Chemical diversity of silverweed (*potentilla erecta* L.) growing at the edges of arable fields

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INTRODUCTION

Silverweed (*Potentilla anserina* L., *Rosaceae)* is a perennial stoloniferous plant occurring mainly in moist meadows and pastures, often situated on the river banks, as well as in some ruderal plant communities. This species relatively rarely appears as a weed on arable fields, but often on their margins, especially if low-input cropping is used [1, 2]. It propagates both generatively and vegetatively, according to environmental conditions [3]. Silverweed herb is used as a medicinal raw material mainly because of its anti-diarrheic and spasmolytic activity. Antimicrobial and antiviral properties of silverweed extracts have been also proved [4-7]. Medicinal usage of silverweed results from the presence of phenolic compounds, especially tannins and flavonoids [7]. So far, possible intraspecific chemical diversity of this species has not been taken into consideration when collecting raw material from wild growing plants. The aim of this preliminary study was to compare the quality of raw material of silverweed growing at the edges of arable fields in three locations in Podlasie area (north-eastern Poland) in respect of the content of phenolic compounds.

RESULTS AND DISCUSSION

The medicinal usage of silverweed herb (*Anserinae herba*) is mainly related to the presence of phenolic compounds. Silverweed herb is considered to be a tannin-rich raw material with the content of these compounds reaching 10%. According to DAC (German Drug Codex Supplement to the Pharmacopoeia), the tannin content in silverweed herb should not be less than 2% [7]. In our study the content of tannins in the silverweed herb appeared to be very low (0.81-0.91%) (Table 1). These compounds reveal antidiarrheic, anti-inflammatory, antibacterial, antioxidant, and antimutagenic activity [9, 10]. Flavonoids constitute another group of active compounds in silverweed herb. They are considered to be responsible for the spasmolytic and choleretic activity of the raw material [11]. The total content of flavonoids in the investigated plant material was rather low (0.48-0.60%). Phenolic acids were present in higher amount, but their content in the raw materials obtained from the studied population differed considerably (1.38-2.26%) (Table 1).

As a result of HPLC analysis of methanolic extracts from the investigated raw materials eight phenolic compounds were identified (Table 2). Three of them were recognised as flavan-3-ols: (+)-catechin, (-)-epicatechin, and (-)-epigallocatechin (Table 2). The presence of (+)-catechin in silverweed herb has been previously reported by Kombal and Glasl [12]. The raw material obtained from the population 1 was distinguished by highest content of this compound. The population 3 was characterised by significantly lower content of all three catechins in comparison with the populations 1 and 2.

Four flavonol glycosides (rutoside, quercetin-3-O-glucoside, isorhamnetin-3-O-glucoside, and kaempferol-3-O-glucoside), and one phenolic acid (ellagic acid) were also identified in the evaluated raw materials. Kombal and Glasl [12] have found eleven flavonol glycosides in the aerial parts of silverweed grown in Germany, including quercetin-3-O-glucoside and kaempferol-3-O-glucoside. Ellagic acid has been previously reported as a constituent of silverweed herb by Krzaczek [13]. The herb collected from the plants of the population 3 was characterised by significantly lower content of all determined flavonol glycosides and ellagic acid in comparison with the raw materials obtained from the populations 1 and 2. The population 1 was characterised by higher content of rutoside, but lower content of quercetin-3-O-glucoside and ellagic acid than the population 2. Distinctly lower content of all determined phenolic compounds in the herb obtained from the population 3 seems to be a result of light conditions. Plants of this population were shaded by broadleaf trees – silver birch (*Betula pendula* Roth) and black poplar (*Populus nigra* L.).

CONCLUSIONS

- Herb collected from the investigated populations of silverweed differed in the content of determined phenolic compounds.
- Population 3 was distinguished by significantly lower content of all identified compounds in comparison with two others. The most considerable differences concerned the content of (+)-catechin (87.0-199.3 mg × 100 g^{-1}), (-)-epigallocatechin (141.9-290.6 mg × 100 g^{-1}), and rutoside (192.3-386.0 mg × 100 g^{-1}).

MATERIALS AND METHODS

Herb of silverweed was obtained from three populations of this species occurring at the edges of arable fields:

population 1 – near Brańsk (N 52° 44.296´ E 22° 49.397´, 120 m above sea level),

population 2 – in Spieszyn (N 52° 39.683´ E 22° 48.959´, 125 m),

population 3 – in Koryciny-Borki (N 52° 39.412´ E 22° 45.987´, 122 m).

Herb was harvested at the beginning of blooming stage (first ten days of June 2008) and dried at 60°C. The content of three groups of phenolic compounds (flavonoids, phenolic acids, and tannins) in the raw material was determined by spectrophotometric methods, according to Polish Pharmacopoeia [8]. Content of flavonoids was expressed as quercetin equivalents, phenolic acids – as caffeic acid, and tannins – as pyrogallol.

For the separation and identification of individual phenolic compounds 1 g of dry raw material was exhaustively extracted with methanol in Büchi B-811 Extraction System. After evaporation of solvent, the residue was dissolved in 5 ml of methanol, filtered (Supelco IsoDisc PTFE 25 mm 0.45 µm), and subjected to HPLC. The analysis was carried out using a Shimadzu chromatograph with SPD-M10A VP DAD detector and a Supelco LC RP 18 column (5 µm, 250 mm 4.6 mm). The gradient of 10% acetonitrile in water and 55% acetonitrile in water at pH 3 was applied.

The following analysis parameters were used: flow rate 1 ml × min^{-1}, oven temperature 28°C, time of analysis 60 min, recorded wave range: 190-450 nm, detection wave length: 206 nm (flavan-3-ols), 254 nm (rutoside, ellagic acid, quercetin-3-O-glucoside, isorhamnetin-3-O-glucoside), and 264 nm (kaempferol-3-O-glucoside). Peaks were identified by comparison of retention time and spectral data with adequate parameters of standards. Quantification was based on the peak area. The content of the determined compounds was calculated in mg × 100 g^{-1} dry matter.

The results were analysed with ANOVA Tukey's HSD test at the 0.05 significance level in Statgraphics Plus for Windows v. 4.1.

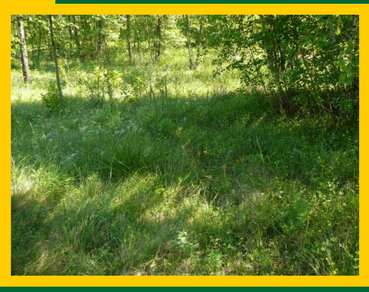
Figure 1. Population 1

near Brańsk
N 52° 44.296´
E 22° 49.397´
120 m above sea level

Figure 2. Population 2

Spieszyn
N 52° 39.683´
E 22° 48.959´
125 m above sea level

Figure 3. Population 3



Koryciny-Borki
N 52° 39.412´
E 22° 45.987´
122 m above sea level

Table 1. Content of determined groups of phenolic compounds in silverweed herb (%)

Group of phenolic compounds	Population 1	2	3
Tannins	0.86 a *	0.91 a	0.81 a
Flavonoids	0.49 a	0.48 a	0.60 b
Phenolic acids	2.26 c	1.60 b	1.38 a

* Values in rows marked with the same letter do not significantly differ at α=0.05 (Tuckey's test)

Table 2. Content of identified phenolic compounds in silverweed herb (mg × 100 g^{-1})

Phenolic compound	Population 1	2	3
(-)-Epigallocatechin	290.6 b*	278.3 b	141.9 a
(+)-Catechin	199.3 c	169.4 b	87.0 a
(-)-Epicatechin	302.3 b	312.8 b	198.1 a
Rutoside	386.0 c	234.7 b	192.3 a
Quercetin-3-O-glucoside	420.2 b	535.9 c	314.1 a
Isorhamnetin-3-O-glucoside	136.9 b	133.5 b	103.3 a
Kaempferol-3-O-glucoside	78.4 b	68.0 ab	58.3 a
Ellagic acid	405.8 b	426.0 c	363.0 a

* Values in rows marked with the same letter do not significantly differ at α=0.05 (Tuckey's test)

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