ARBUTIN IN *SERRATULA QUINQUEFOLIA* M.B. (ASTERACEAE)

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ABSTRACT

Genus *Serratula* is known for the presence of steroid compounds which belong to the group of ecdysones. Former phytochemical works from the late sixties and early seventies indicate, in some *Serratula* species, the occurrence of a phenolic glycoside-arbutin. This has been confirmed in the present work through finding an α,β-arbutin anomer in *Serratula quinquefolia* M.B. New botanical data concerning the classification of genus *Serratula* suggest that the species in question should be regarded as belonging to genus *Klasea* (currently a section of genus *Serratula*). There has been an attempt to find a key to the chemical division within the taxons.

Key words: arbutin, *Serratula*, *Klasea*, Centaureinae, Asteraceae, ecdysones, chemotaxonomy.

INTRODUCTION

The systematics of *Klasea* Cass causes much controversy and is still a subject of discussions among botanists. This taxon is sometimes considered an autonomous genus, or a section of genus *Serratula* L., and it used to be regarded as *Rhaponticum* subgenus. At present, the distinctness of *Klasea* on the level of genus is being emphasized, through most of the current floristic works still tend to follow the traditional classification with the *Klasea* section within *Serratula*. The *Klasea* species occur in the range of places reaching from Southern and Eastern Europe through Northern Africa to western Asia, with special mention of the mountains in Central Asia (Martins 2006). They belong to subtribe Centaureinae (Asteraceae) and they are sister species to the Rhaponticum group (Martins and Hellwig 2005), where mainly sesquiterpene lactones and ecdysones have been found (Nowak 1992; Cis et al. 2006).

On the basis of modern botanical analyses, *Serratula quinquefolia* is ascribed to genus *Klasea* (Martins 2005). This species occurs in Armenia, Azerbaijan, Greece, Iran, Turkey and Russia. It is identified in accordance with the morphological description in Flora SSSR (Borisova1963) and Flora of European part of the former USSR (Tsherepanov1994).

This plant is cultivated in the Garden of Department of Medical and Cosmetic Natural Products, Poznan Medical University of Sciences (Poland). From the dry herb of this species, arbutin (hydroquinone – glucopyranoside) was isolated. By spectral methods: ^{1}H NMR and ^{13}C NMR it was established as a mixture of α and β epimers.

Arbutin is a well known compound occurring in such herbal medicines as *Arctostaphylos uva-ursi* and *Vaccinium vitis-idaea* (Ericaceae), *Majorana hortensis* and *Origanum vulgare* (Lamiaceae) as well as in *Bergenia crassifolia* (Saxifragaceae), *Pyrus communis* (Rosaceae) and *Viburnum opulus* (Caprifoliaceae).

Herbal medicines which include arbutin in the appropriate concentration display a disinfecting activity on urinary tracts, thanks to its hydrolysis, which leads to the formation of active hydroquinone (Weiss and Fintelmann 2000). This compound is also responsible for inhibiting the biosynthesis of melatonin, and applied in the treatment of hyperpigmentation (Maeda and Fakuda 1996). Arbutin has been also found in some species of genus *Serratula* – *S. erucifolia,* S. *gmelini, S. radiata* (Yatsyuk et al. 1968) and *S. sogdiana* (Zatsny 1973). Each of these were classified as belonging to genus *Klasea* in a recent taxonomic study (Martins 2006).

Ecdysones constitute the second group of the compounds to be found in the species of genus *Serratula* – *S. algida* (Novoselskaya et al. 1981), *S. chinensis* (Chen and Wei 1989), *S.* centauroides, *S. coronata* (Gorovits et al. 1974), *S. inermis* (Yatsyuk and Segel 1970), *S. komarovii* (Vorob'eva et al. 2004), *S. lyratifolia* (Ganiev 1980), *S. procumbens* (Novoselskya et al. 1981), *S. quinquefolia* (Goro-

TABLE 1. Ecdysones and arbutin in *Serratula* genus.

Species	Synonyms	Ecd.	Arb.
Serratula algida Iljin	–	+[1]	
S. centauroides L.	*S. pectinata* Turcz. ex Herd. = *Klasea centauroides* (L.) Cass. ex Kitagawa	+[2]	
S. chinensis S. Moore	*Klasea chinensis* (S. Moore) Kitagawa = *Rhaponticum chinense* (S. Moore) L. Martins = *Centaurea missionis* H. Lév.	+[3]	
S. coronata L.	*S. wolffii* Andrae = *S. coronata* L. var. *manshurica* Kitagawa = *S. manshurica*	+[4,19]	
S. erucifolia (L.) Boriss	*S. xeranthemoides* M. Bieb. = *Xeranthemum erucifolium* L.= *Klasea erucifolia* (L.) Gaertner et Wagenitz	+[5]	+[6]
S. gmelinii Tausch	*S. isophylla* Claus = *Klasea gmelinii* (Tausch) Holub		+[7]
S. inermis Gilib.	*S. tinctoria* ssp. *inermis* var. *angustiloba* M. Pop. *S. tinctoria* auct. fl. ross. non L.	+[8]	
S. komarovii Iljin	*Klasea komarovii* (Iljin) Kitagawa = *K. centauroides* ssp. *komarovii* (Iljin) L. Martins	+[9]	
S. lyratifolia Schrenk	*Klasea lyratifolia* (Schrenk) L. Martins	+[10]	
S. procumbens Regel	*S. flexicaulis* Rupr.= *S. depressa* Regel. et Scmalh.= *Klasea procumbens* (Regel) Holub	+[11]	
S. quinquefolia M. Bieb. ex Wild	*Klasea quiquefolia* (M. Bieb.) Cass.= *Klasea quinquefolia* (Wild.) Greuter et Wagenitz	+[13]	+[14]
S. radiata Bunge	*S. bracteifolia* (Iljin ex Grossh.) Stank.= *S. radiata* ssp. *bracteifolia* Iljin ex Grossh.= *Klasea radiata* (Waldst. & Kit.) A. Löve & D. Löve		+[12]
S. sogdiana Bunge	*Klasea sogdiana* (Bunge)	+[15]	+[16]
S. strangulata Iljin	*Klasea strangulata* (Iljin) Kitagawa	+[17]	
S. tinctoria L.	? *S. inermis*	+[18]	

Ecd. = ecdysones
Arb. = arbutin
[1,11]Novoselskaya et al. (1981); [2,13]Gorovits et al. (1974); [3,4]Chen & Wei (1989); [5]Kolodova et al. (1979); [6,7,12]Yatsyuk et al. (1968); [8]Yatsyuk & Segel (1970); [9]Vorob'eva et al. (2004); [10]Ganiev (1980); [14]own study; [15]Zatsny et al. (1971); [16]Zatsny et al. (1973); [17]Dai et al. (2002); [18]Bathori et al. (1986); [19]Akhmed et al. (1990).

vits et al. 1974), *S. sogdiana* (Zatsny et al. 1971), *S. strangulata* (Dai et al. 2002), *S. tinctoria* (Bathori et al. 1986), *S. wolffii* (Akhmed et al. 1990) and *S. xeranthemoides* (Kholodova et al. 1979). Ecdysones are steroid structures with wide pharmacological properties, such as strengthening of the organism as well as decreasing the level of cholesterol (Dinan 2001). Table 1 presents those species of *Serratula*, in which ecdysones and arbutin have been found so far.

The aim of this work was to isolate and identify the dominant compound in *Serratula quinquefolia* and to present the occurrence of arbutin and ecdysones in the *Serratula* genus in the chemotaxonomic aspect.

MATERIAL AND METHODS

Plant material

The *Serratula quinquefolia* were cultivated in the Garden of Department of Medicinal and Cosmetic Natural Products of the University of Medicinal Sciences in Poznan (Poland). Aerial parts of the plant were collected in August 2007 from plants at the flowering stage. A voucher specimen (No.162/89) has been deposited at the above-mentioned garden.

Extraction, isolation and identification

Dried aerial parts of *Serratula quinquefolia* (250 g) were cut into small pieces and exhaustively extracted with methanol at room temperature. The extract was concentrated under reduced pressure providing a residue (15.5 g), which was chromatographed on a silica gel (Merck, Art 7733) column. The column eluted with methylene chloride – methanol mixture (8:1) yielded the substance A (857 mg) in the form of long white needles, m.p. 200.5°C (199.5°C cited by Rolando and Gonzales 2005). Relevant fractions containing the compound A were easily detectable on silica gel TLC plates (Merck, Art. 5553, methylene chloride – methanol 6:1, R_f = 0.14) after spraying with anisaldehyde reagent followed by heating in the temperature of 105°C for 3 minutes.

The structure of the isolated compound was identified on the basis of spectral methods: ^{1}H NMR, ^{13}C NMR, ^{1}H NMR – ^{13}C NMR cosy (Varian, 600 MHz, in CD_3OD).

RESULTS

Our chemical study of methanolic extract from aerial parts of *Serratula quinquefolia* 250 g) led to the isolation of α,β-arbutin anomer (857 mg) (Fig. 1).

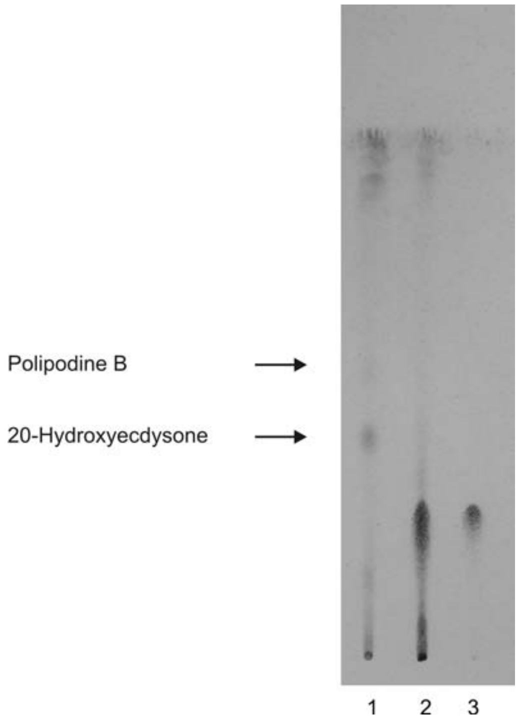

Fig. 1 Chromatogram of methanolic extracts from herbs of *Serratula wolffii, S. quinquefolia* and arbutin.

Methanolic extracts of: 1 – *Serratula wolffii;* 2 – *S. quinquefolia*; 3 – α,β-arbutin. Adsorbent: silica gel. Mobile phase: methylene chloride – methanol 6:1 (V/V). Reagent: anisaldehyde.

Its molecular formula was established on the basis of spectral methods. A comparison of the ^{1}H NMR spectral data with those in literature (Rolando and Gonzales 2005) showed the presence of additional signal at the H-1'(doublet, $J_{1',2'}$=2.2 Hz) what indicates the epimer α and the higher value of coupling constants ($J_{1'',2''}$=7.2 Hz) of the doublet (chemical shift 4.72 ppm) indicates epimer β. The ^{1}H NMR spectra analyse demonstrated (Table 2) that the isolated compound A is arbutin's α,β anomer in the ratio 1:1.9 (Fig. 2).

CONCLUSION

The inclusion of the taxon in question in three different genera in literature (see Table 1), confirms major difficulties in establishing the correct systematics of genus *Serratula*

TABLE 2. Proton NMR spectral data of substance A (600 MHz, CD_3OH) and β-D-arbutin (500 MHz, D_2O).

Protons	Substance A ^{1}H δ (*J*)	β-D-arbutin [1] ^{1}H δ (*J*)
H-1, H-2	6.98	6.94 *d*
H-3, H-5	6.84	6.76 *d*
H-1' β	4.72 *d* $J_{1',2'}$ = 7.7	4.86 *d* $J_{1',2'}$=7.8
H-1'' α	4.72 *d* $J_{1'',2''}$ = 2.2	
H-6' A	3.86	3.81 *d*
H-6' B	3.71	3.65 *dd*
H-2'-H-5'	3.53-3.25 *m*	3.54-3.38 *m*

[1]Rolando and Gonzalez (2005)
d – doublet
m – multiplet

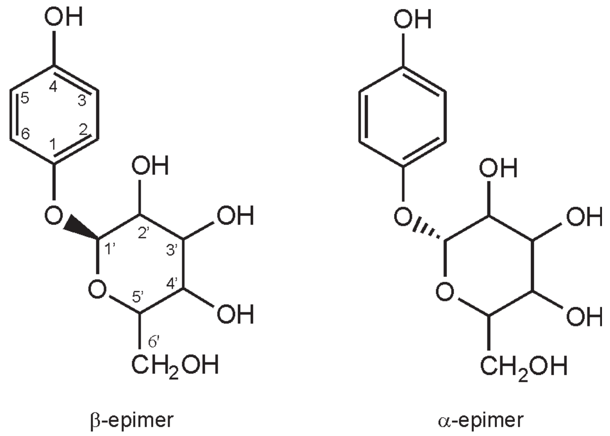

Fig. 2. Chemical structure of arbutin – hydroquinone-α,β-D-glucopyranoside.

If we assume the botanical division into the two separate genera: *Klasea* and *Serratula,* we can make the following remarks.

The phytochemical analyses conducted hitherto indicate that it is impossible to make an adequate division among the species of *Serratula* and those seemingly belonging to *Klasea*. However, there is the following pattern: in *Serratula* arbutin has not been found so far, but in *Klasea* the ecdysones prevail – out of the 11 species analysed, nine contain the very compounds. Arbutin was found in five species (including the results of present work). There are species of *Klasea* containing ecdysones only: *Klasea centauroides*, *K chinensis*, *K. komarovii*, *K. lyratifolia*, *K. procumbens* and *K. strangulata*. The following species contain both: *Klasea erucifolia*, *K. quinquefolia* and *K. sogdiana*.

Arbutin has been isolated from *Serratula quinquefolia* as a α,β-glucoside. Epimer β prevails in the amount of 65%. Ecdysones were not found in *Serratula quinquefolia*, through the methods employed in this present work.

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