



# Iridoid glucosides from *Plantago cornuti*, *Plantago major* and *Veronica cymbalaria*

Rilka Taskova<sup>a</sup>, Nadjalka Handjieva<sup>b,\*</sup>, Ljubka Evstatieva<sup>a</sup>, Simeon Popov<sup>b</sup>

<sup>a</sup>*Institute of Botany, Bulgarian Academy of Sciences, 1113, Sofia, Bulgaria*

<sup>b</sup>*Institute of Organic Chemistry with Centre of Phytochemistry, Bulgarian Academy of Sciences, 1113, Sofia, Bulgaria*

Received 29 April 1998; received in revised form 26 January 1999; accepted 8 February 1999

## Abstract

The new iridoid glucoside from the rare  $\Delta 8,9$  type, 10-acetoxymajoroside, was isolated from *Plantago cornuti* and *P. major* along with known iridoids. The main constituents in *V. cymbalaria* were the known catalpol, amphicoside and verproside together with alpinoside, aucubin, 6-O-veratroylcatalpol and verminoside. The iridoid alpinoside with a 8,9-double bond was found for the first time in genus *Veronica*. © 1999 Elsevier Science Ltd. All rights reserved.

**Keywords:** *Plantago cornuti*; *Plantago major*; *Veronica cymbalaria*; Plantaginaceae; Scrophulariaceae; Iridoid glucosides; 10-Acetoxymajoroside; Alpinoside; 10-Hydroxymajoroside; Majoroside

## 1. Introduction

In most of the recent taxonomic schemes the families Plantaginaceae and Scrophulariaceae are included in one order (Takhtajan, 1987; Dahlgren, 1980; Thorne, 1981). According to Takhtajan (1987) and Hegnauer (1969) a close relation between these families exists. Moreover, a direct origin of Plantaginaceae from Scrophulariaceae is suggested. Many authors on the basis of morphological, embryological and phytochemical data consider that the genus *Veronica* could be the link between the two families (Takhtajan, 1987; Hegnauer, 1969; Willis, 1980).

In continuation of our studies on *Veronica* (Taskova, Peev, Handjieva & Baranovska, 1997; Taskova, Handjieva, Peev & Popov, 1998) and *Plantago* (Handjieva, Taskova & Popov, 1993; Handjieva et al., 1991) the present paper deals with the study of the iridoid glucosides in *Plantago cornuti* Gouan, *Plantago major* L and *V. cymbalaria* Bodard. Previous phytochemical investigations on *V. cymbalaria* have revealed the presence of iridoid glucosides

(Grayer-Barkmajer, 1973) and glucosides of luteolin, apigenin and chrysoeriol (Grayer-Barkmajer, 1978). Only aucubin, 10-hydroxymajoroside and melittoside were reported for *P. cornuti* (Handjieva et al., 1993; Andrzejewska-Golec & Swiatek, 1984), while in *P. major* many constituents as iridoids, phenolic compounds, alkaloids, etc. were found.

## 2. Results and discussion

The methanolic extracts of dried aerial parts of *P. cornuti*, *P. major* and *V. cymbalaria*, were partitioned between water and dichloroethane and the water soluble parts were treated with charcoal and eluted with different solvent mixtures. Chromatography of some of the obtained fractions yielded the new compound **1** besides several known iridoids.

### 2.1. *P. cornuti* and *P. major*

From both species a new compound (**1**) was isolated, besides the previously isolated by us aucubin (**2**) and 10-hydroxymajoroside (**3**) from *P. cornuti*

\* Corresponding author. Fax: +359-2-700-225.

Table 1

$^{13}\text{C}$  NMR spectral data of 10-acetoxymajorside (1) (62.9 MHz,  $\text{D}_2\text{O}$ ) and the model compound 10-hydroxymajorside (3) (62.9 MHz,  $\text{D}_2\text{O}$ )

C	1 <sup>a</sup>	3 <sup>b</sup>
Aglycon moiety		
1	93.7 d	93.8
3	153.9 d	153.9
4	115.2 s	115.3
5	38.2 d	38.1
6	41.9 t	42.1
7	78.4 d	78.7
8	140.1 s	143.9
9	139.3 s	137.6
10	61.3 t	58.2
11	172.2 s	172.5
OMe	54.7 q	54.7
Sugar moiety		
1	101.2 d	101.2
2	75.5 d	75.4
3	79.1 d	78.4
4	72.3 d	72.3
5	79.2 d	79.2
6	63.5 t	63.5
CH <sub>3</sub> CO		
	23.1 q	
	176.8 s	

<sup>a</sup> Multiplicities determined by DEPT, assignments by HMQC.

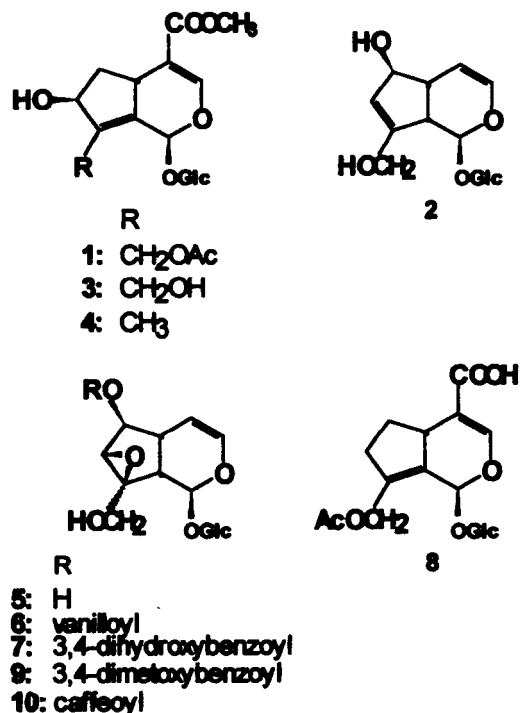
<sup>b</sup> Data from (Handjieva et al., 1993); in  $\text{D}_2\text{O}$ , 62.9 MHz.

(Handjieva et al., 1993) and aucubin (2) and majorside (4) from *P. major* (Handjieva et al., 1991).

Compound (1) possessed NMR data for a  $\text{C}_{10}$  iridoid. The  $^{13}\text{C}$  NMR (Table 1) contained 19 signals, 6 of which were consistent with a glucosidic moiety and aglycone signals similar to 10-hydroxymajorside (3). The  $^1\text{H}$  NMR spectrum resembled that of 3 with the exception of the appearance of a signal for an acetoxy group (three proton singlet at  $\delta 2.12$ ) and deshielding of  $\text{H}_2$ -10, which showed the presence of a 10- $\text{CH}_2\text{OAc}$  group. The  $^{13}\text{C}$  NMR data confirmed this suggestion. Deshielding of C-10 from  $\delta 58.2$  to  $\delta 61.3$  and shielding of C-8 from  $\delta 143.9$  to  $\delta 140.1$  were observed. Hence, 1 is 10-acetoxymajorside.

## 2.2. *V. cymbalaria*

Seven pure compounds were isolated and identified as the known iridoid glucosides 2, 5–10. The major constituents were catalpol (5), amphicoside (6) and verproside (7). Minor components were alpinoside (8) (Jensen, Olsen, Rahn & Rasmussen, 1996), aucubin (2), 6-*O*-veratroylcatalpol (9) and verminoside (10). The known isolated iridoid glucosides were identified by spectral data and comparison with authentic samples. Alpinoside (8) was found for the first time in



genus *Veronica* and 6-*O*-veratroylcatalpol (9) in *V. cymbalaria*.

Till now, only in *Plantago* and *Veronica* were found iridoids with the uncommon 8,9-double bond, most of which in *Plantago*—majorside, 10-hydroxymajorside, 10-acetoxymajorside, alpinoside, hookerioside and desacetylhookerioside (Damtoft, Falkesgaard & Jensen, 1994) and in *Veronica*—anagalloside (Lahloub, 1992) and alpinoside. The presence of iridoids with this structural feature in *Veronica* and *Plantago* is in support of the consideration that the genus *Veronica* is the link between the families Plantaginaceae and Scrophulariaceae.

## 3. Experimental

### 3.1. General procedure

$^1\text{H}$  NMR (250 MHz) and  $^{13}\text{C}$  NMR (62.9 MHz) in  $\text{D}_2\text{O}$ . Reverse phase LPLC: Merck Lobar RP-18 column size C,  $\text{H}_2\text{O}$ – $\text{MeOH}$  mixtures were used as eluent. *P. cornuti* and *P. major* were collected at florescence in Tsarevo (0 m s. m., 1991) and Baltchic (0 m s. m., 1992), respectively, and identified by L. Evstatieva. *Veronica cymbalaria* was collected at florescence in Sozopol (0 m s. m.) in 1996 and identified by R. Taskova. The voucher specimens SOM 151024, 153556 and 153555 were deposited in the herbarium of the Institute of Botany, Bulgarian Academy of Sciences.

### 3.2. Isolation of glucosides

Dry above-ground parts were extracted twice with MeOH and the concd extract partitioned between  $\text{Cl}(\text{CH}_2)_2\text{Cl}$ – $\text{H}_2\text{O}$ . The aq. phase was concd, treated with charcoal and eluted with  $\text{H}_2\text{O}$ ,  $\text{H}_2\text{O}$ –MeOH mixtures (95:5, 70:30, 50:50), MeOH, MeOH– $\text{Me}_2\text{CO}$  (1:1) and MeOH– $\text{Cl}(\text{CH}_2)_2\text{Cl}$  (1:1).

### 3.3. *P. cornuti*

Dry above-ground parts (120 g), total MeOH extract (19 g). The MeOH– $\text{Me}_2\text{CO}$  and the MeOH– $\text{Cl}(\text{CH}_2)_2\text{Cl}$  frs. (0.6 g) after purification on silica gel yielded impure **1** (frs. 6–7, 21 mg), which was additionally purified on a RP-18 Lobar column to give pure **1** (17 mg).

### 3.4. *P. major*

Dry above-ground parts (38 g), total MeOH extract (10 g). The MeOH– $\text{Cl}(\text{CH}_2)_2\text{Cl}$  frs. (80 mg) after purification on silica gel yielded **1** (frs. 7–8, 12 mg), **2** (21 mg) and **3** (9 mg).

### 3.4. 10-Acetoxy*majoroside* (**1**)

$[\alpha] -59^\circ$  (MeOH,  $c$  0.3); UV  $\lambda_{\text{max}}$ , nm 238 (MeOH);  $^1\text{H}$  NMR (250 MHz,  $\text{D}_2\text{O}$ ): 7.38 (d,  $J=2$  Hz, H-3), 6.34 (s, H-1), 4.80 (H<sub>2</sub>-10), 3.85 (s, OMe), 3.55 (H-7), 2.91 (m, H-5), 2.45 (m, H-6a), 2.12 (s, Me), 1.90 (m, H-6b).  $^{13}\text{C}$  NMR: Table 1.

### 3.5. *V. cymbalaria*

Dry above-ground parts (52 g), total MeOH extract (11 g). The MeOH fr. (0.75 g) was separated on silica gel and eluted with  $\text{CHCl}_3$ –MeOH– $\text{H}_2\text{O}$  (60:22:4) to give pure **2** and a mixture of **2** and **5** (frs. 21–23, 23 mg). The MeOH– $\text{Me}_2\text{CO}$  fr. (0.55 g) was chro-

matographed on silica gel and consecutively on a RP-18 Lobar column to afford pure **8** (frs. 25–30, 18 mg). The MeOH– $\text{Cl}(\text{CH}_2)_2\text{Cl}$  fr. (0.77 g) was purified on Lobar to give pure **6** (127 mg), **7** (62 mg), **9** (54 mg) and **10** (7 mg).

### Acknowledgements

This project X-513 has been completed with the financial support of the National Foundation for Scientific Research, which is greatly acknowledged.

### References

- Andrzejewska-Golec, E., & Swiatek, L. (1984). *Herba Polonica*, 30, 9.
- Dahlgren, R. (1980). *Botanical Journal of the Linnean Society*, 80, 91.
- Damtoft, S., Falkesgaard, E., & Jensen, S. R. (1994). *Phytochemistry*, 35, 1367.
- Grayer-Barkmajer, R. J. (1973). *Biochem. Syst. & Ecol.*, 1, 101.
- Grayer-Barkmajer, R. J. (1978). *Biochem. Syst. & Ecol.*, 6, 131.
- Handjieva, N., Taskova, R., & Popov, S. (1993). *Z. Naturforschung*, 48c, 827.
- Handjieva, N., Spassov St, Bodurova, G., Saadi, H., Popov, S., Pureb, O., & Zamjansan, J. (1991). *Phytochemistry*, 30, 1317.
- Hegnauer, R. (1969). *Chemotaxonomie der Pflanzen*, vol. 5 (p. 337). Basel: Birkhäuser.
- Jensen, S. R., Olsen, C. E., Rahn, K., & Rasmussen, J. H. (1996). *Phytochemistry*, 42, 1633.
- Lahloub, M. F. (1992). *Alex. J. Pharm. Sci.*, 6, 134.
- Takhtajan, A. (1987). In *Sistema magnoliifitov* (p. 247). Leningrad: Nauka.
- Taskova, R., Handjieva, N., Peev, D., & Popov, S. (1998). *Phytochemistry*, 49, 1323.
- Taskova, R., Peev, D., Handjieva, N., & Baranovska, L. (1997). *Phytologia Balcanica*, 1, 79.
- Thorne, R. (1981). In D. A. Young, & D. S. Seigler, *Phytochemistry and angiosperm phylogeny* (p. 233). New York: Praeger.
- Willis, J. (1980). In *A dictionary of the flowering plants and ferns* (8th ed.) (p. 911). Cambridge: University Press.