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Iridoid glycosides from *Gmelina philippensis*

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Abstract

Six new iridoids, 6-*O*-α-L-(2″-*O*-, 3″-*O*-, 4″-*O*-tribenzoyl)rhamnopyranosylcatalpol, 6-*O*-α-L-(2″-*O*-, 3″-*O*-dibenzoyl, 4″-*O*-*cis*-*p*-coumaroyl)rhamnopyranosylcatalpol, 6-*O*-α-L-(2″-*O*-, 3″-*O*-dibenzoyl, 4″-*O*-*trans*-*p*-coumaroyl)rhamnopyranosylcatalpol, 6-*O*-α-L-(2″-*O*-benzoyl, 3″-*O*-*trans*-*p*-coumaroyl)rhamnopyranosylcatalpol, 6-*O*-α-L-(2″-*O*-, 3″-*O*-dibenzoyl)rhamnopyranosylcatalpol, and gmephiloside as well as five known monoacyl and diacyl rhamnopyranosylcatalpol derivatives were isolated from the aerial parts of *Gmelina philippensis*. Their structures were established by spectroscopic means. Additionally, the known iridoids catalpol, geniposidic acid, gardoside, and 8-*epi*-loganic acid were identified and quantified by GC and GC–MS. The taxonomic significance of rhamnopyranosylcatalpol derivatives and iridoid acids as chemical characters is discussed.

Keywords: *Gmelina philippensis*; Viticoideae; Scutellarioideae; Lamiaceae; Verbenaceae; Isolation; Chemotaxonomy; Iridoid; Rhamnopyranosylcatalpol esters; Iridoid acids

1. Introduction

The genus *Gmelina* (ca. 40 species) is widely distributed in the tropical and subtropical regions of Australia, Asia and Africa. Briquet (1885) and Moldenke (1971) included *Gmelina* and *Premna* along with *Vitex* and some smaller genera in the tribe Viticeae of the subfamily Viticoideae and family Verbenaceae. Cantino (1992a) and Cantino et al. (1992b) introduced a revised classification and transferred the subfamily Viticoideae along with most genera of the former Verbenaceae to the Lamiaceae s.l. However, recent cladistic analyses of cpDNA sequences by Wagstaff et al. (1998) and Olmstead et al. (1998) suggested that neither the Viticoideae nor the former tribe Viticeae form a monophyletic group. They found a clade consisting of *Premna* and *Gmelina* emerging in a weakly supported position as sister group to a clade consisting of the subfamilies Scutellarioideae, Pogostemonoideae and Lamioideae.

Investigations of several *Premna* species for iridoids revealed a heterogeneous iridoid pattern. Rhamnopyranosylcatalpol esters (e.g. Otsuka et al., 1989, 1990, 1991), other acylated derivatives of catalpol (e.g. Sudo et al., 1997, 1998), iridoid acids and derivatives of iridoid acids (e.g. Bheemasankara Rao et al., 1981; Otsuka et al., 1992) were isolated from different *Premna* species. In contrast to these numerous studies, the distribution of iridoids in *Gmelina* species is only poorly known. Apart from the early chromatographic investigations of Kooiman (Hegnauer and Kooiman, 1978) only one species, *G. arborea* Roxb. (Hosny and Rosazza, 1998), has recently been examined for iridoids. Therefore, we investigated a second species, *G. philippensis* CHAM. (syn. *G. hystrix* Schult. ex Kurz). We here report the isolation and structure elucidation of three new triacyl, two new diacyl and five known monoacyl and diacyl rhamnopyranosylcatalpol derivatives, one new derivative of 8-*epi*-loganic acid as well as the identification of five other known iridoids.

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Table 1
^{1}H NMR spectral data[a] (500 MHz) of compounds **1**–**5** and **16**

H	1[b, c]	2[b, d]	3[b, d]	4[d]	5[b]	16[e]
1	5.13 *d*	5.11 *d*	5.12 *d*	5.11 *d*	5.11 *d*	5.07 *d*
	$J_{1,9}$ = 10	$J_{1,9}$ = 10	$J_{1,9}$ = 10	$J_{1,9}$ = 10	$J_{1,9}$ = 10	$J_{1,9}$ = 10
3	6.43 *dd*	6.41 *dd*	6.42 *dd*	6.39 *dd*	6.41 *dd*	6.35 *dd*
	$J_{3,4}$ = 6	$J_{3,4}$ = 6	$J_{3,4}$ = 6	$J_{3,4}$ = 6	$J_{3,4}$ = 6	$J_{3,4}$ = 6
	$J_{3,5}$ = 2	$J_{3,5}$ = 2	$J_{3,5}$ = 2	$J_{3,5}$ = 2	$J_{3,5}$ = 2	$J_{3,5}$ = 2
4	5.19 *dd*	5.10 *dd*	5.17 *dd*	5.14 *dd*	5.16 *dd*	5.05 *dd*
	$J_{4,3}$ = 6	$J_{4,3}$ = 6	$J_{4,3}$ = 6	$J_{4,3}$ = 6	$J_{4,3}$ = 6	$J_{4,3}$ = 6
	$J_{4,5}$ = 5	$J_{4,5}$ = 5	$J_{4,5}$ = 5	$J_{4,5}$ = 5	$J_{4,5}$ = 5	$J_{4,5}$ = 5
5	2.59 *m*	2.57 *m*	2.57 *m*	2.52 *m*	2.54 *m*	2.38 *m*
6	4.18 *dd*	4.13 *dd*	4.15 *dd*	4.10 *dd*	4.12 *dd*	3.99 *dd*
	$J_{6,5}$ = 8	$J_{6,5}$ = 8	$J_{6,5}$ = 8	$J_{6,5}$ = 8	$J_{6,5}$ = 8	$J_{6,5}$ = 8
	$J_{6,7}$ = 1	$J_{6,7}$ = 1	$J_{6,7}$ = 1	$J_{6,7}$ = 1	$J_{6,7}$ = 1	$J_{6,7}$ = 2
7	3.75 *bs*	3.72 *d*	3.74 *d*	3.68 *bs*	3.71 *bs*	3.62 *d*
		$J_{7,6}$ = 1	$J_{7,6}$ = 1			$J_{7,6}$ = 2
8	–	–	–	–	–	–
9	2.63 *dd*	2.61 *dd*	2.63 *dd*	2.59 *dd*	2.60 *dd*	2.54 *dd*
	$J_{9,1}$ = 10	$J_{9,1}$ = 10	$J_{9,1}$ = 10	$J_{9,1}$ = 10	$J_{9,1}$ = 10	$J_{9,1}$ = 10
	$J_{9,5}$ = 8	$J_{9,5}$ = 8	$J_{9,5}$ = 8	$J_{9,5}$ = 8	$J_{9,5}$ = 8	$J_{9,5}$ = 8
10A	3.83 *d*	3.82 *d*	3.83 *d*	3.81 *d*	3.82 *d*	3.81 *d*
	$J_{10A,10B}$ = 13	$J_{10A,10B}$ = 13	$J_{10A,10B}$ = 13	$J_{10A,10B}$ = 13	$J_{10A,10B}$ = 13	$J_{10A,10B}$ = 13
10B	4.18 *d*	4.16 *d*	4.17 *d*	4.15 *d*	4.16 *d*	4.13 *d*
	$J_{10B,10A}$ = 13	$J_{10B,10A}$ = 13	$J_{10B,10A}$ = 13	$J_{10B,10A}$ = 13	$J_{10B,10A}$ = 13	$J_{10B,10A}$ = 13
1′	4.78 *d*	4.77 *d*	4.78 *d*	4.75 *d*	4.78 *d*	4.77 *d*
	$J_{1',2'}$ = 8	$J_{1',2'}$ = 8	$J_{1',2'}$ = 8	$J_{1',2'}$ = 8	$J_{1',2'}$ = 8	$J_{1',2'}$ = 8
2′	3.27 *dd*	3.26 *dd*	3.27 *dd*	3.26 *dd*	3.26 *dd*	3.25 *dd*
	$J_{2',1'}$ = 8	$J_{2',1'}$ = 8	$J_{2',1'}$ = 8	$J_{2',1'}$ = 8	$J_{2',1'}$ = 8	$J_{2',1'}$ = 8
	$J_{2',3'}$ = 9	$J_{2',3'}$ = 9	$J_{2',3'}$ = 9	$J_{2',3'}$ = 9	$J_{2',3'}$ = 9	$J_{2',3'}$ = 9
3′	3.39 *t*	3.39 *t*	3.39 *t*	3.39 *t*	3.39 *t*	3.38 *t*
	J = 9	J = 9	J = 9	J = 9	J = 9	J = 9
4′	3.24 *dd*	3.24 *dd*	3.25 *dd*	3.24 *dd*	3.25 *dd*	3.24 *dd*
	$J_{4',3'}$ = 8	$J_{4',3'}$ = 8	$J_{4',3'}$ = 8	$J_{4',3'}$ = 8	$J_{4',3'}$ = 8	$J_{4',3'}$ = 8
	$J_{4',5'}$ = 10	$J_{4',5'}$ = 10	$J_{4',5'}$ = 10	$J_{4',5'}$ = 10	$J_{4',5'}$ = 10	$J_{4',5'}$ = 10
5′	3,3 *m*[f]	3,3 *m*[f]	3,3 *m*[f]	3,3 *m*[f]	3,3 *m*[f]	3,3 *m*[f]
6′A	3.62 *dd*	3.62 *dd*	3.62 *dd*	3.62 *dd*	3.62 *dd*	3.61 *dd*
	$J_{6'A,6'B}$ = 12	$J_{6'A,6'B}$ = 12	$J_{6'A,6'B}$ = 12	$J_{6'A,6'B}$ = 12	$J_{6'A,6'B}$ = 12	$J_{6'A,6'B}$ = 12
	$J_{6'A,5'}$ = 6	$J_{6'A,5'}$ = 6	$J_{6'A,5'}$ = 6	$J_{6'A,5'}$ = 6	$J_{6'A,5'}$ = 6	$J_{6'A,5'}$ = 6
6′B	3.91 *dd*	3.91 *dd*	3.91 *dd*	3.91 *dd*	3.91 *dd*	3.91 *dd*
	$J_{6'B,6'A}$ = 12	$J_{6'B,6'A}$ = 12	$J_{6'B,6'A}$ = 12	$J_{6'B,6'A}$ = 12	$J_{6'B,6'A}$ = 12	$J_{6'B,6'A}$ = 12
	$J_{6'B,5'}$ = 2	$J_{6'B,5'}$ = 2	$J_{6'B,5'}$ = 2	$J_{6'B,5'}$ = 2	$J_{6'B,5'}$ = 2	$J_{6'B,5'}$ =2
1″	5.32 *d*	5.27 *d*	5.28 *d*	5.16 *d*	5.19 *d*	4.92 *d*
	$J_{1'',2''}$ = 2	$J_{1'',2''}$ = 2	$J_{1'',2''}$ = 2	$J_{1'',2''}$ = 2	$J_{1'',2''}$ = 2	$J_{1'',2''}$ = 2
2″	5.72 *dd*	5.69 *dd*	5.70 *dd*	5.54 *dd*	5.61 *dd*	3.84 *dd*
	$J_{2'',1''}$ = 2	$J_{2'',1''}$ = 2	$J_{2'',1''}$ = 2	$J_{2'',1''}$ = 2	$J_{2'',1''}$ = 2	$J_{2'',1''}$ = 2
	$J_{2'',3''}$ = 3	$J_{2'',3''}$ = 3	$J_{2'',3''}$ = 3	$J_{2'',3''}$ = 3	$J_{2'',3''}$ = 3	$J_{2'',3''}$ = 3
3″	5.79 *dd*	5.57 *dd*	5.67 *dd*	5.32 *dd*	5.47 *dd*	3.67 *dd*
	$J_{3'',2''}$ = 3	$J_{3'',2''}$ = 3	$J_{3'',2''}$ = 3	$J_{3'',2''}$ = 3	$J_{3'',2''}$ = 3	$J_{3'',2''}$ = 3
	$J_{3'',4''}$ = 10	$J_{3'',4''}$ = 10	$J_{3'',4''}$ = 10	$J_{3'',4''}$ = 10	$J_{3'',4''}$ = 10	$J_{3'',4''}$ = 9
4″	5.64 *t*	5.46 *t*	5.51 *t*	3.76 *t*	3.84 *t*	3.38 *t*
	J = 10	J = 10	J = 10	J = 10	J = 10	J = 9
5″	4.35 *m*	4.15 *m*	4.25 *m*	3.95 *m*	3.99 *m*	3.63–3.69 *m*
6″ (3H)	1.35 *d*	1.31 *d*	1.32 *d*	1.38 *d*	1.41 *d*	1.25 *d*
	$J_{6'',5''}$ = 6	$J_{6'',5''}$ = 6	$J_{6'',5''}$ = 6	$J_{6'',5''}$ = 6	$J_{6'',5''}$ = 6	$J_{6'',5''}$ = 6
Acyl moiety A (R^1)						
2/6	8.08 *m*	8.05 *m*	8.06 *m*	8.05 *m*	8.02 *m*	
3/5	7.55 *m*	7.53 *m*	7.53 *m*	7.51 *m*	7.51 *m*	
4	7.68 *m*	7.66 *m*	7.66 *m*	7.65 *m*	7.64 *m*	
Acyl moiety B (R^2)						
2/6	7.74 *m*	7.82 *m*	7.80 *m*	7.27 *m*	7.89 *m*	
3/5	7.27 *m*	7.29 *m*	7.31 *m*	6.71 *m*	7.35 *m*	
4	7.46 *m*	7.49 *m*	7.49 *m*	–	7.53 *m*	

Table 1 (*continued*)

H	1[b, c]	2[b, d]	3[b, d]	4[d]	5[b]	16[e]
α				6.19 *d* $J_{\alpha,\beta}$ = 16		
β				7.50 *d* $J_{\beta,\alpha}$ = 16		
Acyl moiety C (R^3)						
2/6	7.95 *m*	7.48 *m*	7.36 *m*			
3/5	7.41 *m*	6.63 *m*	6.70 *m*			
4	7.55 *m*	–	–			
α		5.67 *d* $J_{\alpha,\beta}$ = 13	6.20 *d* $J_{\alpha,\beta}$ = 16			
β		6.83 *d* $J_{\beta,\alpha}$ = 13	7.57 *d* $J_{\beta,\alpha}$ = 16			

[a] In CD_3OD. Chemical shift values (δ) in ppm. Coupling constants (J) in Hz.
[b] Assignments were confirmed by $^1H^1H$ COSY.
[c] The positions of the acyl moieties were determined by $^{13}C^1H$ long-range correlation with delay times optimized for coupling constants of 8 Hz.
[d] The positions of the acyl moieties were determined by HMBC.
[e] Data taken from Helfrich and Rimpler (1999).
[f] Overlapped by the CD_2HOD signal.

2. Results

The ethanol–water extract of the aerial parts of *G. philippensis* was separated by a combination of column chromatography (CC) on celite and silica gel, centrifugal partition chromatography (CPC), and medium-pressure (MPLC) as well as high-pressure liquid chromatography (HPLC). The known isolated compounds were identified as 6-*O*-α-L-(3″-*O*-, 4″-*O*-dibenzoyl)-rhamnopyranosylcatalpol (**6**), 6-*O*-α-L-(2″-*O*-, 4″-*O*-dibenzoyl)rhamnopyranosylcatalpol (**7**) (Hosny and Rosazza, 1998), 6-*O*-α-L- (2″-*O*-*trans*-*p*-methoxycinnamoyl)rhamnopyranosylcatalpol (**8**), 6-*O*-α-L- (3″-*O*-*trans*-*p*-methoxycinnamoyl)rhamnopyranosylcatalpol (**9**) (Otsuka et al., 1991), and 6-*O*-α-L- (2″-*O*-*trans*-*p*-coumaroyl)rhamnopyranosylcatalpol (saccatoside) (**10**) (Otsuka et al., 1990) by comparison of the ^1H NMR and ^{13}C NMR spectra with the corresponding published data.

From different fractions of the isolation we identified the known iridoids catalpol (**11**), geniposidic acid (**12**), gardoside (**13**), and 8-*epi*-loganic acid (**14**) by GC and GC–MS.

The ^1H NMR (see Table 1) and ^{13}C NMR (see Table 2) spectra of compound **1** showed the characteristic signals of a 6-*O*-α-L-rhamnopyranosylcatalpol unit bearing three benzoyl moieties. The ESI-MS showed the expected quasimolecular ions at m/z 843 ($[M+Na]^+$) and m/z 859 ($[M+K]^+$), suggesting a molecular formula of $C_{42}H_{44}O_{17}$. The three acyl moieties were attached to the rhamnopyranosyl unit since the H-2″, H-3″ and the H-4″ signals were shifted downfield by 1.88, 2.12 and 2.26 ppm, respectively, compared to the unsubstituted 6-*O*-α-L-rhamnopyranosylcatalpol (**16**) (Helfrich and Rimpler, 1999). The ^1H signals and the corresponding ^{13}C NMR signals of the different benzoyl moieties were assigned to data sets A, B, and C by $^1H^1H$ COSY and $^{13}C^1H$ COSY. The positions of the respective benzoyl groups were determined by $^{13}C^1H$ long-range connectivities: the acyloxy group A was attached to C-2″ since its H-2 and H-6 signals were correlated to the ester carbonyl group at 166.86 ppm which in turn showed a long-range coupling to the H-2″ signal at 5.72 ppm. The ester carbonyl signal at 167.27 ppm was correlated to the H-2 and H-6 signals of the aromatic ring C as well as to the H-4″ signal at 5.64 ppm. The benzoyloxy group C, therefore, was attached to C-4″. Then the benzoyloxy group B must be attached to C-3″, although the expected long-range coupling of the remaining ester carbonyl signal at 166.76 ppm to H-3″ could not be detected. Thus, the structure of **1** was established as 6-*O*-α-L-(2″-*O*-, 3″-*O*-, 4″-*O*-tribenzoyl)rhamnopyranosylcatalpol.

The ESI mass spectra of compounds **2** and **3** showed the same quasimolecular ions at m/z 885 ($[M+Na]^+$) and m/z 901 ($[M+K]^+$), suggesting a molecular formula of $C_{44}H_{46}O_{18}$. The signals of the ^1H NMR spectra (see Table 1) and ^{13}C NMR spectra (see Table 2) were assigned to a basic skeleton of rhamnopyranosylcatalpol, two benzoyl moieties and one *p*-coumaroyl moiety. In **2** the double bond of the *p*-coumaroyl moiety was *cis*-configurated since the two doublets at δ 5.67 (1H, H-α) and δ 6.83 (1H, H-β) showed a coupling constant of 13 Hz whereas the H-α and H-β signals of **3** showed a coupling constant of 16 Hz indicating the *trans*-configuration of this double bond. In **2** the H-2″, H-3″ and H-4″ signals were shifted downfield by 1.85, 1.90 and 2.08 ppm, respectively, compared to **16**. Downfield shifts in the same range (1.86, 2.00 and 2.13 ppm) were also observed for the

H-2″, H-3″ and H-4″ signals of **3**. Therefore, the three acyl groups were linked to the rhamnopyranosyl moiety in both compounds. The ^{1}H signals and the corresponding ^{13}C NMR signals of the different benzoyl groups were assigned to data sets A and B for each compound by $^{1}H^{1}H$ COSY and $^{13}C^{1}H$ COSY. The positions of the corresponding benzoyl moieties as well as of the *p*-coumaroyl moieties were determined by HMBC: the ester carbonyl group at 167.00 ppm showed a long-range coupling to the H-β signal of the *cis*-*p*-coumaroyl moiety as well as to the H-4″-signal of **2**. A long-range coupling was also observed for the ester carbonyl group at 168.38 ppm to the H-β signal of the *trans*-*p*-coumaroyl moiety as well as to the H-4″-signal of **3**. Thus, the *p*-coumaroyl moiety was attached to *O*-4″ in **2** as well as in **3**. The H-2 and H-6 signals of the benzoyloxy groups A of both compounds were correlated to the corresponding ester carbonyl groups as well as to the H-2″ signals. The benzoyloxy group A of **2** as well as of **3**, therefore, was attached to C-2″. Then, the benzoyloxy groups B of both compounds must be connected to C-3″ although the expected long-range couplings of the corresponding carbonyl groups to H-3″ were not observable. Thus, the structures were determined as 6-*O*-α-L-(2″-*O*-, 3″-*O*-dibenzoyl, 4″-*O*-*cis*-*p*-coumaroyl)rhamnopyranosylcatalpol (**2**) and 6-*O*-α-L-(2″-*O*-, 3″-*O*-dibenzoyl, 4″-*O*-*trans*-*p*-coumaroyl)rhamnopyranosylcatalpol (**3**).

Table 2
^{13}C NMR spectral data[a] (125 MHz) of compounds **1**–**5** and **16**

C	1[b,c]	2[d,e]	3[d,e]	4[d,e]	5	16[f]
1	95.18	95.13	95.13	95.15	95.19	95.23
3	142.56	142.55	142.55	142.39	142.41	142.18
4	103.25	103.20	103.24	103.40	103.40	103.64
5	37.28	37.18	37.25	37.24	37.27	37.37
6	85.35	84.82	85.09	84.44	84.53	83.69
7	59.64	59.40	59.53	59.39	59.43	59.38
8	66.61	66.57	66.59	66.55	66.59	nd[h]
9	43.40	43.27	43.33	43.29	43.34	43.35
10	61.46	61.46	61.46	61.48	61.48	61.50
1′	99.75	99.70	99.70	99.71	99.74	99.77
2′	74.86	74.84	74.84	74.84	74.86	74.88
3′	77.72	77.68	77.68	77.69	77.72	77.76
4′	71.82	71.80	71.80	71.81	71.82	71.83
5′	78.69	78.69	78.69	78.67	78.68	78.65
6′	62.99	62.98	62.98	62.98	62.99	62.97
1″	98.05	97.73	97.92	97.73	97.80	100.38
2″	72.09	71.84[g]	71.80	72.16	72.08	72.28[g]
3″	71.56	71.80[g]	71.69	73.03	73.75	72.38[g]
4″	73.13	72.15	71.93	71.94	71.92	73.93
5″	68.29	68.13	68.36	70.33	70.38	70.19
6″	17.91	17.85	17.88	18.12	18.10	17.94
Acyl moiety A (R^1)						
1	130.68	130.67	130.67	130.72	130.73	
2/6	130.79	130.80	130.80	130.78	130.78	
3/5	129.86	129.84	129.84	129.78	129.77	
4	134.86	134.84	134.84	134.71	134.73	
C=O	166.86	167.00	166.84	166.92	166.91	
Acyl moiety B (R^2)						
1	130.38	130.51	130.51	126.45	130.47	
2/6	130.50	130.63	130.58	131.21	130.61	
3/5	129.43	129.53	129.49	117.14	129.40	
4	134.50	134.56	134.53	162.50	134.30	
C=O	166.76	166.84	166.82	168.46	167.30	
α				114.21		
β				147.24		
Acyl moiety C (R^3)						
1	130.47	126.97	126.09			
2/6	130.64	133.78	131.46			
3/5	129.67	116.06	117.31			
4	134.69	161.12	163.14			
C=O	167.27	167.00	168.38			
α		114.91	113.20			
β		147.25	148.15			

[a] In CD_3OD. Chemical shift values (δ) in ppm.
[b] Assignments were confirmed by $^{13}C^{1}H$ COSY.
[c] The positions of the acyl moieties were determined by $^{13}C^{1}H$ long-range correlation with delay times optimized for coupling constants of 8 Hz.
[d] Assignments were confirmed by HMQC.
[e] The positions of the acyl moieties were determined by HMBC.
[f] Data taken from Helfrich and Rimpler (1999).
[g] Values in each column are interchangeable.
[h] nd: signal was not detectable.

The ESI mass spectrum (ESI-MS) of compound **4** showed two quasimolecular ions at *m*/*z* 781 ($[M+Na]^+$) and *m*/*z* 797 ($[M+K]^+$), suggesting a molecular formula of $C_{37}H_{42}O_{17}$. The ^{1}H NMR data (see Table 1) and ^{13}C NMR data (see Table 2) indicated a rhamnopyranosylcatalpol moiety acylated with one benzoyl and one *p*-coumaroyl moiety. The two doublets of the *p*-coumaroyl moiety at δ 6.19 (1 H, H-α) and δ 7.50 (1 H, H-β) with a coupling constant of 16 Hz demonstrated the *trans*-configuration of the double bond. The acyloxy moieties were attached to the C-2″ and C-3″ since the H-2″ and the H-3″ signals were shifted downfield by 1.70 and 1.65 ppm, respectively, compared to **16**. The positions of the acyl units were determined by HMBC: the benzoyloxy group was linked to C-2″ since its H-2 and H-6 signals (δ 8.05) showed a long-range coupling to the ester carbonyl signal at 166.92 ppm which in turn was correlated to the H-2″ signal at 5.54 ppm. The ester carbonyl signal at 168.46 ppm showed the expected long-range coupling to the H-β signal of the *trans*-*p*-coumaroyl moiety as well as the H-3″ signal. Thus, compound **4** was identified as 6-*O*-α-L-(2″-*O*-benzoyl, 3″-*O*-*trans*-*p*-coumaroyl)rhamnopyranosylcatalpol.

The ^{1}H NMR (see Table 1) and ^{13}C NMR (see Table 2) spectra of compound **5** exhibited the signals of a rhamnopyranosylcatalpol moiety as well as of two benzoyl moieties. The ESI-MS showed the expected quasimolecular ions at *m*/*z* 739 ($[M+Na]^+$) and *m*/*z* 755 ($[M+K]^+$), corresponding to a molecular formula of $C_{35}H_{40}O_{16}$. The benzoyloxy moieties were linked to C-2″ and C-3″ since the H-2″ and H-3″ signals showed

the characteristic downfield shifts of 1.77 and 1.80 ppm, respectively, compared to **16**. The ^{1}H signals of the different benzoyl groups were assigned to data sets A and B by ^{1}H^{1}H COSY. The positions of the corresponding benzoyl moieties were determined by comparison of the proton shifts with those of the aromatic protons of **1**: the proton signal with the highest chemical shift was observed for H-2/H-6 of the aromatic ring A (δ 8.08), whereas the H-2/H-6 signals of the aromatic rings B and C showed significantly lower chemical shifts. The H-2/H-6 signal of the benzoyl group A of **5** was observed at 8.02 ppm, indicating the attachment of this benzoyloxy group at C-2″. Then, the benzoyloxy group B must be attached to the C-3″. Thus, compound **5** was identified as 6-*O*-α-L-(2″-*O*-, 3″-*O*-dibenzoyl)rhamnopyranosylcatalpol.

The ^{1}H NMR and ^{13}C NMR data of compound **15** suggested a basic skeleton of 8-*epi*-loganic acid esterified with a second β-D-glucopyranosyl moiety. The ESI-MS showed the expected quasimolecular ions at m/z 561 ($[M+Na]^+$) and m/z 577 ($[M+K]^+$), corresponding to M = $C_{22}H_{34}O_{15}$. The downfield shift of H-1″ (0.86 ppm) compared to H-1′ proved the ester linkage between 8-*epi*-loganic acid and the second glucopyranosyl moiety. In addition, our ^{13}C data of both glucosyl moieties recorded in D_2O were identical with those of asystasioside A (Demuth et al., 1989). Thus, compound **15** was identified as 1-*O*-(8-*epi*-loganoyl)-β-D-glucopyranose. We propose the name gmephiloside for this new compound.

3. Discussion

Gmelina philippensis accumulates a range of rhamnopyranosylcatalpol esters like *G. arborea* (Hosny and Rosazza, 1998). Additionally, *G. philippensis* contains some iridoid acids and an iridoid acid glucosyl ester.

Within Lamiaceae iridoid acids have also been found in species of *Premna* (e.g. Bheemasankara Rao et al., 1981), *Vitex* (e.g. Sehgal et al., 1983), and in several other genera, e.g. in *Holmskioldia* (Helfrich, 2000), *Scutellaria* (e.g. Çalis et al., 1993a), *Clerodendrum* (e.g. Çalis et al., 1994), *Physostegia* (e.g. Nass and Rimpler, 1996), *Prostanthera* and *Pogostemon* (Schmidt, 1997). Thus, the occurrence of these compounds seems to be a less useful character for taxonomic studies within Lamiaceae.

Although rhamnopyranosylcatalpol esters, obviously, evolved several times independently they may be a more useful taxonomic character because of their restricted occurrence in only a few genera of the Lamiaceae and the related families Buddlejaceae (Miyase et al., 1991) and Scrophulariaceae (e.g. Çalis et al., 1988, 1993b; Warashina et al., 1991; Kalpoutzakis et al., 1999; Miyase and Mimatsu, 1999). Within Lamiaceae acylated rhamnopyranosylcatalpol derivatives have only been isolated from *Gmelina* species, several *Premna* species (e.g. Otsuka et al., 1989, 1990, 1991) and *Holmskioldia sanguinea* (Helfrich and Rimpler, 1999).

The accumulation of these compounds in both investigated *Gmelina* species and in several *Premna* species supports a close relationship between these two genera as well as the segregation of both genera from *Vitex*, where no rhamnopyranosylcatalpol derivatives have been found (e.g. Hänsel et al., 1965; Rimpler, 1972a, 1972b; Sehgal et al., 1983; Görler et al., 1985; Kouno et al., 1988; Iwagawa et al., 1993; Suksamran et al., 1999). However, the fact that rhamnopyranosylcatalpol esters are also accumulated in *Holmskioldia* (Scutellarioideae) favours a position of the clade consisting of *Gmelina* and *Premna* as sister group to the Scutellarioideae rather than the weakly-supported position as sister group to the whole clade including Scutellarioideae, Lamioideae and Pogostemonoideae as inferred from cpDNA sequences by Olmstead et al. (1998) and Wagstaff et al. (1998).

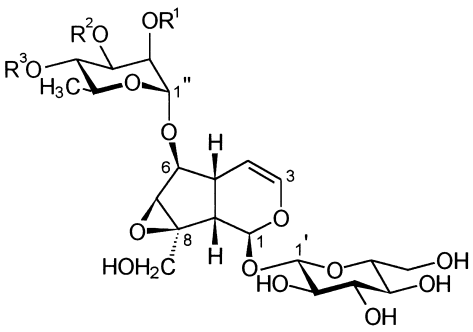
4. Experimental

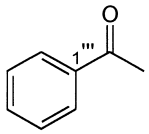
4.1. Plant material

Gmelina philippensis was cultivated in the Botanical Garden, Freiburg. The aerial parts of the plant were collected in the flowering period. A voucher specimen (1271) has been deposited at the herbarium of the Institut für Pharmazeutische Biologie, Freiburg.

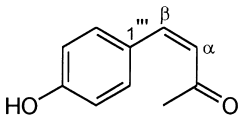
4.2. General

NMR: 500 MHz (^{1}H) or 125 MHz (^{13}C), chemical shifts as δ values (ppm) relative to the CD_2HOD signal at 3.30 ppm (^{1}H) and 49.0 ppm (^{13}C), CD_3OD as solvent. ESI-MS: positive ion mode, 3.8 kV, capillary 250°C, sheath gas 30 psi. GC–MS: EI 70 eV, TMSi derivative, He as carrier gas (6 psi, split vent flow of 10 ml min^{-1}); WCOT Rtx®-200 (trifluoropropylmethyl polysiloxane, 30 m × 0.32 mm), film thickness 0.25 μm; operation conditions: 220°C for 10 min, then 3°C min^{-1} to 290°C. GC: same column and operation conditions; guard column (5 m × 0.32 mm); linear velocity 19 cm min^{-1} (set at 220°C), split ratio 85:1. TLC: silica gel 60 F_{254}, CH_2Cl_2–MeOH–H_2O (70:30:3) and RP-18 MeCN–MeOH–H_2O (25.0:7.7:67.3); spray reagent vanillin (3%) and H_2SO_4 (1%) in EtOH followed by heating at 110° for 5–10 min. CC: silica gel 60, 63–200 μm (Merck); celite 545 (Johns–Manville). HPLC: LiChrospher 100 C-18 (10 μm; 8 × 250 mm), flow 2 ml min^{-1}, pressure 430–480 psi; photodiode array detector, Waters 996, 193–400 nm; solvent systems were

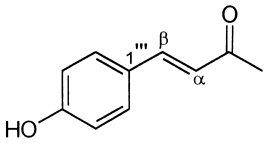




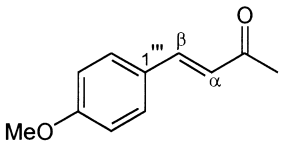
benz



c-coum

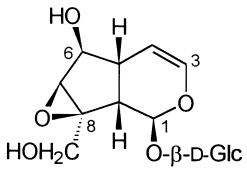


t-coum



t-moc

	R^1 (A)	R^2 (B)	R^3 (C)
1	benz	benz	benz
2	benz	benz	*c*-coum
3	benz	benz	*t*-coum
4	benz	*t*-coum	H
5	benz	benz	H
6	H	benz	benz
7	benz	H	benz
8	*t*-moc	H	H
9	H	*t*-moc	H
10	*t*-coum	H	H
16	H	H	H



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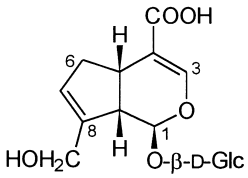
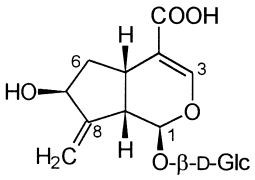
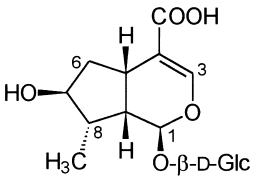


12



13



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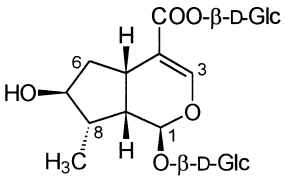

15

optimized with OPTISOLV (Geiger and Rimpler, 1990). MPLC: column I: LiChroprep C-18 (15–25 μm; 18.5 × 480 mm), flow 3 ml min^{-1}, pressure 50–80 psi; column II: LiChroprep C-18 (15–25 μm; 26 × 500 mm), flow 4.5 ml min^{-1}, pressure 50–80 psi; frs. were monitored by UV detection (Uvicord VW 2251, 230 nm). VLC: Eurochrom Bioselect C-18 (25–40 μm; 60 × 230 mm), flow 10 ml min^{-1}, pressure ca 2.5 psi. CPC: ITO multi-layer coil separator–extractor (capacity 380 ml; i.d. 2.6 mm; 700 rpm), $CHCl_3$–MeOH–*iso*-PrOH–H_2O (5:6:1:4) in the descending mode (normal elution mode, NEM) and ascending mode (reversed elution mode, REM), flow 4 ml min^{-1}; frs. were monitored by TLC: silica gel 60 F_{254}, organic layer of $CHCl_3$–MeOH–*iso*-PrOH–H_2O (5:6:1:4); $CHCl_3$ was purified by CC on basic Al_2O_3. Preparation for lyophilization: to minimize transacylations frs. obtained by HPLC and MPLC were diluted with H_2O without removing the organic solvents; frs. obtained by CPC were diluted with H_2O after removal of $CHCl_3$. After dilution the frs. were immediately frozen in liquid N_2 and subsequently lyophilized.

4.3. Isolation

The lyophilized powdered aerial parts (150 g) were extracted by refluxing for 30 min successively with 96%, 80%, 70% and 50% EtOH. The combined

extracts were conc. in vacuo and subsequently lyophilized providing 59 g of crude extract. This extract was partitioned into a lipophilic and hydrophilic fr. (30 g) by CC on Celite with CH_2Cl_2–*n*-Hexan (1:1) and CH_2Cl_2–MeOH (1:1). After freeze-drying 15 g of the hydrophilic fr. were separated by CC on silica gel with CH_2Cl_2–MeOH–H_2O mixtures of increasing polarity [(70:30:3) → (50:50:5)] providing 8 frs. (I–VIII). Frs. I–V were eluted with mixture 70:30:3, frs. VI–VII with mixture 60:40:4 and fr. VIII with mixture 50:50:5. The following yields were obtained after lyophilization: fr. I 0.05 g, fr. II 0.92 g, fr. III 2.29 g, fr. IV 3.52 g, fr. V 2.18 g, fr. VI 2.21 g, fr. VII 1.17 g, and fr. VIII 0.82 g. TLC as well as GC-analysis indicated the presence of iridoids in all frs. except for fr. I. Fr. II was separated (x2 450 mg) by CPC into eight subfractions; frs. II.1–II.4 were obtained by REM, frs. II.5–II.8 by NEM. Purification of fr. II.6 (147 mg) by MPLC (column II) with MeCN–MeOH–H_2O (56.2:7.7:36.1) gave compound **1** (46 mg). Fr. II.8 (54 mg) was separated by MPLC (column I) with MeCN–MeOH–H_2O (43.8:9.5:46.7) providing compounds **2** (1 mg) and **3** (1 mg). HPLC analysis showed that each compound was contaminated with the other. Therefore, the structure elucidation was carried out with **3** containing **2** in a ratio of 4:1. MPLC (column II) of fr. II.2 (123 mg) with MeCN–MeOH–H_2O (37.2:8.1:54.7) gave compound **4** (2 mg). Separation of fr. II.3 (86 mg) by MPLC (column I) with MeCN–MeOH–H_2O (37.2:8.1:54.7) afforded compound **5** (3 mg) and a mixture of **5**, **6** and **7** (5 mg) in a ratio of 1:5:1.5. The subfractions III.5 (320 mg) and III.10 (20 mg) were obtained by separation of fr. III (×3 500 mg) by CPC (NEM). Purification of fr. III.5 by CPC (NEM) gave fr. III.5.2 (140 mg) which was separated by MPLC (column II) with MeCN–-*iso*-PrOH–H_2O (26.6:3.6:69.8) affording a mixture of **8** and **9** (23 mg) in a ratio of 2:5. Compound **10** (2 mg) was obtained by prep. HPLC of fr. III.10 with MeCN–MeOH–H_2O (20.0:6.2:73.8). 500 mg of fr. VI were separated by MPLC with MeCN–MeOH–H_2O (8.4:1.2:90.4) providing **15** (7 mg). Since the isolated amounts of some compounds were too small for structure elucidation 14 g of the hydrophilic fr. were separated in a second isolation procedure providing compounds **2** (2 mg), **3** (5 mg), **4** (7 mg), and the mixture of **6** and **7** (10 mg). The frs. IV–VIII (500 mg of each fr.) were separated by CPC (NEM); the CPC subfractions were investigated by GC–MS and GC co-chromatography with authentic samples. Compound **11** was detected in frs. IV.8 and V.8, **12** in fr. V.7 and **13** in fr. VI.5. Compound **14** was identified in subfractions of the frs. III–VIII. Therefore, the isolation procedure was modified for a quantitative analysis of **11**–**14**.

4.4. Modified extraction

20 g of the lyophilized powdered aerial parts were extracted by refluxing for 30 min successively with 96%, 80%, 70% and 50% EtOH. The combined extracts were conc. in vacuo and partitioned between H_2O and $CHCl_3$. The aq. layer after conc. in vacuo (10 ml) was separated by FC with a H_2O–MeOH gradient (H_2O – MeOH10% – MeOH30% – MeOH50% – MeOH100%). GC–MS and GC co-chromatography with an authentic sample proved the presence of **11** (0.03%) only in the 10% MeOH-fr. (284 mg) as well as of **12** (0.01%) and **13** (0.03%) in the 30% MeOH-fr. (290 mg). Compound **14** (0.07%) was detected in the 30% MeOH-fr. and in the 50% MeOH-fr. (540 mg). The yields given in parenthesis refer to lyophilized plant material. They were quantified by GC with harpagide as internal standard. Salts of the iridoid acids were not detectable in the H_2O-fr (1640 mg).

4.4.1. 6-O-α-L-(2″-O-, 3″-O-, 4″-O-Tribenzoyl)rhamnopyranosylcatalpol (*1*)

White amorphous powder. $[\alpha]_D^{22} - 56^\circ$ (MeOH; *c* 0.11).

4.4.2. 6-O-α-L-(3″-O-, 4″-O-Dibenzoyl)rhamnopyranosylcatalpol (*6*) and 6-O-α-L-(2″-O-, 4″-O-dibenzoyl)rhamnopyranosylcatalpol (*7*)

Pale yellow powder. ESI-MS, *m/z*: 755 $[M+K]^+$, 739 $[M+Na]^+$. ^{13}C NMR: Our data of **6** and **7** were identical with published data (Hosny and Rosazza, 1998). ^1H NMR: our data of **6** as well as of the rhamnopyranosyl moiety and of the acyl moieties of **7** are given here because they differed significantly from published data (Hosny and Rosazza, 1998). (**6**, **7**): δ 1.25 (3 H, *d*, *J* = 6 Hz, **7**: H-6″), 1.27 (3 H, *d*, *J* = 6 Hz, **6**: H-6″), 2.53 (2 H, *m*, **6**, **7**: H-5), 2.60 (1 H, *dd*, *J* = 10, 8 Hz, **6**: H-9), 3.25 (2 H, *dd*, *J* = 10, 8 Hz, **6**, **7**: H-4′), 3.27 (2 H, *dd*, *J* = 9, 8 Hz, **6**, **7**: H-2′), 3.30 (**6**, **7**: H-5′, overlapped by the CD_2HOD signal), 3.40 (1 H, *t*, *J* = 9 Hz, **6**: H-3′), 3.63 (1 H, *dd*, *J* = 12, 6 Hz, **6**: H-6′A), 3.71 (1 H, *br s*, **6**: H-7), 3.83 (1 H, *d*, *J* = 13 Hz, **6**: H-10A), 3.92 (1 H, *dd*, *J* = 12, 2 Hz, **6**: H-6′B), 4.11 (1 H, *m*, **7**: H-5″), 4.11 (1 H, *dd*, *J* = 8, 2 Hz, **6**: H-6), 4.16 (1 H, *d*, *J* = 13 Hz, **6**: H-10B), 4.16 (1 H, *m*, **6**: H-5″), 4.27 (1 H, *dd*, *J* = 3, 2 Hz, **6**: H-2″), 4.35 (1 H, *dd*, *J* = 10, 3 Hz, **7**: H-3″), 4.78 (1 H, *d*, *J* = 8 Hz, **6**: H-1′), 5.09 (1 H, *d*, *J* = 2 Hz, **6**: H-1″), 5.12 (1 H, *d*, *J* = 10 Hz, **6**: H-1), 5.16 (1 H, *dd*, *J* = 6, 5 Hz, **6**: H-4), 5.18 (1 H, *d*, *J* = 2 Hz, **7**: H-1″), 5.36 (1 H, *t*, *J* = 10 Hz, **7**: H-4″), 5.39 (1 H, *dd*, *J* = 3, 2 Hz, **7**: H-2″), 5.52 (1 H, *dd*, *J* = 10, 3 Hz, **6**: H-3″), 5.59 (1 H, *t*, *J* = 10 Hz, **6**: H-4″), 6.41 (1 H, *dd*, *J* = 6, 2 Hz, **6**: H-3), 7.36 (2 H, *m*, $\mathbf{6}_B$: H-3/5) 7.39 (2 H, *m*, $\mathbf{6}_C$: H-3/5), 7.51 (2 H, *m*, $\mathbf{7}_C$: H-3/5), 7.52 (2 H, *m*, $\mathbf{6}_B$: H-4; $\mathbf{6}_C$: H-4), 7.53 (2 H, *m*, $\mathbf{7}_A$: H-3/5), 7.62 (1

H, *m*, $\mathbf{7}_C$: H-4), 7.65 (1 H, *m*, $\mathbf{7}_A$: H-4), 7.92 (2 H, *m*, $\mathbf{6}_B$: H-2/6), 7.93 (2 H, *m*, $\mathbf{6}_C$: H-2/6), 8.08 (2 H, *m*, $\mathbf{7}_C$: H-2/6), 8.13 (2 H, *m*, $\mathbf{7}_A$: H-2/6). Assignments were confirmed by ^{1}H–^{1}H COSY, HMQC and HMBC.

*4.4.3. Gmephiloside (**15**)*

White amorphous powder. ^{1}H NMR: δ 1.04 (3 H, *d*, J = 8 Hz, H-10), 1.88 (1 H, *dt*, J = 14, 5 Hz, H-6α), 2.08 (1 H, *ddd*, J = 14, 9, 5 Hz, H-6β), 2.13 (1 H, *m*, H-8), 2.62 (1 H, *dt*, J = 9, 4 Hz, H-9), 3.06 (1 H, *m*, H-5), 3.18 (1 H, *dd*, J = 9, 8 Hz, H-2′), 3.24 (1 H, *dd*, J = 10, 9 Hz, H-4′), 3.30 (H-5′, overlapped by the CD_2HOD signal), 3.32–3.38 (4 H, *m*, H-2″, H-3″, H-4″, H-5″), 3.42 (1 H, *t*, J = 9 Hz, H-3′), 3.64 (1 H, *dd*, J = 12, 6 Hz, H-6′A), 3.68 (1 H, *dd*, J = 12, 6 Hz, H-6″A), 3.81 (1 H, *m*, H-7), 3.83 (1 H, *dd*, J = 12, 2 Hz, H-6″B), 3.90 (1 H, *dd*, J = 12, 2 Hz, H-6′B), 4.64 (1 H, *d*, J = 8 Hz, H-1′), 5.51 (1 H, *d*, J = 8 Hz, H-1″), 5.54 (1 H, *d*, J = 4 Hz, H-1), 7.54 (1 H, *d*, J = 1 Hz, H-3). ^{13}C NMR: δ 14.35, C-10; 30.85, C-5; 40.93, C-6; 42.95, C-9; 45.12, C-8; 62.29, C-6″; 62.93, C-6′; 71.03, C-4″; 71.70, C-4′; 74.00, C-2″; 74.73, C-2′; 78.02*, C-3″; 78.04*, C-3′; 78.45, C-5′; 78.76, C-5″; 79.32, C-7; 95.42, C-1″; 96.29, C-1; 99.73, C-1′; 113.52, C-4; 153.95, C-3; 167.26, C-11. Assignments were confirmed by ^{1}H–^{1}H COSY and ^{13}C–^{1}H COSY (∗ Data are interchangeable).

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References

Bheemasankara Rao, Ch., Vijayakumar, E.K.S., Vijayalakshmi, K.V., 1981. Iridoids from *Premna latifolia*. Planta Medica 41, 80–83.

Briquet, J., 1885. Engler, A., Prantl, K. (Eds.), Die natürlichen Pflanzenfamilien, vol. IV.3a. W. Engelmann, Leipzig, pp. 132–182.

Çalis, I., Ersöz, T., Saracoglu, I., Sticher, O., 1993a. Scalbidoside and albidoside, two iridoid glycosides from *Scutellaria albida* subsp. *colchica*. Phytochemistry 32 (5), 1213–1217.

Çalis, I., Hosny, M., Yürüker, A., 1994. Inerminosides A1, C and D, three iridoid glycosides from *Clerodendrum inerme*. Phytochemistry 37 (4), 1083–1085.

Çalis, I., Gross, G.-A., Winkler, T., Sticher, O., 1988. Isolation and structure elucidation of two highly acylated iridoid diglycosides from *Scrophularia scopolii*. Planta Medica 54, 168–170.

Çalis, I., Zor, M., Basaran, A.A., Wright, A.D., Sticher, O., 1993b. Karsoside and scropolioside D, two new iridoid glycosides from *Scrophularia ilwensis*. Journal of Natural Products 56 (4), 606–609.

Cantino, P.D., 1992. Evidence for a polyphyletic origin of the Labiatae. Annals of the Missouri Botanical Garden 79, 361–379.

Cantino, P.D., Harley, R.M., Wagstaff, S.J., 1992. In: Harley, R.M., Reynolds, T. (Eds.), Advances in labiate science. Royal Botanic Gardens, Kew, pp. 511–522.

Demuth, H., Jensen, S.R., Nielsen, B.J., 1989. Iridoid glucosides from *Asystasia bella*. Phytochemistry 28 (12), 3361–3364.

Geiger, C., Rimpler, H., 1990. "OPTISOLV" A PC-Program for solvent optimization in liquid chromatography. GIT Fachzeitschrift Labor, Part 1: 11, 1391–1397; Part 2: 12, 1496–1498.

Görler, K., Oehlke, D., Soicke, H., 1985. Iridoidführung von *Vitex agnus-castus*. Planta Medica, 530–531.

Hänsel, R., Leuckert, Ch., Rimpler, H., Schaaf, K.D., 1965. Chemotaxonomische Untersuchungen in der Gattung *Vitex* L. Phytochemistry 4, 19–27.

Hegnauer, R., Kooiman, P., 1978. The taxonomic significance of iridoids of Tubiflorae sensu Wettstein. Planta Medica 33 (1), 1–33.

Helfrich, E., Rimpler, H., 1999. Iridoid glycosides and phenolic glycosides from *Holmskioldia sanguinea*. Phytochemistry 50 (4), 619–627.

Helfrich, E., 2000. Ph.D. thesis. Iridoidglykoside aus *Holmskioldia sanguinea* und *Gmelina philippensis* und deren Bedeutung als taxonomische Merkmale innerhalb der Lamiaceae s.l. Albert-Ludwigs-Universität, Freiburg (in preparation).

Hosny, M., Rosazza, J.P.N., 1998. Gmelinosides A-L, 12 acylated iridoid glycosides from *Gmelina arborea*. Journal of Natural Products 61 (6), 734–742.

Iwagawa, T., Nakahara, A., Nakatani, M., 1993. Iridoids from *Vitex cannabifolia*. Phytochemistry 32 (2), 453–454.

Kalpoutzakis, E., Aligiannis, N., Mitakou, S., Skaltsounis, A.-L., 1999. Verbaspinoside, a new iridoid glycoside from *Verbascum spinosum*. Journal of Natural Products 62 (2), 342–344.

Kouno, I., Inoue, M., Onizuka, Y., Fujisaki, T., Kawano, N., 1988. Iridoid and phenolic glucosides from *Vitex rotundifolia*. Phytochemistry 27 (2), 611–612.

Miyase, T., Akahori, C., Kohsaka, H., Ueno, A., 1991. Acylated iridoid glycosides from *Buddleja japonica* Hemsl. Chemical and Pharmaceutical Bulletin 39 (11), 2944–2951.

Miyase, T., Mimatsu, A., 1999. Acylated iridoid and phenylethanoid glycosides from the aerial parts of *Scrophularia nodosa*. Journal of Natural Products 62 (8), 1079–1084.

Moldenke, H.N., 1989. A fifth summary of the Verbenaceae, Avicenniacea, Stilbaceae, Dicrastylidaceae, Symphoremaceae, Nyctanthaceae, and Eriocaulaceae of the World as to valid taxa, geographic distribution, and synonomy, vol. 1–2. Published by the author, Wayne, New Jersey.

Nass, R., Rimpler, H., 1996. Distribution of iridoids in different populations of *Physostegia virginiana* and some remarks on iridoids from *Avicennia officinalis* and *Scrophularia ningpoensis*. Phytochemistry 41 (2), 489–498.

Olmstead, R.G., Reeves, P.A., Lepschi, B.J., 1998. Confirmation of a monophyletic Chloanthoideae (Lamiaceae) comprising tribes Chloantheae and Prostanthereae. Lamiales Newsletter 6, 7–10.

Otsuka, H., Kubo, N., Yamasaki, K., Padolina, W.G., 1989. Two

iridoid glycoside caffeoyl esters from *Premna odorata*. Phytochemistry 28 (2), 513–515.

Otsuka, H., Sasaki, Y., Yamasaki, K., Takeda, Y., Seki, T., 1990. Iridoid diglycoside monoacyl esters from the leaves of *Premna japonica*. Journal of Natural Products 53 (1), 107–111.

Otsuka, H., Sasaki, Y., Kubo, N., Yamasaki, K., Takeda, Y., Seki, T., 1991. Isolation and structure elucidation of mono- and diacyl iridoid diglycosides from leaves of *Premna japonica*. Journal of Natural Products 54 (2), 547–553.

Otsuka, H., Kashima, N., Hayashi, T., Kubo, N., Yamasaki, K., Padolina, W.G., 1992. Premnaodorosides A, B, and C, iridoid glucoside diesters of an acyclic monoterpenediol from leaves of *Premna odorata*. Phytochemistry 31 (9), 3129–3133.

Rimpler, H., 1972a. Phytoecdysones and iridoids from *Vitex megapotamica*. Archiv der Pharmazie 305 (10), 746–751.

Rimpler, H., 1972b. Iridoids and ecdysones from *Vitex* species. Phytochemistry 11, 2653–2654.

Schmidt, E.-M., 1997. Ph.D. thesis. Verwandtschaftsbeziehungen bei Lamiaceae: Eine kladistische Analyse der Verbreitung von Iridoiden und von morphologischen Merkmalen. Albert-Ludwigs-Universität, Freiburg.

Sehgal, C.K., Taneja, S.C., Dhar, K.L., Atal, C.K., 1983. 6′-*p*-hydroxybenzoylmussaenosidic acid-an iridoid glucoside from *Vitex negundo*. Phytochemistry 22 (4), 1036–1038.

Sudo, H., Ide, T., Otsuka, H., Hirata, E., Takushi, A., Takeda, Y., 1997. 10-*O*-acylated iridoid glucosides from leaves of *Premna subscandens*. Phytochemistry 46 (7), 1231–1236.

Sudo, H., Ide, T., Otsuka, H., Hirata, E., Takushi, A., Takeda, Y., 1998. Iridoid glucosides with different acyl moieties from globularinin and globularimin from leaves of *Premna subscandens*. Phytochemistry 49 (3), 783–786.

Suksamran, S., Kumcharoen, S., Suksamran, A., 1999. Iridoids from *Vitex limonifolia*. Planta Medica 65, 392.

Wagstaff, S.J., Hickerson, L., Spangler, R., Reeves, P.A., Olmstead, R.G., 1998. Phylogeny in Labiatae s.l., inferred from cpDNA sequences. Plant Systematics and Evolution 209, 265–274.

Warashina, T., Miyase, T., Ueno, A., 1991. Iridoid glycosides from *Verbascum thapsus* L. Chemical and Pharmaceutical Bulletin 39 (12), 3261–3264.