PERGAMON

Phytochemistry 54 (2000) 959–964

PHYTOCHEMISTRY

www.elsevier.com/locate/phytochem

Alkaloids from *Isopyrum thalictroides* L.[☆]

Ralitsa S. Istatkova, Stefan A. Philipov*

Institute of Organic Chemistry with Center of Phytochemistry, Bulgarian Academy of Sciences, Acad. G. Bonchev Street, Building 9, 1113 Sofia, Bulgaria

Received 7 September 1999; received in revised form 8 February 2000

Abstract

Three new bisbenzylisoquinoline alkaloids, isopyruthaldine, isopythaldine and isothalmidine, containing new type of bridges were isolated from roots and rhizomes of *Isopyrum thalictroides*. The known isoquinoline alkaloids, reticuline, isocorydine, columbamine and palmatine, obtained from the aerial parts, are new for the genus *Isopyrum*. All structures were elucidated by spectroscopic analysis. © 2000 Elsevier Science Ltd. All rights reserved.

Keywords: Isopyrum thalictroides; Ranunculaceae; Bisbenzylisoquinoline alkaloids; Isopyruthaldine; Isopythaldine; Isothalmidine

1. Introduction

In a previous report (Philipov and Istatkova, 1997, 1999) we presented the isolation and the structural elucidation of five new and one known bisbenzylisoquinoline alkaloids from roots and rhizomes of *Isopyrum thalictroides* (Ranunculaceae), growing in Bulgaria. Further investigations on roots and rhizomes, and aerial parts of these species comprised, in addition, several new and known isoquinoline alkaloids. In the present phytochemical study, we report on the isolation and the structural elucidation of three new bisbenzylisoquinoline alkaloids, named isopyruthaldine (**1**), isopythaldine (**2**) and isothalmidine (**3**) from roots and rhizomes of *Isopyrum thalictroides*. The known isoquinoline alkaloids reticuline, isocorydine, columbamine and palmatine, obtained from the aerial parts, were found for the first time in the genus *Isopyrum*. The structures of the three new alkaloids were established using of ^{1}H and ^{13}C NMR spectra, DEPT experiment, 2D techniques COSY, NOESY, HMQC and HMBC, as well as by the UV, IR, MS and $[\alpha]_D$ data.

2. Results and discussion

Isopyruthaldine (**1**) was isolated as a yellow amorphous solid. The CI mass spectrum showed a peak at *m*/*z* 769 $[M + H]^+$, which correspond to the molecular formula $C_{44}H_{52}N_2O_{10}$. The base peak in the EI mass spectrum at *m*/*z* 236 represented the rings A and B. The peak at *m*/*z* 533 due to the loss of the rings A and B from the molecule was also observed. These data, as well as the weak molecular ion at *m*/*z* 768, suggested the structure of a bisbenzylisoquinoline alkaloid. The ^{1}H NMR spectrum of **1** exhibited singlets for seven aromatic protons, sharp singlets for nine OCH_3 groups, and a three-proton singlet for one NCH_3 group (see Table 1). The lack of second signal for NCH_3 group and the presence of an absorption for NH at 3390 cm^{-1} in the IR spectrum proved that in one of the benzylisoquinoline moiety of the molecule the nitrogen atom was secondary. The most downfield signal in the spectrum at δ 10.68 was for a CHO group. A confirmation for the presence of a formyl function was the absorption at 1721 cm^{-1} in the IR

[☆] Part 3 in the series "Alkaloids from *Isopyrum thalictroides*". For Part 2 see Philipov and Istatkova (1999).

* Corresponding author. Tel.: +359-2-7334244; fax: +359-2-700-225.

E-mail address: philipov@orgchm.bas.bg (S.A. Philipov).

PII: S0031-9422(00)00068-6

spectrum. In the same spectrum, an intramolecular H-bond at 3452 cm^{-1} (CCl$_4$; c 1.0×10^{-3} M/l) between NH and CHO groups was observed, which indicated that these groups were located closely and were on the same side of the molecule. The data based on the ^{13}C NMR spectrum (Table 1) and on the DEPT experiment showed the presence of five upfield secondary carbons, corresponding to C-3, C-4, C-3′, C-4′ and to one of the α-carbons. On the other hand, in the IR spectrum an absorption at 1629 cm^{-1} for a double bond was observed. From the HMQC and HMBC data the double bond was located between C-1′ and C-α′. All these evidences placed the CHO group at C-α′. The combination of a double bond placed like this and substituted with a formyl function was detected for the first time for bisbenzylisoquinoline alkaloids. ^{1}H NMR spectrum of **1** contained a broad two-proton singlet at δ 5.07 corresponding to a CH$_2$ group. A downfield signal for secondary carbon was also observed in the ^{13}C NMR spectrum and by the DEPT experiment. The analyses of the HMQC and HMBC data led to the assignment of this CH$_2$ group, connected the two benzylisoquinoline moieties of **1** between C-10 and C-12′. To our knowledge, this is the

Table 1
^{1}H NMR, ^{13}C NMR, HMQC, HMBC, NOESY and COSY data of compound **1**[a]

Position	^{1}H (J Hz)	^{13}C and HMQC	HMBC	NOESY	COSY
1	3.94 *m*	63.7 d		2-NCH$_3$H-8	H-α
2-NCH$_3$	2.55 *s*	41.9 *q*	C-1, C-3	H-1	
3	2.81 *m*, 3.28 *m*	45.7 *t*	C-1		H-4
4	3.28 *m*	21.1 *t*	C-4*a*, C-8*a*		H-3
4*a*		119.3 *s*			
5		150.8 *s* or 150.9 *s*			
6		140.6 *s*			
7		151.1 *s*			
8	5.98 *s*	106.9 *d*	C-1, C-4*a*, C-6, C-7	7-OCH$_3$H-14, H-1	
8*a*		122.3 *s*			
α	3.28 *m*	21.1 t	C-8*a*, C-9, C-10		H-1
9		136.5 *s*			
10		123.5 *s*			
11	7.31 *s*	104.8 *d*	C-9, C-10, C-12, C-13	12-OCH$_3$, H-11′	
12		158.4 *s*			
13		153.0 *s*			
14	7.96 *s*	108.0 *d*	C-9, C-10, C-12, C-13	13-OCH$_3$, H-8	
1′		146.9 *s*			
3′	2.81 *m*	19.2 *t*	C-1′, C-4′*a*		H-4′
4′	2.81 *m*, 3.28 *m*	34.1 *t*	C-5′, C-4′*a*, C-8′*a*		H-3′
4′*a*		121.9 *s*			
5′	6.80 *s*	114.6 *d*	C-1′, C-4′, C-6′, C-7′	6′-OCH$_3$	
6′		145.7 *s*			
7′		148.5 *s*			
8′	6.51 *s*	103.6 *d*	C-1′, C-4′*a*, C-6′, C-7′	7′-OCH$_3$, H-10′, 14′ -OCH$_3$	
8′*a*		122.3 *s*			
α′		151.7 *s*			
9′		137.4 *s*			
10′	6.92 *br s*	109.0 *d*	C-α′, C-9′, C-12′, C-14′	H-8′, H-11′	
11′	7.89 *br s*	118.1 *d*	C-9′, C-12′	H-10′, H-11	
12′		123.5 *s*			
13′		150.9 *s* or 150.8 *s*			
14′		145.7 *s*			
α′-CHO	10.68 *s*	147.4 *d*	C-9′	14′-OCH$_3$	
5-OCH$_3$	3.82 *s* or 3.97 *s*	60.4 *q* or 56.3 *q*	C-5		
6-OCH$_3$	3.79 *s*	56.3 *q*	C-6		
7-OCH$_3$	3.57 *s*	55.7 *q*	C-7	H-8	
12-OCH$_3$	4.13 *s*	57.0 *q*	C-12	H-11	
13-OCH$_3$	4.10 *s*	57.1 *q*	C-13	H-14	
6′-OCH$_3$	3.86 *s*	56.3 *q*	C-6′	H-5′	
7′-OCH$_3$	3.80 *s*	60.8 *q*	C-7′	H-8′	
13′-OCH$_3$	3.97 *s* or 3.82 *s*	56.3 *q* or 60.4 *q*	C-13′		
14′-OCH$_3$	3.99 *s*	61.3 *q*	C-14′	H-8′, α′-CHO	
CH$_2$-bridge	5.07 *br s*	54.3 *t*	C-10, C-12′		

[a] Carbon multiplicities were established by DEPT data.

first example for a CH_2 bridge in bisbenzylisoquinoline alkaloids. The NOESY experiment, carried out, were in agreement with the proposed structure of **1**. The cross peaks between H-8 (δ 5.98) and OCH_3 group at C-7 (δ 3.57), H-5′(δ 6.80) and OCH_3 group at C-6′ (δ 3.86), H-8′ (δ 6.51) and OCH_3 group at C-7′ (δ 3.80) were observed. The NOESY spectrum showed significant correlations between H-11 (δ 7.31) and OCH_3 group at C-12 (δ 4.13) and between H-14 (δ 7.96) and OCH_3 group at C-13 (δ 4.10). NOE between H-10′ (δ 6.92) and H-11′ (δ 7.89) was visible, but these protons showed no correlations with any methoxy groups. The spatial relationships between H-11 (δ 7.31) and H-11′ (δ 7.89), as well as between H-10′ (δ 6.92) and H-8′ (δ 6.51) were also observed. Further important NOE in the NOESY spectrum are given in Table 1. The assignments of the chemical shifts of all carbons and hydrogens were made using HMQC and HMBC experiments (see Table 1). Compound **1** is a racemate, since the specific rotation is 0°. All these data suggested the structure of the new bisbenzylisoquinoline alkaloid *racem.* isopyruthaldine as **1**.

Isopythaldine (**2**) was isolated as a yellow amorphous solid. The CI mass spectrum exhibited a $[M + H]^+$ peak at *m*/*z* 753, corresponding to the molecular formula $C_{43}H_{48}N_2O_{10}$. The base peak in the EI mass

Table 2
^{1}H NMR, ^{13}C NMR, HMQC, HMBC, NOESY and COSY data of compound **2**[a]

Position	^{1}H (*J* Hz)	^{1}C and HMQC	HMBC	NOESY	COSY
1	3.92 *m*	63.7 *d*		2-NCH_3, H-8	H-α
2-NCH_3	2.56 *s*	42.0 *q*	C-1, C-3	14-OCH_3, H-1	
3	2.82 *m*, 3.15 *m*	45.6 *t*	C-4*a*		H-4
4	3.15 *m*	27.4 *t*	C-4*a*, C-5	5-OCH_3	H-3
4*a*		119.3 *s*			
5		150.9 *s*			
6		140.5 *s*			
7		151.0 *s*			
8	5.95 *s*	107.0 *d*	C-1, C-4*a*, C-6, C-7	7-OCH_3, 14-OCH_3, H-1	
8*a*		122.0 *s* or 131.3 *s*			
α	3.15 *m*	21.1 *t*	C-1, C-9, C-10		H-1
9		138.0 *s*			
10	6.91 *br s*	109.9 *d*	C-9, C-12, C-14	H-11, H-8′	
11	7.91 *br s*	118.9 *d*	C-9, C-12, C-13	H-10, H-11′	
12		123.9 *s*			
13		150.8 *s*			
14		146.3 *s*			
1′		147.3 *s*			
3′	2.82 *m*	19.2 *t*	C-1′		H-4′
4′	2.82 *m*, 3.15 *m*	34.0 *t*	C-4′*a*, C-5′		H-3′
4′*a*		121.5 *s*			
5′	6.78 *s*	114.6 *d*	C-1′, C-4′, C-6′, C-7′	6′-OCH_3	
6′		145.6 *s*			
7′		148.5 *s*			
8′	6.44 *s*	102.9 *d*	C-1′, C-4′a, C-6′, C-7′	7′-OCH_3, H-10	
8′*a*		131.3 *s* or 122.0 *s*			
α′		156.6 *s*			
9′		138.7 *s*			
10′		124.9 *s*			
11′	7.31 *s*	102.9 *d*	C-α′, C-9′, C-10′, C-12′	H-11	
12′		151.2 *s* or 151.3 *s*			
13′		151.3 *s* or 151.2 *s*			
14′	7.83 *s*	105.9 *d*	α′-CHO, C-α′, C-9′, C-12′	α′-CHO	
α′-CHO	10.60 *s*	147.9 *d*	C-9′, C-10′, C-14′	H-14′	
OCH_2O	6.27 *s*	103.4 *t*	C-12′, C-13′		
5-OCH_3	3.96 *s*	61.2 *q*	C-5	H-4	
6-OCH_3	3.80 *s*	60.8 *q*	C-6		
7-OCH_3	3.56 *s*	55.6 *q*	C-7	H-8	
13-OCH_3	3.82 *s*	60.4 *q*	C-13		
14-OCH_3	3.97 *s*	61.2 *q*	C-14	2-NCH_3, H-8	
6′-OCH_3	3.86 *s*	56.3 *q*	C-6′	H-5′	
7′-OCH_3	3.77 *s*	56.3 *q*	C-7′	H-8′	
CH_2-bridge	5.03 *br s*	54.4 *t*	C-12, C-10′		

[a] Carbon multiplicities were established by DEPT data.

spectrum at *m*/*z* 236 was for the rings A and B. A weak peak at *m*/*z* 516 due to the loss of the rings A and B from the molecule was also observed. The weak molecular ion at *m*/*z* 752 and the displayed mass spectral fragmentation proved a bisbenzylisoquinoline alkaloid. The ^{1}H NMR spectrum of **2** showed singlets for seven aromatic protons, seven sharp singlets for OCH$_3$ groups, a two-proton singlet for one methylenedioxy group and a three-proton singlet for one NCH$_3$ group (Table 2). For compound **2** the same set of NMR experiments was carried out, like for **1**. The obtained spectral data of **2** were related to those of **1**. The alkaloid contained a NH group and a double bond with attached CHO function to it, as well as an intramolecular H-bond at 3535 cm^{-1} (CCl$_4$; *c* 1.0 × 10^{-3} M/l). The arrangement of the groups were made according to the interpretations of the spectral results as in **1**. Compound **2** also had a new type of CH$_2$ bridge, which was placed between C-12 and C-10′. The assignment of the chemical shifts of all protons, carbons and groups of **2** were in agreement with the NOESY, HMQC, HMBC and COSY experimental data (Table 2). The alkaliod **2** is a racemate, since the specific rotation is 0°. According to the spectral analyses the structure **2** represented the new bisbenzylisoquinoline alkaloid *racem.* isopythaldine.

(+)-Isothalmidine (**3**) was isolated as a pale yellow solid. The [M + H]$^+$ peak at *m*/*z* 621 in the CI mass spectrum was in agreement with the molecular formula $C_{37}H_{36}N_2O_7$. In accordance with this composition was the base molecular ion at *m*/*z* 620 in the EI mass spectrum, which corresponds to a bisbenzylisoquinoline alkaloid. The ^{1}H NMR spectrum of **3** displayed a three-proton singlet at δ 2.35 for one NCH$_3$ group. The most downfield singlet at δ 8.39 was for a proton of a NCHO group, which was confirmed by

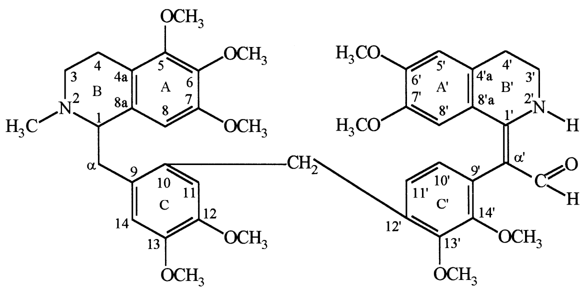
Table 3
^{1}H NMR, ^{13}C NMR, HMQC, HMBC, NOESY and COSY data of compound **3**[a]

Position	^{1}H (*J* Hz)	^{13}C and HMQC	HMBC	NOESY	COSY
1	3.72 *m*	61.8 *d*		2-NCH$_3$, H-α	H-α
2-NCH$_3$	2.35 *s*	42.3 *q*	C-1, C-3	H-1	
3	2.87 *m*, 3.53 *m*	44.4 *t*	C-4*a*		H-4
4	2.58 *m*, 2.87 *m*	21.9 *t*	C-4*a*, C-8*a*		H-3
4*a*		132.9 *s*			
5	6.31 *s*	104.9 *d*	C-4, C-6, C-7, C-8a	6-OCH$_3$	
6		145.6 *s*			
7		134.4 *s*			
8		141.4 *s*			
8*a*		122.7 *s*			
α	2.58 *m*	41.4 *t*	C-8*a*, C-9, C-10	H-1	H-1
9		122.7 *s*			
10	6.22 *d* (2.2)	132.4 *d*	C-11, C-12, C-α		
11		145.4 *s*			
12		149.8 *s*			
13	5.97 *d* (7.7)	110.2 *d*	C-9, C-11, C-14	H-14	H-14
14	6.52 *dd* (7.7, 2.2)	115.8 *d*	C-9, C-12, C-13, C-α	H-13	H-13
1′	5.61 *dd* (5.6, 10.8)	53.8 *d*	C-4′*a*, C-8′	2′-NCHO, H-8′, H-14′	H-α′
2′-NCHO	8.39 *s*	161.2 *d*	C-1′, C-3′	H-1′	
3′	6.90 *s*	123.0 *d*	C-4′*a*, C-1′	4′-OCH$_3$	
4′		147.2 *s*			
4′*a*		134.4 *s*			
5′	6.57 *s*	123.0 *d*	C-1′, C-3′, C-4′, C-6′, C-8′*a*	6′-OCH$_3$	
6′		149.3 *s*			
7′		125.8 *s*			
8′	6.21 *s*	120.1 *d*	C-1′, C-6′, C-7′, C-8′*a*	H-1′, H-10′	
8′*a*		125.8 *s*			
α′	2.87 *m*, 3.04 *m*	37.8 *t*	C-1′, C-9′, C-10′		H-1′
9′		132.9 *s*			
10′	6.86 *dd* (8.2, 2.2)	111.5 *d*	C-9′, C-14′	H-8′, H-11′	H-11′
11′	6.79 *dd* (8.2, 2.6)	121.9 *d*	C-9′, C-12′, C-13′	H-10′	H-10′
12′		154.1 *s*			
13′	7.15 *dd* (8.2, 2.6)	122.1 *d*	C-9′, C-11′, C-12′	H-14′	H-14′
14′	7.45 *dd* (8.2, 2.2)	130.3 *d*	C-10′, C-12′, C-α′	H-1′, H-13′	H-13′
6-OCH$_3$	3.78 *s*	56.1 *q*	C-6	H-5	
4′-OCH$_3$	3.93 *s*	56.1 *q*	C-4′	H-3′	
6′-OCH$_3$	3.42 *s*	56.2 *q*	C-6′	H-5′	

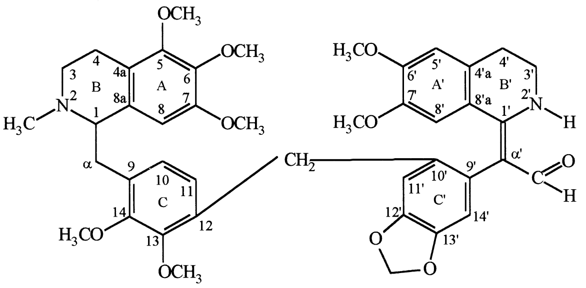
[a] Carbon multiplicities were established by DEPT data.

the absorption at 1683 cm^{-1} in the IR spectrum. The downfield *dd* signal at δ 5.61 for H-1′ also supported this assertion (Tantisewie et al., 1990). The signal for H-1 was visible, too. Furthermore the ^1H NMR spectrum contained three sharp singlets for OCH_3 groups and four singlets for aromatic protons. In the aromatic region an ABX spin system for the ring C and an AA′XX′ spin system for the 1,4-disubstituted benzene ring C′ was observed (see Table 3). Only four signals for secondary carbons in the ^{13}C NMR spectrum were detected. It means that in **3** the ring B or B′ contains a double bond between C-3 and C-4 or between C-3′ and C-4′. In addition, the IR spectrum displayed an absorption at 1630 cm^{-1} for a double bond. According to the data from NOESY, HMQC and HMBC experiments, the double bond was situated in ring B′ and it was substituted with one OCH_3. The HMBC correlations between the proton from the amide group at 2′-place and C-3′, between H-5′ and C-4′, as well as the NOE between H-3′ and OCH_3 group determined the place of the OCH_3 group at C-4′ (see Table 3). This is the first example for a bisbenzylisoquinoline alkaloid with a C-3′–C-4′ double bond. From the mass and the spectral data discussed above it might be proposed that **3** contains two bridges — one ether and one C –C bridge. The HMQC, HMBC and NOESY correlations led to the final location of the C–C bridge between C-7 and C-7′ and of the ether bridge between C-11 and C-12′ (Fig. 3). To our knowledge, **3** displayed a bisbenzylisoquinoline alkaloid with a new type of C–C bridge. The combination of all spectral data proved that the structure of the new bisbenzylisoquinoline alkaloid (+)-isothalmidine is **3**.

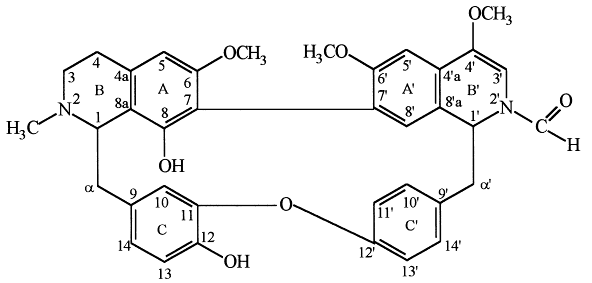
The isoquinoline alkaloids reticuline, isocorydine, columbamine and palmatine were indentified by direct comparison of their UV, IR, ^1H NMR and mass spectral data with these of authentic samples (Sidjimov and Marekov, 1982; Kuzmanov et al., 1992; Ivanovska and Philipov, 1996). The alkaloids were new for the genus *Isopyrum*.



1



2



3

3. Experimental

3.1. General

UV: EtOH. IR: $CHCl_3$. ^1H, ^{13}C NMR spectra and 2D experiments: 250 MHz in $CDCl_3$ with TMS as international standard. MS: 70 eV. CC: silica gel (Merck, Kieselgel 60, 70–230 mesh) and neutra alumina (Merck, Aluminiumoxid 90, act. II–III Brockmann, 70–230 mesh). PTLC: silica gel GF_{254}. Spray reagent for TLC: Dragendorff's reagent.

3.2. Plant material

Isopyrum thalictroides L. was collected in May 1995 during the time of flowering from Ljulin mountain near Sofia. A voucher specimen (SOM-320) is deposited in the Herbarium of the Institute of Botany, Bulgarian Academy of Sciences.

3.3. Extraction and isolation

3.3.1. Roots and rhizomes

From the crude mixture of tertiary alkaloids, described in (Philipov and Istatkova, 1999), by consecutive use of CC on silica gel with $CHCl_3$–MeOH (increasing polarity), then on neutra aluminia with hexane–Me_2CO (increasing polarity) and finally by prep. TLC with $CHCl_3$–MeOH–NH_4OH (30:8:0.4) **1** (10.4 mg) and **2** (13.4 mg), and by prep. TLC with petrol-$CHCl_3$–Me_2CO–MeOH (4:4:1:1) **3** (9.3 mg) were obtained.

3.3.2. Aerial parts

Air-dried and ground aerial parts (500 g) were worked up to obtain 0.4 g crude mixture of tertiary alkaloids (A) (Philipov and Istatkova, 1997). Then, the ammonia–alkaline water phase was acidified (pH 6–6.5) with concentrated. HCl, 65 ml satd. solution of KJ was added and extracted with $CHCl_3$ (6 × 200 ml). The combined $CHCl_3$ extracts were dried (Na_2SO_4) and evaporated under reduced pressure to give 0.06 g of crude mixture of quaternary alkaloids (B). By the consecutive use of CC on silica gel with $CHCl_3$–MeOH (increasing polarity) and then by prep. TLC with petrol–$CHCl_3$–Me_2CO–MeOH (4:4:1:1) from A reticuline (1.0 mg) and isocorydine (5.2 mg) were isolated. By consecutive use of CC on silica gel with $CHCl_3$-MeOH (increasing polarity) and then by prep. TLC with $CHCl_3$–MeOH–NH_4OH (30:8:0:4) from B columbamine (1.6 mg) and palmatine (2.0 mg) were obtained.

3.4. Isopyruthaldine (*1*)

UV λ_{max} nm (log ε): 215 (6.72), 242 (6.66), 250 *sh* (6.59), 260 *sh* (6.44), 282 *sh* (6.56), 292 (6.65). IR ν_{max} cm^{-1}: 3390 (NH), 1721 (C=O), 1629 (C=C). ^1H NMR: see Table 1. ^{13}C NMR: see Table 2. CIMS (NH_3) *m/z* (rel. int.): 769 $[M + H]^+$ (100). EIMS *m/z* (rel. int.): 768 (4), 533 (40), 236 (100), 206 (10).$[\alpha]_D$ 0° ($CHCl_3$; *c* 0.16).

3.5. Isopythaldine (*2*)

UV λ_{max} nm (log ε): 215 (6.64), 242 (6.61), 250 *sh* (6.54), 260 *sh* (6.40), 282 *sh* (6.48), 292 (6.56). IR ν_{max} cm^{-1}: 3388 (NH), 1720 (C=O), 1629 (C=C). ^1H NMR: see Table 1. ^{13}C NMR: see Table 2. CIMS (NH_3) *m/z* (rel. int.): 753 $[M + H]^+$ (100). EIMS *m/z* (rel. int.): 752 (2), 516 (4), 236 (100), 206 (10). $[\alpha]_D$ 0° ($CHCl_3$; *c* 0.21).

3.6. (+)-Isothalmidine (*3*)

UV λ_{max} nm (log ε): 210 (6.52), 240 *sh* (6.17), 290 (5.77), 312 (5.77). IR ν_{max} cm^{-1}: 3542 (OH), 3530 (OH), 1683 (C=O), 1630 (C=C). ^1H NMR: see Table 1. ^{13}C NMR: see Table 2. CIMS (NH_3) *m/z* (rel. int.): 621 $[M + H]^+$ (100). EIMS *m/z* (rel. int.): 620 (100), 605 (18), 589 (8), 417 (20), 393 (64), 365 (72), 319 (14), 310 (8). $[\alpha]_D$ + 14° ($CHCl_3$; *c* 0.46).

Acknowledgements

This work was supported by a grant from the National Fund for Scientific Research, Bulgaria. We thank Dr. P. Denkova for the NMR spectra and her fruitful discussion. We are indebted to Dr. D. Tsankov and Mrs. A. Poneva for the IR spectra and Mrs. P. Tuleva for her technical assistance.

References

Ivanovska, N., Philipov, S., 1996. Study on the anti-inflammatory action of *Berberis vulgaris* root extract, alkaloid fractions and pure alkaloids. Int. J. Immunopharmac. 18, 553–561.

Kuzmanov, B.A., Philipov, S.A., Deligiozova-Gegova, I.B., 1992. Comparative phytochemical and chemosystematic research of populations of *Glaucium flavum* Crantz in Bulgaria. Fitologija 43, 52–58.

Philipov, S., Istatkova, R., 1997. Alkaloids from *Isopyrum thalictroides*. Phytochemistry 44, 1591–1594.

Philipov, S., Istatkova, R., 1999. Alkaloids from *Isopyrum thalictroides*. Phytochemistry 51, 1161–1165.

Sidjimov, A., Marekov, N., 1982. Biosynthesis of thalicarpine in *Thalictrum minus*. Phytochemistry 21, 871–874.

Tantisewie, B., Pharadai, T., Freyer, A.J., Guinaudeau, H., Shamma, M., 1990. Four amidic bisbenzylisoquinoline alkaloids from *Cyclea atjehensis*. J. Nat. Prod. 53, 553–558.