 h and 6 h, resp. After removal of the solvents of the combined extracts by evaporation under reduced pressure, the total extract (570.0 g) was suspended in distilled H_2O. The resulting aq. layer was extracted with AcOEt (5×), and the extract was concentrated under reduced pressure. A part of the AcOEt extract (100.0 g) was subjected to CC (SiO_2; 1. hexane/AcOEt 1:0 → 0:1; 2. AcOEt/MeOH 9:1; 3. MeOH): eight subfractions (*Fr.A – H*). Further fractions of *Fr.B* were eluted using a step gradient of hexane/AcOEt: *Fr.1 – 35* (each *ca*. 150 ml). From *Fr.10*, compound **4** was isolated; *Fr.12 – 16* gave **1** and **2**; and *Fr.25 – 30* afforded **3**. The remaining fractions (*Fr.31 – 35*) gave a mixture of the common sterols stigmasterol and β-sitosterol, which were identified by comparison with authentic samples.

Plumericin (= *Methyl (3*E*,3a*R*,4a*S*,7a*R*,9a*R*,9b*R*)-3-Ethylidene-3,3a,7a,9b-tetrahydro-2-oxo-2*H*,4a*H*-1,4,5-trioxadicyclopenta[*a,hi*]indene-7-carboxylate* (*ent*-**1**)). Colorless crystals. M.p. 205 – 208° (dec.; lit. 211.5 – 212.5° [5]). UV (MeCN): 237 (sh, 11400), 213 (sh, 18900), 200 (21000). $[\alpha]_D^{20} = +179$ ($c = 14.6$, $CHCl_3$; lit. +173 ($c = 1.10$, $CHCl_3$) [10]). CD (MeCN, $c = 0.68$ mM): 268 (−0.13), 237 (12.91), 214 (−4.7), 191 (14.73). IR ($CHCl_3$): 1755 (α,β-unsat. lactone), 1796 (ester C=O), 1635, 1524 (C=C). ^{1}H-NMR ($CDCl_3$, 500 MHz)[1]): 2.10 (*d*, *J* = 7.5, Me(14)); 3.45 (*dd*, *J* = 9.5, 6.0, H−C(9)); 3.75 (*s*, MeOOC); 4.01 (*td*, *J* = 9.5, 2.5, H−C(5)); 5.11 (*s*, H−C(10)); 5.55 (*d*, *J* = 6.0, H−C(1)); 5.65 (*dd*, *J* = 5.5, 2.5, H−C(6)); 6.05 (*dd*, *J* = 5.5, 2.5, H−C(7)); 7.15 (*dq*, *J* = 7.5, 1.5, H−C(13)); 7.49 (*s*, H−C(3)). ^{13}C-NMR ($CDCl_3$, 125 MHz)[1]): 16.2 (*q*, C(14)); 38.4 (*d*, C(5)); 51.7 (*q*, C(16)); 53.7 (*d*, C(9)); 80.3 (*d*, C(10)); 102.3 (*d*, C(1)); 104.6 (*s*, C(4)); 109.2 (*s*, C(8)); 126.3 (*d*, C(6)); 127.4 (*s*, C(11)); 141.1 (*d*, C (7)); 145.6 (*d*, C(13)); 152.3 (*d*, C(3)); 166.6 (*s*, C(15)); 168.2 (*s*, C(12)). EI-MS: 290 (100, M^+, $C_{15}H_{14}O_6^+$), 258 (87), 230 (90), 201 (70), 193 (80), 160 (73), 139 (80), 57 (78).

Isoplumericin (= *Methyl (3*Z*,3a*R*,4a*S*,7a*R*,9a*R*,9b*R*)-3-Ethylidene-3,3a,7a,9b-tetrahydro-2-oxo-2*H*,4a*H*-1,4,5-trioxadicyclopenta[*a,hi*]indene-7-carboxylate* (*ent*-**2**)). Colorless crystals. M.p. 207 – 211° (dec.; lit. 199 – 200° [10]). UV (MeCN): 238 (sh, 11900), 210 (20400). $[\alpha]_D^{20} = +179$ ($c = 14.6$, $CHCl_3$; lit. +189 ($c = 0.61$, $CHCl_3$) [10]). CD (MeCN, $c = 0.53$ mM): 267 (−0.28), 235 (12.28), 216 (−0.87), 193 (18.53). ^{1}H-NMR ($CDCl_3$, 500 MHz)[1]): 2.35 (*d*, *J* = 6.5, Me(14)); 3.45 (*dd*, *J* = 9.5, 6.0, H−C(9)); 3.75 (*s*, MeOOC); 4.01 (*td*, *J* = 9.5, 6.0, H−C(5)); 5.11 (*s*, H−C(10)); 5.55 (*d*, *J* = 6.0, H−C(1)); 5.65 (*dd*, *J* = 5.5, 2.2, H−C(6)); 6.05 (*dd*, *J* = 5.5, 2.2, H−C(7)); 6.84 (*dq*, *J* = 7.5, 1.5, H−C(13)); 7.44 (*s*, H−C(3)). ^{13}C-NMR ($CDCl_3$ 125 MHz)[1]): 14.8 (*q*, C(14)); 38.4 (*d*, C(5)); 51.6 (*q*, C(16)); 51.7 (*d*, C(9)); 83.7 (*d*, C(10)); 101.3 (*d*, C(1)); 104.1 (*s*, C(4)); 109.2 (*s*, C(8)); 126.1 (*d*, C(6)); 125.4 (*s*, C(11)); 141.1 (*d*, C (7)); 145.6 (*d*, C(13)); 152.3 (*d*, C(3)); 166.6 (*s*, C(15)); 168.2 (*s*, C(12)). EI-MS: 290 (100, M^+, $C_{15}H_{14}O_6^+$), 258 (87), 230 (32), 201 (74), 193 (82), 160 (73), 139 (81), 57 (78).

Plumieride (= *Methyl 1-(Glucopyranosyloxy)-4a,7a-dihydro-4′-(1-hydroxyethyl)-5′-oxo-1*H*,5′*H*-spiro[cyclopenta[c]pyran-7,2′-furan]-4-carboxylate*; **3**). Colorless powder. M.p. 205 – 220° (lit. 225 – 226° [9]). UV (MeCN): 282 (sh, 0.6), 238 (sh, 11.9), 210 (20.4). $[\alpha]_D^{20} = -71.7$ ($c = 1.4$, MeOH; lit. −68 ($c = 0.9$, MeOH [9]). CD (MeCN, $c = 0.23$ mM): 264 (0.58), 226 (−20.19), 196 (16.46). CD (MeOH, $c = 0.32$ mM): 269 (0.27), 228 (−20.27), 193 (18.68). ^{1}H-NMR ((D$_5$)pyridine, 500 MHz)[1]): 1.72 (*d*, *J* = 6.5, Me(14)); 3.15 (*d*, *J* = 10.5, 6.0, H−C(9)); 3.75 (*s*, Me(16); 4.5 – 3.90 (glucosyl); 5.11 (*m*, H−C(13)); 5.43 (*d*, *J* = 8.0, H−C(1′)); 5.49 (*dd*, *J* = 5.5, 3.0, H−C(7)); 5.66 (*d*, *J* = 5.5, H−C(1)); 6.55 (*dd*, *J* = 5.5, 2.2, H−C(6)); 7.60 (*s* H−C(3)); 7.70 (*d*, *J* = 2.0, H−C(10)). ^{13}C-NMR ($CDCl_3$, 125 MHz): 23.9 (*q*, C(14)); 43.1 (*d*, C(5)); 50.1 (*d*, C(9)); 52.1 (*q*, COO*Me*); 63.1 (*t*, C(6′)); 63.6 (*d*, C(13)); 71.7 (*d*, C(4′)); 75.6 (*d*, C(2′)); 79.1 (*d*, C(3′)); 79.7 (*d*, C(5′)); 94.9 (*d*, C(1)); 97.4 (*s*, C(8)); 101.7 (*d*, C(1′)); 110.5 (*s*, C(4)); 130.1 (*d*, C(7)); 139.6 (*s*, C(11)); 141.8 (*d*, C(6)); 150.0 (*d*, C(10)); 152.3 (*d*, C(3)); 167.6 (*s*, C(15)); 171.3 (*s*, C(12)). EI-MS: 290 (100, M^+), 258 (87), 230 (90), 201 (70), 193 (80), 160 (73), 139 (80), 57 (78).

Fulvoplumierin (= *Methyl (7*E*)-1,7-Dihydro-7-[(2*E*)-but-2-en-1-ylidene]-1-oxo-cyclopenta[c]pyran-4-carboxylate*; **4**). Orange crystals. M.p. 140 – 142° (dec.; lit. 150 – 152° [10]). ^{1}H-NMR ($CDCl_3$, 500 MHz): 2.02 (*d*, *J* = 7.5, Me(11)); 3.92 (*s*, MeOOC)); 6.55 (*dq*, *J* = 14.5, 11.5, H−C(10)); 6.85 (*dq*, *J* = 14.5, 11.5, 1.5, H−C(9)); 7.23 (*d*, *J* = 5.3, H−C(5)); 7.35 (*d*, *J* = 5.3, H−C(6)); 7.96 (*d*, *J* = 11.5, H−C(8)); 8.29 (*s*, H−C(3)). ^{13}C-NMR ($CDCl_3$, 125 MHz): 19.5 (*q*, C(11)); 52.0 (*q*, *Me*OOC)); 109.9 (*s*, C(6)); 113.3 (*s*, C(4)); 127.4 (*d*, C(6)); 129.6 (*d*, C(9));

130.2 (*d*, C(5)); 136.8 (*q*, C(7)); 143.4 (*d*, C(8)); 145.5 (*d*, C(10)); 150.3 (*s*, C(4a)); 156.5 (*d*, C(3)), 157.6 (*s*, MeOO*C*), 164.3 (*s*, C(1)). EI-MS: 244 (n, M^+, $C_{14}H_{12}O_4^+$), 212 (38), 149 (79), 115 (15), 44 (100).

X-Ray Crystal-Structure Determination[2]). Data of compounds *ent*-**1** and *ent*-**2** were collected on a *Bruker AXS SMART APEX* diffractometer with graphite monochromated (MoK_α) radiation ($\lambda = 0.71073$ Å). Both compounds crystallized in noncentrosymmetric space groups; however, in the absence of significant anomalous scattering effects, the *Flack* parameters are essentially meaningless. Accordingly, *Friedel* pairs were merged. Structures were solved by direct methods, with full-matrix least-squares refinements based on F^2. All but the H-atoms were refined anisotropically. The H-atoms were derived from difference *Fourier* maps, and refined at idealized positions, riding on their attached C-atoms, with isotropic displacement parameters $U_{iso}(H) = 1.5U_{eq}(Me)$ or $1.2U_{eq}(C)$. All Me groups were allowed to rotate, but not to tip. The *Bruker* software package [39] was used. The molecular structures of *ent*-**1** and *ent*-**2** are shown in *Fig. 2*, and selected crystallographic data are summarized in *Table 2*.

Table 2. *Crystallographic Data of Compounds* **1** *and* **2**

	1	**2**
Empirical formula	$C_{15}H_{14}O_6$	$C_{15}H_{14}O_6$
Molecular weight	290.26	290.26
Temperature [K]	120(2)	120(2)
Wavelength [Å]	0.71073	0.71073
Crystal system	Monoclinic	Orthorhombic
Space group	$P\,2_1$	$P\,2_12_12_1$
Unit-cell dimensions:		
a [Å]	4.7594(3)	4.7564(2)
b [Å]	7.8798(5)	8.0058(4)
c [Å]	17.3684(10)	34.5066(17)
β [°]	96.443(1)	–
Volume [Å^3]	647.26(7)	1314.0(1)
Z	2	4
D_{calc} [Mg/cm^3]	1.489	1.467
Absorption coefficient [mm^{-1}]	0.116	0.115
$F(000)$	304	608
Crystal size [mm^3]	$0.45 \times 0.15 \times 0.15$	$0.50 \times 0.38 \times 0.20$
Data-collection range	$2.36 \leq \Theta \leq 28.23°$	$2.36 \leq \Theta \leq 28.24°$
Index ranges (h, k, l)	$-6/6,\ -10/10,\ -22/22$	$-6/6,\ -10/10,\ -42/45$
Reflections collected	8198	13850
Independent reflections	1717 [R(int) = 0.0180]	1945 [R(int) = 0.0468]
Absorption correction	Semi-empirical from eq.	Semi-empirical from eq.
Max. and min. transmission	0.983 and 0.949	0.977 and 0.945
Refinement method	Full-matrix least-sq. on F^2	Full-matrix least-sq. on F^2
Data/restraints/parameters	1717/1/192	1945/0/192
Goodness-of-fit on F^2	1.114	1.164
R_{final} ($I > 2\sigma(I)$)	$R1 = 0.0322$, $wR2 = 0.0835$	$R1 = 0.0359$, $wR2 = 0.0957$
R (all data)	$R1 = 0.0327$, $wR2 = 0.0839$	$R1 = 0.0373$, $wR2 = 0.0966$
Largest diff. peak and hole [eÅ^{-3}]	0.363 and -0.155	0.337 and -0.162

A. S. and *T. K.* thank the *National Science and Research Foundation*, Hungary (OTKA, T-049436, F-043536) for financial support. *T. K.* is indebted to the *Hungarian Academy of Sciences* for the *Bolyai Postdoctoral Fellowship.*

---

[2]) The crystallographic data of *ent*-**1** and *ent*-**2** have been deposited with the *Cambridge Crystallographic Data Centre* as supplementary publication numbers CCDC-258400 (*ent*-**1**) and -258401 (*ent*-**2**). The data can be obtained, free of charge, *via* the internet at http://www.ccdc.cam.ac.uk/data_request/cif.

REFERENCES

[1] A. Bianco, in 'Studies in Natural Products Chemistry: Structure and Chemistry, Part. A', Ed. Atta-ur-Rahman, Elsevier, Amsterdam, 1990, Vol. 7, p. 439 – 497.
[2] A. Bianco, *Pure Appl. Chem.* **1994**, *66*, 2335.
[3] H. Schmid, H. Bickel, T. M. Meijer, *Helv. Chim. Acta* **1952**, *35*, 415.
[4] O. Halpern, H. Schmid, *Helv. Chim. Acta* **1958**, *41*, 1109.
[5] G. Albers-Schönberg, H. Schmid, *Helv. Chim. Acta* **1961**, *44*, 1447.
[6] R. K. Chaudhuri, F. U. Afifi-Yazar, O. Sticher, T. Winkler, *Tetrahedron* **1980**, *36*, 2317.
[7] J. J. W. Coppen, A. L. Cobb, *Phytochemistry* **1983**, *22*, 125.
[8] F. Abe, R.-F. Chen, T. Yamauchi, *Chem. Pharm. Bull.* **1988**, *36*, 2484.
[9] M. S. Abdel-Kader, J. Wisse, R. Evans, H. van der Werff, D. G. I. Kingston, *J. Nat. Prod.* **1997**, *60*, 1294.
[10] F. Abe, T. Mōri, T. Yamauchi, *Chem. Pharm. Bull.* **1984**, *32*, 2947.
[11] S. M. Kupchan, A. L. Dessertine, B. T. Blaylock, R. F. Bryan, *J. Org. Chem.* **1974**, *39*, 2477.
[12] T. Yamauchi, F. Abe, M. Taki, *Chem. Pharm. Bull.* **1981**, *29*, 3051.
[13] M. de Fátima Vanderlei, M. S. da Silva, *J. Braz. Chem. Soc.* **1991**, *2*, 51.
[14] D. Basu, A. Chatterjee, *Indian J. Chem.* **1973**, *11*, 297.
[15] L. B. S. Kardono, S. Tsauri, K. Padmawinata, J. M. Pezzuto, A. D. Kinghorn, *J. Nat. Prod.* **1990**, *53*, 1447.
[16] K. E. B. Parkes, G. Pattenden, *J. Chem. Soc., Perkin Trans. 1* **1988**, 1119.
[17] S. Jolad, J. J. Hoffmann, R. M. Wiedhopf, J. R. Cole, R. B. Bates, G. R. Kriek, *Tetrahedron Lett.* **1976**, *46*, 4119.
[18] S. M. Kupchan, A. L. Dessertine, B. T. Blaylock, R. F. Bryan, *J. Org. Chem.* **1974**, *39*, 2478.
[19] M. P. Dobhal, G. Li, A. Gryshuk, A. Graham, A. K. Bhatanager, S. D. Khaja, Y. C. Joshi, M. C. Sharma, A. Oseroff, R. K. Pandey, *J. Org. Chem.* **2004**, *69*, 6165.
[20] J. J. W. Coppen, *Phytochemistry* **1983**, *22*, 179.
[21] K. Jewers, J. J. W. Coppen, H. Manchanda, A. H. Paw, A. Castillo, *Pahlavi Med. J.* **1975**, *6*, 52.
[22] Y. Lin, X. Wu, S. Feng, G. Jiang, J. Luo, S. Zhou, L. L. P. Vrijmoed, E. B. G. Jones, K. Krohn, K. Steingröver, F. Zsila, *J. Org. Chem.* **2001**, *66*, 6252.
[23] G. Bringmann, S. Busemann, K. Krohn, K. Beckmann, *Tetrahedron* **1997**, *53,* 1655.
[24] K. Krohn, U. Flörke, M. John, N. Root, K. Steingröver, H.-J. Aust, S. Draeger, B. Schulz, S. Antus, M. Simonyi, F. Zsila, *Tetrahedron* **2001**, *57*, 4343.
[25] G. Bringmann, S. Busemann, in 'Natural Product Analysis', Eds. P. Schreier, M. Herderich, H.-U. Humpf, W. Schwab, Vieweg, Braunschweig, Wiesbaden, 1998, p. 195 – 212.
[26] R. Sanduja, A. J. Weinheimer, K. L. Euler, M. Alam, *J. Nat. Prod.* **1985**, *48*, 335.
[27] A. de Sá Barreto, M. G. de Carvalho, I. de Almeida Nery, L. Gonzaga, M. A. Coelho Kaplan, *J. Braz. Chem. Soc.* **1998**, *9*, 430.
[28] G. E. Martin, R. Sanduja, M. Alam, *J. Org. Chem.* **1985**, *50*, 2383.
[29] L. J. El-Naggar, J. L. Beal, *J. Nat. Prod.* **1980**, *43*, 649.
[30] B. M. Trost, J. M. Balkvec, M. K.-T. Mao, *J. Am. Chem. Soc.* **1983**, *105*, 6755.
[31] D. A. Lightner, J. E. Gurst, 'Organic Conformational Analysis and Stereochemistry from Circular Dichroism Spectroscopy', J. Wiley & Sons, New York, 2000, p. 337 – 394.
[32] L.-F. Tietze, U. Niemeyer, P. Marx, K.-H. Glüsenkamp, L. Schwenen, *Tetrahedron* **1980**, *36*, 735.
[33] S.-S. Lee, S. M. Chang, C.-H. Chen, *J. Nat. Prod.* **2001**, *64*, 1548.
[34] J. W. Downing, DZDO Program, Version 4.23 (ZDO Program, Version of 22.06.2001), Department of Chemistry and Biochemistry, University of Colorado, Boulder, USA.
[35] Spartan SGI, Version 5.1.3, *Wavefunction, Inc.*, Irvine, USA.
[36] D. A. Yowell, H. Schmid, *Experientia* **1964**, *20*, 250.
[37] E. K. Adesogan, *Phytochemistry* **1979**, *18*, 175.
[38] Bruker (2002), SMART (Version 5.62), SAINT (Version 6.02), SHELXTL (Version 6.10), and SADABS (Version 2.03); *Bruker AXS, Inc.*, Madison, Wisconsin, USA.

Received December 16, 2004