EVALUATION AND COMPARISON OF THE CONTENT OF TOTAL POLYPHENOLS AND ANTIOXIDANT ACTIVITY IN ONION, GARLIC AND LEEK

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ABSTRACT
Onion, leek and garlic as onion family are a great source of freely available health-promoting and chemoprotective compounds (polyphenols, flavonoids, fructooligosaccharides, thiosulfimates and other sulfur compounds, vitamins). Chemoprotective compounds belong between natural components. Onion, garlic and leek have high nutritional value. They are an important component of our human diet and we used them as a preventive factor for many diseases of civilization (cancer, coronary heart diseases, and atherosclerosis). In this work we watched and evaluated content of total polyphenols and antioxidant activity in onion, garlic and leek. Samples of plant material (onion, garlic and leek) we collected at the stage of full maturity in the area of Pružina. Pružina is area without negative influences and emission sources. Samples of fresh onion, garlic and leek were homogenized and were prepared an extract: 25 g cut onion, garlic and leek extracted by 50 ml 80% ethanol according sixteen hours. These extracts were used for analyses. The content of the total polyphenols was determined by using the Folin-Ciocalteu reagent (FCR). Antioxidant activity was measured by using a compound DPPH’ (2,2-diphenyl-1-picrylhydrazyl). In the present experiment it was detected, that total polyphenols content in samples ranges from 210.67 mg/kg (leek) - 429.58 mg/kg (onion). Statistically significant highest value of total polyphenols was recorded in onion (389.64 to 429.58 mg/kg). Statistically significant the lowest content of total polyphenols was recorded in leek (201.67 - 254.80 mg/kg). Another indicator that has been evaluated and compared was the antioxidant activity of onion, garlic and leek. Statistically significant highest value of antioxidant activity was recorded in onion (20.22 - 25.76%). Statistically significant the lowest value of antioxidant activity was recorded in garlic (4.05 - 5.07%). Based on the measured values of AOA in onion, garlic and leek samples can be classified as follows: onion (20.22 - 25.76%) > leek (8.55 - 12.92%) > garlic (4.05 - 5.07%).

Keywords: 5 Keywords: onion; garlic; leek; total polyphenols; antioxidant activity

INTRODUCTION
Vegetables are important components in the human diet and protect against many diseases of civilization especially cardiovascular diseases, high blood pressure, stroke, neurodegeneration, cancer, obesity and diabetes. Family belongs Liliaceae among the most important vegetable observation and demonstrated effects on human organism.

Onions, garlic, leeks, shallots and other members of the onion family are commonly grown in fresh market gardens. All plants in the Allium are herbaceous, biennial vegetables that are known as annuals. Plants in the family Alliaceae are derived their characteristic flavor from the enzyme alliinase that acts on sulfur compounds. Alliums with their shallow roots system, grown the best in well-prepared soil (Delamaunt, 2003).

Onion (Allium cepa var. cepa) is member of the lily family or Amaryllidaceae and it is classified by the scientific name Allium cepa. Onions (Allium cepa var. cepa) are an important source of bioactive compounds including phenolic compounds, flavonoids, fructooligosaccharides (FOS), thiosulfimates and other sulfur compounds, and many of these compounds have potential beneficial properties for human health Soinen et al., (2012). Phenolic compounds in onions, garlics and leeks are an essential part of the human diet and they are of considerable interest due to their antioxidant properties. These compounds possess an aromatic ring bearing one or more hydroxyl groups and their structures may range from that of a simple phenolic molecule to that of a complex high-molecular weight polymer Balasundram et al., (2005). Polyphenols are reported to in vitro inhibit cancer cell proliferation, reduce vascularization, protect neurons, stimulate vasodilation and improve insulin secretion Rio-Del et al., (2010).

Onion (Allium cepa var. cepa) represents a source of cysteine derivatives, which makes it a good antioxidant additive for food Ostrowska et al., (2004). Antioxidants are important components, which they protect against free radicals. Free radicals are known to be the major contributors to degenerative diseases of aging and they are recognised as major factors causing cancer, cardiovascular disorders and diabetes Calucci et al., (2003).

Garlic (Allium sativum var. sativum) belongs among the oldest of all cultivated plants. It has been used as a medicinal agent for thousands of years. It is a remarkable plant, which has multiple beneficial effects.
such as antimicrobial, antithrombotic, hypolipidemic, antiarthritic, hypoglycemic and antitumor activity.

Recently, it has been observed that aged garlic extract exhibited radical scavenging activity. The two major compounds in garlic, S-allylcysteine and S-allylimercapto-L-cysteine, have the highest radical scavenging activity therefore the consumption of garlic may provide some kind of protection from cancer development Thomson et al., (2003).

Leeks (Allium ampeloprasum var. porrum) are robust winter hardy biennials that do not form a hard bulb like onions or garlic. Leeks are sweeter than onions and have a creamy texture when cooked (Delamaunt, 2003).

Leeks (Allium ampeloprasum var. porrum) contain vitamin C, vitamin B, vitamin E, copper, potassium, iron, carotenoids, chlorophyll (green tops) and flavonoids. The major flavonoid in leek is kaempferol, with only a small amount of quercetin like spring onions, leeks contain carotenoids and chlorophyll mainly in the green tops.

MATERIAL AND METHODOLOGY

Samples of plant material were collected at full maturity stages from area of Pružina. The samples of soil and plant material were analyzed individually by selected methodologies, and were used fresh material on analysis. Pružina is located under the Strážovské hills - Strážov. The attitude of the village is in the middle of 381 m.a.s.l. Pružina belongs to the mild cold climate zone, average annual air temperature is 7 °C, annual rainfall is 800 - 1000 mm. Pružina is area without negative influences, emission sources (carbon), relatively pure from content of risk permissible forms point of view (Table 1 and Table 2). Samples of fresh onion, garlic and leek were homogenized and were prepared an extract: 25 g cut onion, garlic and leek extracted by 50 ml 80% ethanol according sixteen hours. These extracts were use for analyze.

Determination of total polyphenols

Total polyphenols were determined by the method of Lachman, et al., (2003) and expressed as mg of gallic acid equivalent per kg fresh mater. Gallic acid is usually used as a standard unit for phenolics content determination because a wide spectrum of phenolic compounds. The total polyphenol content was estimated using Folin-Ciocalteau assay. The Folin-Ciocalteau phenol reagent was added to a volumetric flask containing 100 µL of extract. The content was mixed and 5 mL of a sodium carbonate solution (20%) was added after 3 min. The volume was adjusted to 50 mL by adding of distilled water. After 2 hours, the samples were centrifuged for 10 min. and the absorbance was measured at 765 nm of wavelength against blank. The concentration of polyphenols was calculated from a standard curve plotted with known concentration of gallic acid.

Determination of antioxidant activity

Antioxidant activity was measured by the Brand and Williams et al., (1995) method-using a compound DPPH• (2,2-diphenyl-1-pikrylhidrazyl)). 2,2-diphenyl-1-pikrylhydrazyl (DPPH•) was pipetted to cuvette (3.9 m³) then the value of absorbance, which corresponded to the initial concentration of DPPH• solution in time Ao was written. Then 0.1 cm² of the followed solution was added and then the dependence A = f(t) was immediately started to measure. The absorbance of 1, 5 and 10 minutes at 515.6 nm in the spectrophotometer Shimadzu UV/VIS-1240 was mixed and measured. The percentage of inhibition reflects how antioxidant compound are able to remove DPPH• radical at the given time. Inhibition (%) = (Ao - At / Ao) x 100

RESULTS AND DISCUSSION

Onion, garlic and leek belong among the most valuable kinds of vegetables. They are rich on polyphenols, flavonoids, vitamins and protein. It also contains of phosphorus, calcium and carbohydrates. Plants in the onion family have antihelmintic, antioxidant, antiinflammatory and antiseptic effects. The total content of polyphenolic compounds contained in onion, garlic and leek are quite variable, may be affected by post-harvest and climatic conditions. In this work the content of total polyphenols in onion, garlic and leek was watched and evaluated.

In the present experiment it was detected, that total polyphenols content in samples ranges from 210.67 ±16.63 (in variety of leek) to 429.58 ±17.44 (in variety of onion). Statistically significant highest value of total polyphenols was recorded in onion (variety of Lusy, Štugarská and Všetana). Statistically significant the lowest content of total polyphenols was recorded in

Table 1 Agrochemical characteristic of soil substrate in mg/kg

| pH (H₂O) | pH (KCl) | Cox (%) | Humus (%) |
|----------|----------|---------|-----------|
| 7.70     | 5.60     | 1.38    | 2.39      |

Table 2 Agrochemical characteristic of soil substrate in mg/kg (heavy metals)

| Heavy metals | Zn | Cu | Mn | Fe | Cr | Cd | Pb | Co |
|--------------|----|----|----|----|----|----|----|----|
| aqua regia   | 85.00 | 22.4 | 615.0 | 2642 | 34.6 | 1.28 | 24.40 | 14.0 |
| limit value  | **100.00** | **60.00** | - | - | **70.00** | **0.40** | **70.00** | **15.00** |
| HNO₃(c=2 mol.dm⁻³) | 6.50 | 10.50 | 454 | 1500 | 0.50 | 0.14 | 3.10 |
leek (variety of Arkansas, Nobel, Hannibal). Andrejiová et al., (2011) said that the content of total polyphenols in onion was in the interval from 105 to 134 mg/kg. Armand et al., (2012) reported that the content of polyphenols in onion was 620 mg/kg. Our values of total polyphenols in onion were in the range from 389.64 ±20.31 mg/kg to 429.58 ±17.44 mg/kg. Polyphenolic compounds have antioxidant properties and play an important role in preventing many of lifestyle diseases. In the work the content of total polyphenols in leek and garlic was also watched. Our values of total polyphenols in leek were 210.67 ±16.63 mg/kg to 254.80 ±10.09 mg/kg.

In the case of garlic were measured lower levels of content of polyphenols (260.62 ±25.56 mg/kg to 279.74 ±16.18 mg/kg) than in onion. Priecina et al., (2013) reported that the polyphenols in garlic was in the amounts from 272.28 to 1818.81 mg/kg. From the results there is obvious that the highest content of total polyphenols is characteristic for onion (389.64 ±20.31 mg/kg to 429.58 ±17.44 mg/kg), followed by garlic with the measured value of 260.62 ±25.56 mg/kg to 279.74 ±16.18 mg/kg. While the lowest content of total polyphenols was recorded in leek (210.67 ±16.63 mg/kg to 254.80 ±10.09 mg/kg. Polyphenolic compounds contained in onions, garlic and leeks are one of the largest categories of phytonutrients, it is essentially the most represented group of secondary plant metabolites. Another indicator that has been evaluated and compared was the antioxidant activity of onion, garlic and leek. The antioxidant activity of vegetables is often associated with a ß-carotene, L-ascorbic acid, vitamin E and present polyphenolic compound such as quercetin, rutin.

**Table 3** Average content of total polyphenols (mg/kg) in onion, garlic, leek.

| vegetable | variety  | TPC (mg/kg) |
|-----------|----------|-------------|
| onion     | Lusy     | 389.64 ±20.3 d |
|           | Všetana  | 396.71 ±18.10 de |
|           | Štutgarská | 429.58 ±17.44 e |
| garlic    | Makoi    | 260.62 ±25.56 bc |
|           | Mirka    | 268.09 ±16.61 c |
|           | Matin    | 279.74 ±16.18 c |
| leek      | Arkansas | 225.51 ±20.04 ab |
|           | Nobel    | 210.67 ±16.63 a |
|           | Hannibal | 254.80 ±10.09 bc |
|           | HD<sub>0.05</sub> | 26.6589 |
|           | HD<sub>0.01</sub> | 35.9988 |

**Table 4** Average values of antioxidant activity (% inhibition) in onion, garlic, leek.

| vegetable | variety  | AOA (%) |
|-----------|----------|---------|
| onion     | Lusy     | 25.76 ±0.53 h |
|           | Všetana  | 23.09 ±0.79 g |
|           | Štutgarská | 20.22 ±0.53 f |
| garlic    | Makoi    | 4.45 ±0.28 ab |
|           | Mirka    | 5.07 ±0.47 b |
|           | Matin    | 4.05 ±0.20 a |
| leek      | Arkansas | 8.55 ±0.54 c |
|           | Nobel    | 10.82 ±0.53d |
|           | Hannibal | 12.92 ±0.66 e |
|           | HD<sub>0.05</sub> | 0.768657 |
|           | HD<sub>0.01</sub> | 1.037950 |
In the present work it was detected, that antioxidant activity in samples ranges from 4.05% ±0.20% (garlic) to 25.76% ±0.53% (onion). Statistically significant highest value of antioxidant activity was recorded in onion (20.22% ±0.53% to 25.76% ±0.53%). Statistically significant the lowest value of antioxidant activity was recorded in garlic (4.05% ±0.20% to 5.07% ±0.47%).

Prakash et al., (2007) published that the value of antioxidant activity in onion was in interval from 3.6% to 84.1%. Our results of antioxidant activity in onion was higher than the results Karadeniz et al., (2005), which found that the value of antioxidant activity was 12.5%.

In the case of garlic was measured the lowest level of antioxidant activity (in interval from 4.05% ±0.20% to 5.07% ±0.47%) than in onion. Priecina et al., (2013) reported that value of antioxidant activity in garlic was 11.98%. Based on the measured values of AOA in onion, garlic and leek can be samples classified as follows: onion > leek > garlic. Antioxidants in onion, garlic and leek catch reactive form of oxygen, inhibition of enzymes responsible for superoxide anion production, chelation of transition metals involved in process forming radicals and prevention of the peroxidation process by reducing alcoxyl and peroxyl radicals (Biesaga, 2011).
CONCLUSION
The contribution focuses on the total of polyphenol content and antioxidant activity in onion, garlic and leek. The results suggest that the highest value of total polyphenols and antioxidant activity was in onion. In generally these vegetables are also characterized by a high content of chemoprotective compounds (polyphenols, flavonoids, anthocyanins, quercetin, and many more) and minerals (zinc, calcium, iron, selenium, and phosphorus), which positively affecting the human body. The content of total polyphenols and antioxidant activity may be affected by many factors for example postharvest (storage) and climatic conditions (altitude, rainfall, mean annual temperature) and the agrochemical composition of the soil (humus of content, nutrients) and type varieties.

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Acknowledgments:
This work was supported by grant VEGA 1/0290/14, VEGA 1/0456/12.

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