INVASIVE SOLIDAGO CANADENSIS L. AS A RESOURCE OF VALUABLE BIOLOGICAL COMPOUNDS

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ABSTRACT
The phytochemical characteristics of alien species have not yet been fully studied. Meanwhile, the reserves of their raw materials in the secondary distribution range are very large and can be used as new sources of functional ingredients for food, nutraceuticals, cosmeceuticals, medicines, and other applications. Particular attention is attracted by species which have closely related native plants that are included in the official pharmacopeia. Solidago canadensis L. in Slovakia has already formed powerful thickets, and a similar species Solidago virgaurea L. is used as a medicinal plant. The goal of our study is to examine biologically active compounds from leaves and inflorescences of Solidago canadensis collected in some invasive populations along the Nitra river and Gron river. Leaves and inflorescences of 3 populations have been taken for analysis. In addition, we tested herbal tea was made by a traditional procedure using 2 types of fermentation. The following parameters have been understudying: total dry matter, ash and protein content, total lipid, saccharides, vitamin C content, total carotenoid content, amino acids content, elemental analysis, and antioxidant activity. Mean values and variations of these parameters are given understudying: total dry matter, ash and protein content, total lipid, saccharides, vitamin C content, total carotenoid content, amino acids content, elemental analysis, and antioxidant activity. Mean values and variations of these parameters are given in the article. The results demonstrated that S. canadensis can be a valuable raw material resource for many sectors of the economy with the possibility of its wider application in the future.

Keywords: Solidago canadensis; goldenrod; phytochemical analyses; economic value

INTRODUCTION
Valuable phytochemicals with antioxidant, antimicrobial and other health benefits are synthesized by plants in the process of secondary metabolism. Therefore, medicinal and aromatic plants have been used in health care, food, and cosmetics since ancient times. The importance of herbal medicines in the XXI century increases significantly due to the rapid development of functional foods and nutraceuticals, which became the main trend in food science and technology, as well as in nutrition and disease prevention (Nitrayová et al., 2014; Kraujaliene, Pakalskas and Venskutonis, 2017). Many plant species have not been sufficiently studied from the phytochemical point of view. This suggestion applies specially to alien plant species, the phytochemistry of which in the secondary distribution range may differ significantly, due to a change in the soil and climatic conditions during the expansion the secondary distribution range. Evaluation of widespread invasive species will, perhaps, reveal new sources of functional ingredients for food, nutraceuticals, cosmeceuticals, medicines, and other applications. Invasive species can be a valid resource and instead of eliminating them, there could be a great benefit obtained.

Canadian goldenrod (Solidago canadensis L.) is a highly aggressive alien plant native to North America. Once established, it can reduce biodiversity or locally out-compete all native plants. It is considered as the worst invasive alien weed in Europe (Lambdon, 2008; Vinogradova et al., 2010). Nowadays it is widely spread in China, Russia (from Kaliningrad to the Far East), Japan, Taiwan, Europe and Australia (Waleck et al, 1999; Weber, 2000; Vinogradova, Mayorov and Chroon, 2010). Complex Solidago canadensis is a highly variable and its taxonomic status is not clear and difficult to assess (Semple and Cook, 2006; Melville and Morton, 2011; Vinogradova, Ryabchenko and Mayorov, 2013; Semple et al., 2013; Semple et al., 2015; Vinogradova et al., 2017). Invasive S. canadensis may acquire spreading advantage in non-native habitat by using “novel weapons” to inhibit not only local plants but also soilborne pathogens (Zhang et al., 2009). Thus, invasion success of Solidago canadensis is depended on allelopathic compounds which plant can release (Abilasha et al., 2008). It has been found that acetone extracts of S. canadensis showed allelopathic effects on the growth of other weeds (Solyomyi, 1994).

Numerous organic compounds were reported for the genus Solidago, for example, flavonoids, phenolic acids and glucosides, polysaccharides, diterpenes, triterpenoid saponosides, tannins, and essential oils. These results highlighted the potential of using S. canadensis extracts as...
a natural antimicrobial and antioxidant substances for food applications (Deng et al., 2015). The flowers were used in traditional medicine as an analgesic, burns and ulcers treatment, febrifuge, gastrointestinal and liver aids (Vinogradova and Kuklina, 2012; Zihare and Blumberga, 2017). This species has been used in traditional medicine as a urological and antiphlogistic medicament (Apati et al., 2003). Many herbal preparations exploited in traditional medicine contain a significant number of polysaccharides or their glycoconjugates. It has been found that polyphenolic–polysaccharide–protein complexes isolated from medicinal plants of Asteraceae and Rosaceae families showed anticoagulant activity (Pawlacyzk et al., 2009). Special medicales "Goldenrod" with antispasmodic, diuretic and anti-inflammatory effects is produced in Ukraine and Russia. Available for sale “Prostanorm” – for the treatment of prostate diseases, "Marelin" and "Fitolizin" – from urolithiasis (Vinogradova and Kuklina, 2018).

In terms of bioeconomics, large populations of S. canadensis are important for honey production (Botta-Dukat and Dancza, 2008). Our earlier data confirm the availability of Solidago canadensis: its aerial part contains from 0.1 to 0.7% of essential oil and from 0.1 to 0.4% of essential oil in the inflorescens. A-pinene (1.3 – 61.2%), limonene (0.5 – 22.5%), bornyl acetate (3.4 – 29.8%) and germacrene D (1.8 – 39.2%) were the major compounds detected in oil samples of S. canadensis. Samples from inflorescences contained the maximal percentage of monoterpen hydrocarbons, while the leaves' samples showed the maximal cumulative percentage of sesquiterpene and monoterpen hydrocarbons (Shelepova et al., 2018). The threats and benefits to food production from S. canadensis in the Nitra river basin are described in detail (Fehér et al., 2016).

The aim of this article was to investigate the composition and structural features of the complex isolated from leaves and inflorescence of S. canadensis, growing in Slovakia for to assess the possibility of using this species for bioeconomics in the future.

Scientific hypothesis
Worldwide, works are being carried out the estimation of the content and accumulation of compounds during the growth of Solidago. The main research is done in the field of extracts. Essential oil is the next largest researches object. The scientific hypothesis of this study was to examine the leaves and inflorescences of Solidago canadensis due to its biologically active compounds. They may serve as potential sources of phytochemical compounds in foods and health promoting ingredients for humans.

MATERIAL AND METHODOLOGY

Biological material
Three populations from different regions of Slovakia have been observed: 1) nearby Nitra (N48.3355, E18.0468), 2) along the Gron river nearby Hronovce (N47.9915, E18.6620) and 3) along the Gron river nearby Zvolen (N48.5637, E19.1159). Populations of Solidago are very dense and occupy large areas (Figure 1). Thus, there is no shortage of this plant as a biological resource. Material has been dried in the shade, temperature 20 – 30 °C. Because of our earlier studies proved the minimal concentration of functional ingredients in stems (Vinogradova and Kolesnikov, 2007; Akimova, Kolesnikov and Vinogradova, 2008), only leaves and inflorescences have been taken for analysis.

Preparation of herbal tea from S. canadensis leaves
The tested herbal tea was made by a traditional procedure. Herbal tea infusions were obtained by air-dried and freeze-drying leaves and activation using 2 types of fermentation – fermentation of leaves after deep freezing and fermentation of fresh leaves.

Chemicals
All the chemicals used were of analytical grade and were purchased from Sigma-Aldrich (Steinheim, Germany), Merck (Darmstadt, Germany) and CentralChem (Slovakia).

Phytochemical analyses
Total dry matter, ash and protein content were determined according to EN method (ČSN EN 12145, 1997). Total lipid content was determined according to methods specified in ISO method (ISO 659:1998).

For determination of saccharides, 1 g of sample was extracted with 10 mL of extraction solution (ultrapure water and ethanol mixed in ration 4:1) in a 50 mL centrifugation tube placed on vertical shake table (GFL, Germany). After 1 h of extraction, samples were centrifuged for 4 min at 6000 rpm in a centrifuge (EBA 21, Hettich, Germany); the supernatant was filtered using a filter with 0.45 μm pore size (Labicom, Czech Republic) and filled up to 50 mL in a volumetric flask with ultrapure water. An Agilent Infinity 1260 liquid chromatograph (Agilent Technologies, USA) equipped with ELSD detector was used for determination of saccharides. A Prevail Carbohydrates ES column (250/4.6 mm) was used.
as a stationary phase and acetonitrile (VWR) mixed with water in 75:25 volume ratio was used as the mobile phase. Total carotenoid content expressed as beta-carotene was analysed at a wavelength of 445 nm spectrophotometrically (VIS spectrophotometer UV Jenway Model 6405 UV/VIS). Sample (1 g) was disrupted with sea sand and extracted with acetone until complete discoloration. Petroleum-ether was added and then water, in purpose to the separation of phases. After the separation, the petroleum ether-carotenoid phase was obtained, and the absorbance was measured (ČSN 560053, 1986). HPLC method of L-ascorbic acid (vitamin C) content estimation (Staan, Soran and Marutoiu, 2014) was used by the help of Shimadzu HPLC model LC2010 with PDA detector, for separation was used RP C18 column, mobile phase was methanol: water (5:95, v/v), PDA detector was adjusted to 243 nm. Sample for elemental analysis was prepared using wet ashing method in a microwave oven (Milestone 1200, Milestone, Italy). Total of 0.25 g sample matrix was decomposed in a mixture of nitric acid (6 mL) (Analytika Praha spol. s.r.o., Czech Republic) and hydrochloric acid (2 mL) (Analytika Praha spol. s.r.o., Czech Republic). After the decomposition sample was filtered using a filter with 0.45 µm pore size and filled up to 25 mL in a volumetric flask with ultrapure water. Elemental analysis was performed using ICP-OES (Ultima 2, Horiba Scientific, France) according to the procedure described by Divis et al. (2015).

Amino acids were determined by ion-exchange liquid chromatography (Model AAA-400 amino acid analyser, Ingos, Czech Republic) using post-column derivatization with ninhydrin and a VIS detector. A glass column (inner diameter 3.7 mm, length 350 mm) was filled manually with a strong cation exchanger in the LG ANB sodium cycle (Laboratory of Spolchemie) with average particles size 12 µM and 8% porosity. The column was tempered within the range 35 to 95 °C. Elution of the studied amino acids took place at a column temperature set to 74 °C. A double-channel VIS detector with the inner cell volume of 5 µL was set to two wavelengths: 440 and 570 nm. A solution of ninhydrin was stored in an inert atmosphere (N₂) in darkness at 4 °C. The flow rate was 0.25 mL·min⁻¹ and the reactor temperature was 120 °C.

**Antiradical activity**

Biochemical analysis of antioxidant activity detection was conducted according to Brand-Williams, Cuvelier and Berset (1995). Plant extracts were prepared in following solvents: methanol, ethanol and distilled water. 1 g of dry powder of plant sample was mixed with 25 mL of each solvent. Extraction was carried out during 12 hours at continuous stirring. Preparing of the radical solution was following: 25 mg of DPPH-radical (2,2-diphenyl-2-picrylhydrazyl) was solved in methanol (in 100 mL volumetric flask) and used for following dilution (1:10). 0.1 mL of investigated plant extract was added to 3.9 mL of a radical solution. The optical density of the radical solution was measured immediately and after 10 min of incubation in the dark after adding a sample. The measurement was conducted at 515 nm on the spectrophotometer (Genesis 20, Germany). Obtained data calculated using a formula:

\[
\% \text{ Inh} = \frac{A_0 - A_1}{A_0} \times 100
\]

**Statistical analysis**

Student t-test was used for the statistical analysis of the obtained results. Data are presented as mean ± standard error of the mean (SEM). p < 0.05 was considered statistically significant. Significance of p < 0.05 and p < 0.01 is shown by one and two asterisks, respectively.

**RESULTS AND DISCUSSION**

Phytochemicals possess various bioactivities and may serve as potential sources of antioxidants in foods and health promoting ingredients in humans. Chemical analyses of Solidago canadensis leaves and inflorescences revealed the presence of protein (12.2 wt/wt%), carbohydrates (4 wt/wt%), lipids (5.1 wt/wt %) and inorganic material (6.7 wt/wt%) (Table 1a and 1b). Monosaccharide analysis of neutral carbohydrate part showed the presence of one main sugar – fructose (8.8 g·kg⁻¹), while other saccharides as maltose, sucrose, and lactose were found in low amounts only (<0.5 g·kg⁻¹). However, this may not always be the case. So, according to Štová et al. (2013), S. canadensis complex (hot alkaline extraction of flowers) revealed the presence of harnemose (~23 wt/wt%), arabinoxyl (~20 wt/wt%), galactose (~17 wt/wt) and glucose (~14 wt/wt%). In our study, we did not detect these monosaccharides.

S. canadensis contains vitamin C (L-ascorbic acid (1.53 µmol·g⁻¹)) and beta carotene (93.9 mg·kg⁻¹) (Table 1a and 1b). The vitamin E family includes four tocopherols, α-tocopherol being the most potent member of this family (Shahidi and Ambigaipalan, 2015). The major quantititative tocopherol in S. canadensis leaves and inflorescences was α-tocopherol (48.8 ±3.2 mg·kg⁻¹ DWP). It is in agreement with Kraujaliene, Pukalskas and Venskutonis (2017). Vitamin E is known as a reducing agent and may contribute to antioxidant activity by reducing the oxidized state of the phenolic antioxidant compounds. Thus, these compounds may be regenerated, allowing their antioxidant capacity to increase. Carotenoids have been reported to show chain-breaking antioxidant activity. These compounds are also effective singlet oxygen quenchers (Demir, 2009).

Lipids are another important group of S. canadensis phytochemicals. The oil contents were 5.1% dry weight plant material (Table 1a and 1b). It is reasonable because the majority of botanical materials (leaves and inflorescences) contain low amounts of lipids. The fatty acids composition was comprised of saturated fatty, monounsaturated and polyunsaturated fatty acids (34.0; 14.3 and 44.8 g.100g⁻¹ oil, respectively). The lipophilic fraction contains of 20 fatty acids; four acids (linoleic acid, C-18:2; oleic acid, C-18:1; palmitic acid, C-16, linolenic acid, C-18:3) is dominated. Of these acids, the first two, the unsaturated C-18 acids, amounted to 57.2% of the total (Figure 2).
The contents of some phytochemical compounds in leaves and inflorescences of *Solidago canadensis* L.

### Table 1

<table>
<thead>
<tr>
<th>Components</th>
<th>mean ±SD</th>
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<th>mean ±SD</th>
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</thead>
<tbody>
<tr>
<td>Total dry matter (%)</td>
<td>91.07 ±1.14</td>
<td>Saturated fatty acids (g.100g⁻¹ oil)</td>
<td>34.0 ±1.0</td>
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<tr>
<td>Total content of protein (%)</td>
<td>12.15 ±0.74</td>
<td>Monounsaturated fatty acids (g.100g⁻¹ oil)</td>
<td>14.3 ±0.80</td>
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<td>Total content of ash (%)</td>
<td>6.68 ±0.18</td>
<td>Polyunsaturated fatty acids (g.100g⁻¹ oil)</td>
<td>44.8 ±2.10</td>
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<tr>
<td>Total content of lipids (%)</td>
<td>5.11 ±0.11</td>
<td>Fructose (g.kg⁻¹)</td>
<td>8.8 ±0.50</td>
</tr>
<tr>
<td>Beta carotene (mg.kg⁻¹)</td>
<td>93.9 ±5.30</td>
<td>Maltose (g.kg⁻¹)</td>
<td>&lt;0.50</td>
</tr>
</tbody>
</table>

Note: mean – arithmetic mean; SD – standard error of the mean.

### Table 2

<table>
<thead>
<tr>
<th>Components</th>
<th>mean ±SD</th>
<th>Components</th>
<th>mean ±SD</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sucrose (g.kg⁻¹)</td>
<td>&lt;0.50</td>
<td>Lactose (g.kg⁻¹)</td>
<td>&lt;0.50</td>
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<tr>
<td>Vitamin A (retinyl acetate) (mg.kg⁻¹)</td>
<td>&lt;0.10</td>
<td>Vitamin E (α-tocopherol) (mg.kg⁻¹)</td>
<td>48.8 ±3.20</td>
</tr>
<tr>
<td>L-ascorbic acid (vitamin C) (µmol.g⁻¹)</td>
<td>1.53 ±0.01</td>
<td></td>
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</table>

Note: mean – arithmetic mean; SD – standard error of the mean.

### Table 3

<table>
<thead>
<tr>
<th>Specimen</th>
<th>Methanol extracts, %</th>
<th>Ethanol extracts, %</th>
<th>Water extracts, %</th>
</tr>
</thead>
<tbody>
<tr>
<td>Freeze-drying leaves</td>
<td>54.66 ±1.09</td>
<td>35.67 ±0.48</td>
<td>34.92 ±1.04</td>
</tr>
<tr>
<td>Air-dried leaves</td>
<td>83.17 ±0.52</td>
<td>83.78 ±0.62</td>
<td>18.59 ±0.84</td>
</tr>
<tr>
<td>Fermentation of fresh leaves</td>
<td>76.32 ±1.15</td>
<td>55.55 ±1.25</td>
<td>66.91 ±2.52</td>
</tr>
<tr>
<td>Fermentation of leaves after deep freezing</td>
<td>74.78 ±2.83</td>
<td>56.80 ±0.65</td>
<td>58.20 ±1.05</td>
</tr>
</tbody>
</table>

Figure 2: Fatty acid composition from leaves and inflorescences of *Solidago canadensis* L. (g.100g⁻¹ oil). Note: Minor components (<0.1): Eicosene C20:1, Arachidonic C20:4, Erucic C22:1, Lignoceric C24:0, Tetracosenoic C24:1, Heptadecanoic C17:1 and Dicosadiene C22:2 are in the right column, their total amount is 0.7 g.100g⁻¹ oil.

Table 3: Antioxidant activity of experimental herbal tea from *Solidago canadensis* L. leaves by DPPH method.


S. canadensis fatty acid profiles showed the presence of high amounts of linoleic acid (35.5%) (Figure 2). From linoleic acid is synthesized γ-linolenic acid and is the first intermediate in the conversion of linoleic acid to arachidonic acid. γ-linolenic acid prevents or alleviates a wide variety of human diseases, and it is important as a dietary and cosmetic component. Palmitic acid is the second quantitatively major compound (21.7%).

Amino acid analysis has shown that the studied S. canadensis leaves and inflorescences contained 18 amino acids (10 essential and 8 non-essential) (Figure 3). Aspartic acid was found to be the dominant free amino acid (12.2 g.kg\(^{-1}\)) followed by a proline (11.0 g.kg\(^{-1}\) and glutamic acid (10.9 g.kg\(^{-1}\)), respectively. Proline content of S. canadensis inflorescences could be one of the parameters in the determination of S. canadensis honey authenticity and could be a good indicator of the botanical origin of honey. Amino acids found in high levels were: lysine, leucine, valine, arginine, phenylalanine – average content in from 7.1 to over 5.1 g.kg\(^{-1}\). Lower but also important amounts of glycine, isoleucine, threonine, serine, alanine, histidine, tyrosine, cystine, methionine, and tryptophan (ranging from 1.5 to 4.9 g.kg\(^{-1}\)) were detected in the study. The total number of amino acids was 97.10 g.kg\(^{-1}\) DM, including total essential amino acids (45.60 g.kg\(^{-1}\) DM) and percentage of total essential amino acids (46.96%).

The results of the elemental analysis of S. canadensis leaves and inflorescences are summarized in Table 2; these values are averages of three independent measurements having ±SD. Concentration of various elements decreases in the order: K>Ca>P>S>Mg>Zn>Mn>Fe>Cu>Na>Al>Ni>As>Cr>Se>Pb>Cd>Hg.

Among the various elements As, Cr, Se, Pb, Cd, Hg are found to be present at the trace level. Zn, Mn, Fe, Cu, Na, Al, and Ni are at the minor level and K, Cu, P, S and Mg are at the major levels. This result is supported by the results of Pytlakowska et al. (2012).

We note here that our focus is not to compare but rather to explore the mineral composition in the selected plants and to discuss their importance in human health. Due to a deficiency of these minerals in the human diet, most of these minerals are often taken as supplements (Agarwal et al., 2011) for their important role in human health.

In fact, the chemical constituents present in plants are responsible for their medicinal as well as toxic properties which include vegetable bases comprising of alkaloids and amines, glycosides, essential oils responsible for their characteristic odor, toxic substances known as toxalbumin, resins, and antibiotics. Whereby the trace elements play a very important role in the formation of these compounds.

One important factor for the formation of active constituents in plants are the trace elements because they are known to play an important role in plant metabolism and active constituents of medicinal plants are metabolic products of plant cells. Among the various elements estimated, the potassium and sodium ions play an important role in the diseases related to renal disorder. Potassium and sodium salts are partially responsible for the diuretic action of some drugs. Potassium is readily excreted by the kidneys both by glomerular filtration and by tubular excretion. Potassium salts act as osmotic diuretics and deficiency of potassium causes diabetic acidosis. Calcium ion concentration also plays an important role in the urinary tract system. Hypercalcemia causes renal failure and calcium stones in the urinary tract. Iron deficiency is common in uremic patients, it causes substantial blood losses. Iron may bind to the dialyzer membrane. Some reports indicate that dysgeusia, poor food intake, and impaired sexual function, which are common problems of uremic patients, may be improved by zinc supplements (Rajurkar and Damane, 1998). Magnesium is reported to have a curative effect in more than 300 health disorders, including headache and fatigue (Daur, 2015). Mn ameliorates some of the symptoms of diabetes and plays a role in the function of connective tissue, bones, and blood clotting factors Similarly Cr, Mn, Fe, Co, Cu, and Zn have been reported as essential or beneficial to human health (Kozlowska et al., 2015).

The total antioxidant activity of extracts from freeze-drying leaves of S. canadensis was not high and amounted to 34.9% (aqueous extracts), 35.7% (ethanol extracts) and 54.7% (methanol extracts) (Table 3). While the total antioxidant activity of extracts from air-dry leaves was significantly higher for methanol and ethanol extracts and amounted to 83.2 and 85.8%, respectively. But it was lower for the water extract – 18.6%. The total antioxidant activity of the herbal tea samples from the 2 types of fermentation was generally similar and significantly higher than the values are averages of three independent measurements having ±SD. Concentration of various elements decreases in the order: K>Ca>P>S>Mg>Zn>Mn>Fe>Cu>Na>Al>Ni>As>Cr>Se>Pb>Cd>Hg.

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Figure 3 Amino acid composition from leaves and inflorescences of Solidago canadensis L. (g.kg\(^{-1}\) DM).
native samples for aqueous extracts (66.9 – 58.2%). The indices of alcoholic extracts of herbal tea were lower than those for dry leaves but higher than those of native samples (55.6 – 56.8% (ethanol extract) and 74.8 – 76.6% (methanol extract)). Deng et al. (2015) determined antioxidant activity from leaves and bark of S. canadensis using DPPH radical. The capacity ranged from 25.3 – 37.2% (ultrasound-assisted extraction) to 18.8 – 21.1% (ethanol extraction) by DPPH method. Demir et al. (2009) reported results antioxidant capacity of young shoots with leaves for S. virgaurea using DPPH method. The antioxidant capacity ranged from 30.7% (aqueous extracts) to 64.3% (methanol extract) of DPPH inhibition.

CONCLUSION

The results demonstrated that S. canadensis can be a valuable raw material resource for many sectors of the economy. Many high added value products can be obtained from this species. This proves the value of S. canadensis for the bioeconomy and the possibility of its wider application in the future.

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ISO 659:1998. Oil seeds determination of hexane extract (or light petroleum extract), called “oil content”.


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