ISSN 1644-0692 www.acta.media.pl

ARUM POLO <sup>2</sup> Acta Sci. Pol. Hortorum Cultus, 14(5) 2015, 41-53

# **BIOLOGICAL VALUE OF Eruca sativa Mill. LEAVES** UNDER THE DIFFERENT PLANT NUTRITION **BY NITROGEN AND POTASSIUM**

Renata Nurzyńska-Wierdak

University of Life Sciences in Lublin

Abstract. Vegetables have a significant place in the healthy eating pyramid. It is recommended that vegetables should be consumed as often as possible to provide nutritional and biologically active substances. The aim of the present study was to determine the biological value of the leaves of rocket as affected by different regimes of plant nitrogen and potassium nutrition. Plants were grown in a peat-based medium under greenhouse conditions. After harvest, the contents of L-ascorbic acid, chlorophyll, flavonoids, glucosinolates and essential oil were determined in the rocket leaves. The average content of chlorophyll a + b per 100 g of rocket fresh biomass was 1.16 mg, while the L-ascorbic acid concentration - 92.66 mg. 100 g of dried rocket leaves contained on average 0.84 g of flavonoids and 0.15 ml of essential oil, while 1 g of dry plant material was characterized by the presence of 10.56 µmol of glucosinolates, on average. Potassium chloride proved to be an interesting source of potassium; its application significantly increased the concentration of glucosinolates in the rocket leaves. However, this form of potassium was not found to have a significant effect on the accumulation of L-ascorbic acid, chlorophyll, flavonoids, and essential oil. An increased rate of nitrogen contributed to a decrease in the content of L-ascorbic acid and glucosinolates. The presented results show that it is possible to modify the chemical composition of rocket leaves by using an appropriate system of plant mineral nutrition.

Key words: Brassicaceae, vegetables, rocket, active substances, L-ascorbic acid

# **INTRODUCTION**

Proper nutrition is a direct factor that guarantees the maintenance of well-being and health. At the same time, the interest in the preservation of good health and in the slowing down of aging processes is growing among an ever larger group of consumers. As

Corresponding author: Renata Nurzyńska-Wierdak, Department of Vegetable Crops and Medicinal Plants, Faculty of Horticulture and Landscape Architecture, University of Life Sciences in Lublin, Poland, e-mail: renata.nurzynska@up.lublin.pl

<sup>©</sup> Copyright by Wydawnictwo Uniwersytetu Przyrodniczego w Lublinie, Lublin 2015

a result of that, the demand for, among others, food with the desired health-promoting effects on the human body is continually increasing. The concept of functional food originates from philosophical tradition of the Far East in which there is no clear distinction between a medicine and food. Functional foods are specially designed food products that show a documented beneficial effect on health beyond the one that results from the presence in such food of nutritional components which are traditionally considered to be necessary. The latest scientific research focused on the issues associated with the structure of active compounds in edible plants and their pharmacological properties leads to the development of functional food and nutraceuticals [Kopeć et al. 2015, Liu et al. 2015].

Vegetables are important dietary components due to the presence of mineral compounds, dietary fibre, sugars, and protein [Žnidarčič et al. 2013, Kadivec et al. 2015, Sinkovič et al. 2015]. In addition to the above-mentioned nutritional constituents, they contain in their tissues different bioactive components, such as vitamins, flavonoids, glycosides, chlorophyll, or essential oils. The biosynthesis of these substances, associated with different variation factors, determines the biological activity of a plant material [Aires et al. 2011, Narits 2011]. Many commercial plant species native to the Mediterranean region, rich in nutritional and biologically active components, show healthpromoting properties [Vardavas et al. 2006, Vig et al. 2009, Michael et al. 2011, Srianta et al. 2012, Strgar et el. 2013]. The effect of Mediterranean diet (MD) is particularly evident in the prevention of heart and cardiovascular diseases as well as tumors. The identification of MD active components is important for developing proper nutrition guidelines [Ortega 2006]. Green leafy vegetables, characterized by high antioxidant activity, are now perceived as important nutraceuticals and pharmacological products [Saeed et al. 2012, Romojaro et al. 2013, Yadav et al. 2013, Patricia et al. 2014].

In Europe rocket (*Eruca sativa* Mill.) is primarily grown as a salad plant with extremely aromatic leaves harvested throughout the entire growing season. The peculiar aroma of rocket is a consequence of the presence of glucosinolates: glucoerucin, glucoraphanin, gluociberin, and glucocochlearin, synthesized from phenylalanine and tyrosine, the precursor of which is methionine, while the sulphur atom is derived from cysteine [Fahey et al. 2001, Falk et al. 2004, Chun et al. 2013]. Some glucosinolate degradation products probably participate in the development of enzymatic systems in human organism which are directed against carcinogenic factors. It is probable that some glucosinolate hydrolysis products have "Janus properties": both cancerogenic and chemoprotective. Janus carcinogens are considered to be carcinogenic compounds which under various conditions (tissue type or dose) can instead act as anticarcinogens [Polat 2010]. Moreover, glucosinolates exhibit bactericidal, fungicidal, nematicidal, allelopathic, antioxidant, and antimutagenic activity [Fahey et al. 2001, Falk et al. 2004, Vig et al. 2009, Khoobchandani et al. 2010]. The antimicrobial activity of rocket leaves is not associated only with the presence of glucosinolates, but also other phytochemical compounds [Filocamo et al. 2010]. The latest research shows [Michael et al. 2011] that the flavonoids in rocket may prove to be a potential factor of antitumor protection. As a result of the presence of vitamin C, tocopherols, lutein, β-carotene, glucosinolates, and phenolics in rocket leaves, they exhibit antioxidant activity [Kim and Ishii 2004, Vardavas et al. 2006]. Due to their chemical composition and biological activity, rocket leaves are considered to be potential nutritional and health- promoting powerhouses [Bukhsh et al. 2007, Tassi and Amaya-Farfan 2008, Filocamo et al. 2010, Michael et al. 2011, Villatoro-Pulido et al. 2012].

The biosynthesis of bioactive substances shows genetic, ontogenetic and environmental variation [Bennett et al. 2006, De Pascale et al. 2007]. Turnip plants accumulated in their roots more glucoraphanin during the spring-summer season than in the summer-autumn and autumn-winter seasons [Zhang et al., 2008]. Environmental stresses may increase the concentration of glucosinolates, L-ascorbic acid and phenolic compounds in Brassica plants, but they do not increase the yield of these compounds per unit area [Antonious i in. 2009]. The content and chemical profile of some biosubstances can also be modified by agronomic factors, among which the level of mineral nutrition is particularly evident [Michałojć and Dzida 2012, Nurzyńska-Wierdak et al. 2012]. The glucosinolates in oilseed rape exhibit a linear relationship with the level of sulphur [Hassan et al. 2007]. Similarly to rocket [Singh et al. 1999], the application of sulphur increased the content of protein, sinigrin and glucosinolates in its seed. Furthermore, sulphur fertilization significantly increased the antioxidant activity of turnip, which was associated with a genotypically varying reduction in nitrate content in turnip leaves [De Pascale et al. 2007]. The biosynthesis of glucosinolates in rocket is strictly related to nitrogen nutrition and the differences caused by the form of nitrogen used most probably result from two pathways of their formation. Moreover, Nartis [2011] showed that higher nitrogen application increased the glucosinolate content in agrimony seeds, while the rate of nitrogen did not have a significant effect. Similar relationships were found for other plant substances. A higher rate of nitrogen contributed to an increase in dry matter production and in the level of chlorophyll in mustard green plants [Vyas et al. 1995]. The aim of the present study was to determine the potential biological value of the leaves of rocket (Eruca sativa Mill.) as affected by different regimes of plant nitrogen and potassium nutrition.

### MATERIAL AND METHODS

The present study was conducted over the period 2010–2012 by carrying out a plant growth experiment in a detached heated greenhouse, situated in the north-south direction and belonging to the Department of Vegetable Crops and Medicinal Plants of the University of Life Sciences in Lublin (Poland). The study material consisted of garden rocket (*Eruca sativa* Mill., Brassicaceae) plants. Seeds acquired from a seed production company, PNOS Ożarów Mazowiecki, were sown individually in 2 dm<sup>3</sup> pots filled with a peat-based medium with a pH of 6.5. The experiment was set up as a completely randomized design in 14 replicates; one pot in which 3 plants were grown was one replicate. The seeds were seeded around 20 March, having been first dressed with Funaben T (5 g kg<sup>-1</sup> seed). The experiment used the following plant mineral nutrition regime: two rates (g dm<sup>-3</sup> medium) of nitrogen applied as Ca(NO<sub>3</sub>)<sub>2</sub>: 0.3 N (including 0.37 Ca); 0.6 N (including 0.74 Ca), as well as three rates of potassium applied as K<sub>2</sub>SO<sub>4</sub>: 0.3 K (including 0.27 Cl); 0.6 K (including 0.54 Cl); 0.9 K (including 0.81 Cl); P – 0.4;

Hortorum Cultus 14(5) 2015

Mg – 0.2, as well as the following micronutrients (mg dm<sup>-3</sup> medium): Fe – 8.0; Cu – 13.3; Mn – 5.1; B – 1.6; Mo – 3.7; Zn – 0.74. Phosphorus was applied in the form of granulated triple superphosphate (20% P), magnesium as MgSO<sub>4</sub> H<sub>2</sub>O, iron in the form of chelate, copper, manganese, zinc as sulphates, boron as boric acid, and molybdenum as ammonium molybdate. Fertilizer doses were divided into 3 equal parts and provided as root-applied nutrient solution in the following way: one day before sowing, 30 days from sowing, and 10 days before harvest. The nutrients whose quantity and form were not differentiated were applied only once before sowing. The plants were watered once or twice per day, as necessary. A single dose of water received by the plants was about 250 ml. Leaf rosettes were harvested around 10 May (48–54 days from sowing). After harvest, the fresh leaf yield and the biological value of rocket leaves were determined, taking into account the contents of L-ascorbic acid (in the fresh rocket leaves), chlorophyll, flavonoids, glucosinolates, and essential oil (in the leaves dried at a temperature of 35°C).

**L-ascorbic acid content.** L-ascorbic acid was extracted from a sample using a solution of metaphosphoric acid. A reducing solution was used to convert dehydro-L(+)-ascorbic acid to L(+)ascorbic acid. The total L(+)-ascorbic acid content was determined by HPLC with UV detection at 265 nm using an external standard. The limit of detection for this method was 1.5 ug ml<sup>-1</sup>, while the limit of quantification was 5 ug ml<sup>-1</sup>.

Flavonoid content. Flavonoids, expressed as quercetin equivalents, were determined spectrophotometrically on a Cary 50 Varian spectrophotometer, according to Polish Pharmacopoeia VIII [2008], after they had been extracted from the plant material and the absorbance of the solutions had been measured at a wavelength of  $\lambda = 425$  nm. The limit of detection for this method was 4.01 mg 100 g<sup>-1</sup>, while the limit of quantification  $-4.76 \text{ mg } 100\text{g}^{-1}$ . 10 g of medium powdered plant material (using a 0.315 mm mesh sieve) was weighed and placed into a round-bottom flask, 20 ml of acetone, 2 ml of HCl (281 g  $l^{-1}$ ) and 1 ml of a methenamine solution (5 g  $l^{-1}$ ) were added, and the mixture was maintained for 30 min. in a boiling water bath under a reflux condenser. The hydrolysate was filtered through cotton wool into a 100 ml volumetric flask, the sediment, together with the cotton wool, was placed in the flask, 20 ml of acetone was added, and the whole was kept boiling again for 10 min. The digestion was repeated once again. The extracts were filtered to the same volumetric flask and the volume was made up with acetone. 20 ml of the solution was measured into a separator, 20 ml of water was added, and the solution was extracted with ethyl acetate in portions of 15 ml and three times in a 10 ml portion. The combined organic layers were washed twice with 40 ml of water, filtered through into a 50 ml volumetric flask, and the volume was made up with ethyl acetate. Two samples were prepared for determination: 2 ml of aluminium chloride solution (20 g  $l^{-1}$ ) was added to 10 ml of the stock solution and the volume was made up with a mixture (1 : 19) of acetic acid  $(1.02 \text{ kg } l^{-1})$  and methanol up to 25 ml. To prepare a reference solution, 10 ml of the stock solution was made up with a mixture (1 : 19) of acetic acid (1.02 kg  $l^{-1}$ ) and methanol up to 25 ml volume. After 45 minutes, the absorbance of the solutions was measured at 425 nm, using the reference solution as reference. The total flavonoid content (%) was expressed as quercetin equivalents according to the following formula:

$$X = \frac{A \times k}{m}$$

where:

A – absorbance of the test solution,

$$k$$
 – quercetin equivalents  $k = 0.875 \cdot \left( a \frac{l\%}{lcm} = 714 \right)$ 

m – the weighed amount of plant material in g.

Glucosinolate content. Glucosinolates (GL) were extracted with methanol and subsequently they were purified and subjected to enzymatic desulfation on ion exchange resin. The GL content was determined on a high-performance liquid chromatograph (HPLC), equipped with an ultraviolet detector, using reverse phase columns and gradient elution. To determine the total glucosinolate content (TGL), the following reagents were used: sinigrin monohydrate at a concentration of 5 mmol 1<sup>-1</sup> (Sigma-Aldrich) buffered to a pH of 5.8; Helix pomatia sulfatase type H-1 (Sigma-Aldrich); 70% (v/v) methyl alcohol (HPLC-grade purity) (Baker); sodium acetate (0.02 M solution with a pH of 4.0); sodium acetate (0.2 M solution), imidazole formate (6 M solution); ion exchange resin Sepharose DEAE CL-6B; water (HPLC-grade purity, Baker) - mobile phase A; acetonitrile (HPLC-grade purity), water (HPLC-grade purity, Baker), phosphoric acid - mobile phase B. A HPLC system (Shimadzu), a dual-beam spectrophotometer (Shimadzu) equipped with quartz cuvettes, an ASE 150 system (Dionex), an ultra centrifuge (Beckman Coulter L-90 K), and a Rotofix 32 A centrifuge were used for the analysis. A 15 cm long chromatographic column, with a diameter of 3.2 cm, a grain size of 4  $\mu$ m, filled with C<sub>18</sub> (Restek-Pinnacle II), and an Amicon Ultra 4 ultrafiltration unit (Millipore) were also used. The TGL determination procedure included the following: purification of the sulfatase, investigation of its activity, determination of the moisture and volatile content as well as extraction of the plant material by the methanol/water phase 70/30 (50 cm<sup>3</sup>) in an ASE 150 system, centrifugation of the extract in a Rotenta 360 centrifuge at 5000 g for 15 min, and the performance of a blank test. The chromatographic analysis of the sample was performed with a mobile-phase flow rate of 0.75 ml min<sup>-1</sup>, a detector wavelength of 229 nm, an injection volume of 20  $\mu$ l, and a column temperature of 30°C. Elution gradient: 0-2.5 min - 100 (phase A, isocratically), 2.5-20.5 min -100-0 phase A and 0-100 phase B (by gradient elution), 20.5-25.5 min - 100 phase B (isocratically), 25.5-28.5 - 0-100 phase A and 100-0 phase B (by gradient elution), 28.5–32.5 min – 100 phase A (isocratically). The extract was filtered through a syringe filter to a tightly capped vial and the sample was injected into the chromatograph. The total glucosinolate content (TGL) was calculated according to the following formula:

$$TGL = \frac{Ag}{ms} \times \frac{n}{m} \times \frac{100}{100 - w}$$

where:

 $A_g$  – the aggregate area of the peaks with area > 1% of the total peak area in integrator units,

 $A_s$  – the peak area in integrator units of desulfosinigrin,

m – the weight of the analytical sample g,

n – the amount of the internal standard added to the tube in µmol,

w – the moisture and volatile content as % of the sample weight.

The essential oil content was determined from air-dried powdered material (30 g) by hydrodistillation in a Clevenger-type apparatus, conducting the process for 3 hours, following Farmakopea Polska VIII guidelines [2008].

All assays were conducted in three replicates. The results were statistically analysed by analysis of variance at a significance level of 0.05.

### **RESULTS AND DISCUSSION**

The fresh leaf yield of rocket was on average 111.2 g and it was significantly higher in the plants fed with KCl than in those supplied with K<sub>2</sub>SO<sub>4</sub>. The plants that received the higher rates of nitrogen and potassium (the  $2^{nd}$  and  $3^{rd}$  rates) were characterized by significantly higher fresh matter yield than those fertilized at the lower rate. Rocket leaves are a valuable nutritional product, because they have a significant proportion of dry matter (on average 11.17%) [Nurzyńska-Wierdak 2015]. They were shown to be characterized by a high concentration of L-ascorbic acid (on average 92.66 mg  $100 \text{ g}^{-1}$ ). Brassica vegetables are marked by a large amount of L-ascorbic acid and protein, which are the main determinants of their nutritional value [Acikgoz 2011]. The results obtained in the present experiment are comparable to the values determined in cabbage plants [Acikgoz 2011] and partially in mustard greens [Ng et al. 2012] as well as they exceed those found for most green leafy vegetables [Ng et al. 2012, Yadav et al. 2013]. The rocket leaves also proved to be a valuable source of other biocomponents. The average content of chlorophyll a + b per 100 g of rocket fresh biomass was 1.16 mg. The chlorophyll a : b ratio in the rocket leaves was 0.98. Žnidarčič et al. [2011] found a higher concentration of chlorophyll and a higher chlorophyll a : b ratio in garden rocket and wild rocket. In most plants, the chlorophyll a to chlorophyll b ratio is 3 : 1 [Kopsell et al. 2004, Lisiewska et al. 2006, Vivek et al. 2013]. The concentration of chlorophyll pigments increases with plant growth, while the chlorophyll a : b ratio decreases [Lisiewska et al. 2006]. The above differences could have been caused by the different stages at which the plants were harvested, but they could also have resulted from the variation within the species and varieties [Kopsell et al. 2004, Mitić et al. 2013]. The chlorophyll content in leafy vegetables can be from 0.04 to 2.70 mg  $g^{-1}$  of fresh weight [Kamga et al. 2013, Mitić et al. 2013, Vivek et al. 2013]. The chlorophyll content in the fresh biomass of rocket should therefore be considered to be high, enhancing its nutritional and health-promoting value. It is reported that the average daily

Acta Sci. Pol.

consumption of 300–400 g of green vegetables and fruit in whole supplements chlorophyll and that plant products containing this component should be used in daily diet as a source of nutrient supplementation [Vivek et al. 2013]. Chlorophyll pigments are used as food pigments as well as in pharmaceutical and cosmetic preparations in which their antioxidant, bacteriostatic and tissue regeneration enhancing properties are utilised. Moreover, the results of the study by Kopsell et al. [2004] suggest that the chlorophyll content in green leafy vegetables can be used to estimate the concentration of lutein and  $\beta$ -carotene.

| K<br>source<br>(A)               | Ν           | K                |                           | Lagoarhia                      | Chlorophyll             |        |       |
|----------------------------------|-------------|------------------|---------------------------|--------------------------------|-------------------------|--------|-------|
|                                  | dose<br>(B) | dose<br>(C)      | Yield of fresh<br>biomass | acid<br>mg 100 g <sup>-1</sup> | а                       | b      | a+b   |
|                                  | g d         | lm <sup>-3</sup> | g poi                     | f.m.                           | mg g <sup>-1</sup> f.m. |        |       |
| K <sub>2</sub> SO <sub>4</sub> - | 0.3         | 0.3              | 95.4                      | 106.87                         | 0.61                    | 0.62   | 1.23  |
|                                  |             | 0.6              | 104.1                     | 75.91                          | 0.60                    | 0.60   | 1.20  |
|                                  |             | 0.9              | 94.5                      | 120.74                         | 0.50                    | 0.51   | 1.01  |
|                                  |             | 0.3              | 100.0                     | 107.57                         | 0.60                    | 0.62   | 1.22  |
|                                  | 0.6         | 0.6              | 115.8                     | 76.74                          | 0.60                    | 0.60   | 1.20  |
|                                  |             | 0.9              | 140.7                     | 72.84                          | 0.59                    | 0.59   | 1.18  |
| Mean (A)                         |             |                  | 108.4b                    | 93.45a                         | 0.58a                   | 0.59a  | 1.17a |
| KCl -                            |             | 0.3              | 106.5                     | 98.18                          | 0.64                    | 0.64   | 1.28  |
|                                  | 0.3         | 0.6              | 116.1                     | 102.55                         | 0.45                    | 0.45   | 0.91  |
|                                  |             | 0.9              | 100.2                     | 111.16                         | 0.46                    | 0.47   | 0.93  |
|                                  | 0.6         | 0.3              | 103.5                     | 102.48                         | 0.67                    | 0.68   | 1.35  |
|                                  |             | 0.6              | 131.1                     | 77.55                          | 0.58                    | 0.60   | 1.18  |
|                                  |             | 0.9              | 126.3                     | 59.23                          | 0.62                    | 0.63   | 1.25  |
| Mean (A)                         |             |                  | 114.0a                    | 91.86a                         | 0.57a                   | 0.58a  | 1.15a |
| Mean (B)                         |             | 0.3              | 95.17b                    | 102.57a                        | 0.54b                   | 0.59a  | 1.17a |
|                                  |             | 0.6              | 119.7a                    | 82.74b                         | 0.61a                   | 0.62a  | 1.15a |
| Mean (C)                         |             | 0.3              | 101.4b                    | 103.78a                        | 0.63a                   | 0.64a  | 1.09b |
|                                  |             | 0.6              | 117.0a                    | 83.19b                         | 0.56ab                  | 0.56ab | 1.23a |
|                                  |             | 0.9              | 115.2a                    | 90.99ab                        | 0.54b                   | 0.55b  | 1.09b |

Table 1. Bioactive substances in the fresh matter (f.m.) of rocket leaves (mean from 2010–2012)

The other active components of rocket were also at a relatively high level. 100 g of dried rocket leaves contained on average 0.84 g of flavonoids and 0.15 ml of essential oil, while 1 g of dry plant material was characterized by the presence of 10.56  $\mu$ mol of glucosinolates, on average. A similar content of flavonoids was determined for mustard greens [Ng et al. 2012], while at the same time a correlation was shown between the antioxidant groups (phenolics and flavonoids) and the antioxidant capacity. The glucosinolate content in the leaves of plants of the genera *Brassica*, *Degenia* and *Eruca* (Brassicaceae) can range 9.9–31.4  $\mu$ mol g<sup>-1</sup> DW [De Pacale et al. 2007, De Nicola et al. 2011,

Hortorum Cultus 14(5) 2015

Omirou et al. 2012] and is associated with the amount of nitrogen used and harvest timing [Omirou et al. 2012]. Furthermore, high ontogenetic variation in the profile of glucosinolates and flavonoids was found in rocket [Bennett et al. 2006]. The results of the study also reveal high variation in the essential oil concentration in rocket [Blažević and Mastelić 2008, Mastelić et al. 2008], which may be attributable to the biodiversity of the material analyzed and the variety of analytical methods. In analyzing the essential oil of rocket by the method of hydrodistillation–adsorption and hydrodistillation, Mastelić et al. [2008] showed an increase by 104.7% (flowers) and 27.9% (leaves together with green pods) in the yield of distilled oil in favour of the former method.

| K source                       | N dose<br>(B)      | K dose<br>(C) | Dry<br>matter*        | Glucosinolates            | Flavonoids                 | Essential oil               |
|--------------------------------|--------------------|---------------|-----------------------|---------------------------|----------------------------|-----------------------------|
| (A)                            | g dm <sup>-3</sup> |               | %                     | µmol g <sup>-1</sup> d.m. | g 100 g <sup>-1</sup> d.m. | ml 100 g <sup>-1</sup> d.m. |
| K <sub>2</sub> SO <sub>4</sub> |                    | 0.3           | 936                   | 8.60                      | 0.98                       | 0.13                        |
|                                | 0.3                | 0.6           | 9.44                  | 10.02                     | 0.71                       | 0.18                        |
|                                |                    | 0.9           | 8.45                  | 9.37                      | 0.73                       | 0.18                        |
|                                | 0.6                | 0.3           | 7.56<br>12.10<br>7.85 | 10.10                     | 0.78                       | 0.13                        |
|                                |                    | 0.6           |                       | 8.47                      | 0.86                       | 0.10                        |
|                                |                    | 0.9           |                       | 8.50                      | 0.97                       | 0.18                        |
| Mean (A)                       |                    |               | 13.33a                | 9.18b                     | 0.84a                      | 0.15a                       |
| KCI                            | 0.3                | 0.3           | 9.78                  | 8.48                      | 0.86                       | 0.15                        |
|                                |                    | 0.6           | 9.17                  | 16.47                     | 0.88                       | 0.15                        |
|                                |                    | 0.9           | 10.16                 | 11.92                     | 0.79                       | 0.10                        |
|                                | 0.6                | 0.3           | 8.82                  | 14.96                     | 0.86                       | 0.18                        |
|                                |                    | 0.6           | 7.95                  | 8.96                      | 0.62                       | 0.13                        |
|                                |                    | 0.9           | 8.20                  | 10.83                     | 1.00                       | 0.15                        |
| Mean (A)                       |                    |               | 9.01b                 | 11.94a                    | 0.84a                      | 0.14a                       |
| Mean (B)                       |                    | 0.3           | 9.39a                 | 12.31a                    | 0.83a                      | 0.15a                       |
|                                |                    | 0.6           | 8.75a                 | 10.30b                    | 0.85a                      | 0.15a                       |
|                                |                    | 0.3           | 8.88ab                | 10.54a                    | 0.87a                      | 0.15a                       |
| Mean (C)                       |                    | 0.6           | 9.67a                 | 10.98a                    | 0.77b                      | 0.15a                       |
|                                |                    | 0.9           | 8.67b                 | 10.16a                    | 0.87a                      | 0.15a                       |

| Table 2. Bioactive substances in t | the dry matter (d | l. m.) of rocket leaves ( | (mean from 2010–2012) |
|------------------------------------|-------------------|---------------------------|-----------------------|
|------------------------------------|-------------------|---------------------------|-----------------------|

\*(Nurzyńska-Wierdak 2015)

The fresh biomass and the content of glucosinolates in rocket fed with chloride sodium were significantly higher than in the plants fertilized with potassium sulphate (tab. 2). Thus, the plants that received more sulphur (fed with  $K_2SO_4$ ) did not accumulate more glucosinolates than those that received less of this nutrient (supplied with KCl). This is in agreement with the results of the study by De Pascale et al. [2005] who did not find sulphur to affect the accumulation of glucosinolates. The immediate precursors in the biosynthesis of glucosinolates are amino acids, both protein and non-protein ones, mainly tryptophan, methionine, and phenylalanine [Graser et al. 2000, Falk et al. 2004]. The rocket plants fertilized with potassium chloride accumulated significantly more methionine and phenylalanine (as well as most of the other amino acids) [Nurzyńska-Wierdak 2015] and glucosinolates compared to the other plants. The form of potassium was not found to significantly affect the accumulation of L-ascorbic acid, chlorophyll, flavonoids, and essential oil. Likewise, the level of sulphur did not influence the vitamin C content in spinach and pepper [Smatanová i in. 2004]. However, other relationships were shown in an earlier study conducted during the autumn season [Nurzyńska-Wierdak 2009]; in this study, the L-ascorbic acid content in the rocket leaves was significantly higher when the plants were fed with potassium chloride compared to potassium sulphate. These differences may be attributable to different light conditions (the spring and autumn seasons), which particularly strongly affect the metabolism of plants with a short growing season, and may also be a result of the high requirement of rocket plants for sulphur.

The applied rates of nitrogen and potassium significantly modified the content of some biocomponents in rocket. The higher rate of nitrogen contributed to an increase in fresh plant biomass, at the same time causing a decrease in the content of L-ascorbic acid and glucosinolates. The research by Omirou et al. [2012] shows that the glucosinolate content in rocket leaves is significantly modified by the rate of nitrogen and growth stage. Moreover, various groups of glucosinolates exhibited a different response to an increase in the rate of nitrogen and the form of potassium [Kim et al. 2006, Omirou et al. 2012] as well as to water [Zhang et al. 2008] and environmental stress [Antonious et al. 2009]. It seems that under optimal plant growth conditions the glucosinolate content is more stable. In the present study, the plants that received more potassium were characterized by higher fresh biomass compared to the other plants. The L-ascorbic acid content, in turn, was highest at the lowest and highest rates of potassium. Similar relationships were demonstrated in an earlier study [Nurzyńska-Wierdak 2009]. Furthermore, Antonious et al. [2009] proved that environmental stress during plant growth may increase the concentration of L-ascorbic acid in *Brassica* shoots, but without increasing the yield of this substance per unit area.

## CONCLUSIONS

To sum up, it can be concluded that rocket leaves are a rich and very promising source of health-promoting substances and should be taken into consideration as functional diet components. The possibility of obtaining fresh plant produce during the spring, spring-summer and autumn-winter seasons is of great importance in this respect. The short growth period of rocket, the continuity of yields, and the duration of the yielding period, given the high biological value of rocket, make this plant a perfect component of safe and easily available food that allows a meal to be prepared efficiently and in various situations associated with human activity. The presented results of the study concerning the accumulation of L-ascorbic acid, glucosinolates, flavonoids, and essential oil prove that it is possible to increase the biological value of rocket leaves by using an appropriate level of plant mineral nutrition. The form of potassium was not found to significantly affect the accumulation of L-ascorbic acid, chlorophyll, flavonoids, and essential oil. Nevertheless, potassium chloride, the application of which significantly increased the concentration of glucosinolates in the leaves, can be indicated as a good source of potassium in growing rocket. On the other hand, an increase in the rate of nitrogen contributed to a decrease in the content of L-ascorbic acid and glucosinolates.

## ACKNOWLEDGMENTS

This research was financially supported by the Polish Ministry of Science and Higher Education funds, research grant No N N310 210537.

### REFERENCES

- Acikgoz, F.E., (2011). Mineral, vitamin C and crude protein contents in kale (*Brassica oleraceae* var. *acephala*) at different harvesting stages. Afr. J. Biotech., 10, 17170–17174.
- Aires, A., Fernandes, C., Carvalho, R., Bennett, R.N., Saavedra, M.J., Rosa, E.A.S. (2011). Seasonal effects on bioactive compounds and antioxidant capacity of six economically important *Brassica* vegetables. Molecules, 16, 6816–6832.
- Antonious, G.F, Bomford, M., Vincelli, P. (2009). Screening *Brassica* species for glucosinolate content. J. Environ. Sci. Health., 44, 311–316.
- Blaževic, I., Mastelic, J. (2008). Free and bound volatiles of rocket (*Eruca sativa Mill.*). Flav. Fragr. J., 23, 278–285.
- Bukhsh, E., Malik, S.A., Ahmad, S.S. (2007). Estimation of nutritional value and trace elements 21 content of *Carthamus oxyacantha*, *Eruca sativa* and *Plantago ovata*. Pak. J. Bot. 39, 4, 1181–1187.
- Chun, J.-H., Arasu, M.V., Lim, Y.-P., Kim, S.-J. (2013). Variation of major glucosinolates in different varieties and lines of rocket salad. Hort. Environ. Biotechnol., 54, 206–213.
- De Nicola, G.R., Blazevic, I., Montaut, S., Rollin, P., Mastelic, J., Iori, R., Tatibouet, A. (2011). Glucosinolate distribution in aerial parts of *Degenia velebitica*. Chem. Biodivers, 11, 2090–2096.
- De Pascale, S., Maggioa, A., Pernice, R., Fogliano, V., Barbieri, G. (2007). Sulphur fertilization may improve the nutritional value of *Brassica rapa* L. subsp. sylvestris. Europ. J. Agron., 26, 418–424.
- Fahey, J.W., Zalcmann, A.T., Talalay, P. (2001). The chemical diversity and distribution of glucosinolates and isothiocyanates among plants. Phytochemistry, 56, 5–51.
- Falk, K.J., Vigel, C., Textor, S., Bartram, S., Hick, A., Pickett, J.A., Gerhenzon, J. (2004). Glucosinolate biosynthesis: demonstration and characterization of the condensing enzyme of chain elongation cycle in *Eruca sativa*. Phytochemistry, 65, 1073–1084.
- *Eruca sativa* Mill.: Phytochemical profile and antimicrobial properties of rocket leafy salads. Planta Med., 76, doi: 10.1055/s-0030-1264205.
- Graser, G., Schneider, B., Oldham, N.J., Gershenzon, J., (2000). The methionine chain elongation pathway in the biosynthesis of glucosinolates in *Eruca sativa* (Brassicaceae). Arch. Biochem. Biophys. 378, 411–419.
- Hassan, F.U., Manaf, A., Qadir, G., Basra, S.M.A. (2007). Effects of sulphur on seed yield, oil, protein and glucosinolates of canola cultivars. Int. J. Agri. Biol., 9, 504–508.

- Kadivec, M., Kopjar, M., Žnidarčič, D., Požrl, T. (2015). Potential of eggplant peel as by-product. Acta Alimen. Hung., 44, 126–131.
- Kamga, R.T., Kouame, C., Atangana, A.R., Chagomoka, T., Ndango, R. (2013). Nutritional evaluation of five African indigenous vegetables. J. Hort. Res., 21, 99–106.
- Kim, S.-J., Ishii, G. (2006). Glucosinolate profiles in the seeds, leaves and roots of rocket salad (*Eruca sativa* Mill.) and anti-oxidative activities of intact plant powder and purified 4-methoxyglucobrassicin. Soil Sci. Plant Nutr., 52, 394–400.
- Kopeć, A., Piątkowska, E., Bieżanowska-Kopeć, R., Pysz, M., Koronowicz, A., Kapusta-Duch, J., Smoleń, S., Rakoczy, R., Skoczylas, Ł., Leszczyńska, T., Ledwożyw-Smoleń, I. (2015). Effect of lettuce biofortified with iodine by soil fertilization on iodine concentration in various tissues and selected biochemical parameters in serum of Wistar rats. J. Funct. Foods, 14, 479–486.
- Khoobchandani, M., B.K. Ojeswi, B.K., Ganesh, N., Srivastava, M.M., Gabbanini, S., Matera, R., Iori, R., Valgimigli, L. (2010). Antimicrobial properties and analytical profile of traditional *Eruca sativa* seed oil: Comparison with various aerial and root plant extracts. Food Chem. 120, 217–224.
- Kopsell, D.A., Kopsell, D.E., Lefsrud, M. Curran-Celentano, J., Dukach, L.E. (2004). Variation in lutein, β-caritene, and chlorophyll concentrations among *Brassica oleracea* cultigens and seasons. Hort. Sci., 39, 361–364.
- Lisiewska, Z., Kmiecik, W., Korus, A. (2006). Content of vitamin C, carotenoids, chlorophylls and polyphenols in green parts of dill (*Anethum graveolens* L.) depending on plant height. J. Food Comp. Analys., 19, 134–140.
- Liu, J.-Y., Chen, X.-X., Tang, S.C.-W., Lao, L.-X., Sze, S. C.-W., Lee, K.-F., Zhang, K.Y.-B. (2015). Edible plants from traditional Chinese medicine is a promising alternative for the management of diabetic nephropathy. J. Funct. Foods, 14, 12–22.
- Mastelić, J., Jerković, I., Blažević, I., Radonić, A., Krstulović L. (2008). Hydrodistillationadsorption method for the isolation of water-soluble, non-soluble and high volatile compounds from plant materials. Talanta 76, 885–891.
- Michael, H.N., Shafik, R.E., Rasmy, G.E. (2011). Studies on the chemical constituents of fresh leaf of *Eruca sativa* extract and its biological activity as anticancer agent *in vitro*. J. Med. Plants Res., 5, 1184–1191.
- Michałojć, Z., Dzida, K. (2012). Yielding and biological value of sweet pepper fruits depending on foliar feeding using calcium. Acta Sci. Pol., Hortorum Cultus, 11, 255–264.
- Mitić, V., Jovanović, V.S., Dimitrijević, M., Cvetković, J., Petrović, G., Stojanović, G. (2013). Chemometric analysis of chlorophyll a, b and carotenoid content in green leafy vegetables. Biol. Nyss., 4, 49–55.
- Narits, L. (2011). Effect of top-fertilizing of raw protein and glucosinolates content of winter turnip rape. Agron. Res., 9, 451–454.
- Ng, X.N., Chye, F.Y., Mohd Ismail, A. (2012). Nutritional profile and antioxidative properties of selected tropical wild vegetables. Internat. Food Res. J., 19, 1487–1496.
- Nurzyńska-Wierdak, R. (2009). Growth and yield of garden rocket (*Eruca sativa* Mill.) affected by nitrogen and potassium fertilization. Acta Sci. Pol., Hortorum Cultus, 8, 23–33.
- Nurzyńska-Wierdak, R. (2015). Protein nutritional value of rocket leaves and possibilities of its modification during plant growth. Turk. J. Agric. For., 39 (in press).
- Nurzyńska-Wierdak, R., Dzida, K., Rożek, E., Jarosz, Z. (2012). The effect of nitrogen and potassium on N-NH<sub>4</sub> and N-NO<sub>3</sub> accumulation and nutrient contents in rocket (*Eruca sativa* Mill.) leaves. Acta Sci. Pol., Hortorum Cultus, 11, 211–221.
- Omirou, M., Papastefanou, C., Katsarou, D., Papastylianou, I., Passam, H.C., Ehaliotis, C., Papadopoulou, K.K. (2012). Relationships between nitrogen, dry matter accumulation and glucos-

inolates in *Eruca sativa* Mills. The applicability of the critical N-NO<sub>3</sub> levels approach. Plant Soil, 354, 347–358.

- Ortega, R.M. (2006). Importance of functional foods in the Mediterranean diet. Pub. Health Nutr., 9, 1136–1140.
- Patricia, O., Zoue, L., Megnanou, R.-M., Doue, R., Niamke, S. (2014). Proximate composition and nutritive value of leafy vegetables consumed in Northern Côte d'Ivoire. Eur. Sci. J., 10, 212–227.
- Polat, U. (2010). The effects on metabolism of glucosinolates and theirs hydrolysis products. J. Biol. Environ. Sci., 4, 39–42.
- Polish Pharmacopoeia VIII
- Romojaro, A., Ángeles Botella, A., Obón, C., Pretel, M.T. (2013). Nutritional and antioxidant properties of wild edible plants and their use as potential ingredients in the modern diet. Int. J. Food Sci. Nutr., 64, 944–952.
- Saeed, M.K., Anjum, S., Ahmad, I., Ali, S., Zia, A., Ali, S. (2012). Nutritional facts and free radical scavenging activity of turnip (*Brassica rapa*) from Pakistan. World Appl. Sci. J., 19, 3, 370–375.
- Singh, B., Singh, J., Kumar, A., Yadav, Y.P., Singh, S. (1999). Response of Brassicas to sulphur for yield. Indian J. Agric. Sci., 69, 427–429.
- Sinkovič, L., Demšar, L., Žnidarčič, D., Vidrih, R., Hribar, J., Treutter, D. (2015). Phenolic profiles in leaves of chicory cultivars (*Cichorium intybus L.*) as influenced by organic and mineral fertilizers. Food Chem., 166, 507–513.
- Smatanovà, M., Richter R., Hlušek J. (2004). Spinach and red pepper response to nitrogen and sulphur fertilization. Plant Soil Environ. 50, 303–308.
- Srianta, I., Arisasmita, J.H., Patria, H.D., Epriliati, I. (2012). Ethnobotany, nutritional composition and DPPH radical scavenging of leafy vegetables of wild *Paederia foetida* and *Erechtites hieracifolia*. Internat. Food Res. J., 19, 245–250.
- Strgar, J., Pilih, M., Pogačnik, M., Žnidarčič, D. (2013). Knowledge of medicinal plants and their uses among secondary and grammar school students: a case study from Slovenia. Arch. Biol. Sci., 65, 1123–1129.
- Tassi, E.M.M., Amaya-Farfan, J. (2008). Carotenoid uptake by human triacylglycerol-rich lipoproteins from the green leafy vegetable *Eruca sativa*. Ecol. Food Nutr. 47, 1, 77–94.
- Vardavas, C.I., Majchrzak, D., Wagner, K.-H., Elmadfa, I., Kalatos, A. (2006). The antioxidant and phylloquinone content of wildly grown greens in Recte. Food Chem., 99, 813–821.
- Vig, A.P., Rampal, G., Thind, T.S., Arora, S. (2009). Bio-protective effects of glucosinolates A review. LWT – Food Sci. Technol., 42, 1561–1572.
- Villatoro-Pulido, M., Font, R., Saha, S., Obregón-Cano, S., Anter J., Muńoz-Serrano, A., De Haro-Bailón, A., Alonso-Moraga, A., Del Río- Celestino, M. (2012). In vivo biological activity of rocket extracts (*Eruca vesicaria* subsp. *sativa* (Miller) Thell) and sulforaphane. Food Chem. Toxicol. 50, 1384–1392.
- Vivek, P., Prabhakaran, S., Shankar, S.R. (2013). Assessment of nutritional value in selected edible greens based on the chlorophyll content in leaves. Res. Plant Biol., 3, 45–49.
- Vyas, P., Prakash, S., Shivanna, K.R. (1995). Production of wide hybrids and backcross progenies between *Diplotaxis erucoides* and crop brassicas. Theor. Appl. Genet., 90, 549–553.
- Zhang, H., Schonhof, I., Krumbein, A., Gutezeit, B., Li, L., Stützel, H., Schreiner, M. (2008). Water supply and growing season influence glucosinolate concentration and composition in turnip root (*Brassica rapa* ssp. *rapifera* L.). J. Plant Nutr. Soil Sci. 171, 255–265.
- Žnidarčič, D., Ban, D., Šircelj, H. (2011). Carotenoid and chlorophyll composition of commonly consumed leafy vegetables in Mediterranean countries. Food Chem., 129, 1164–1168.

Žnidarčič, D., Šircelj, H., Kacjan-Maršić, N. (2013). The influence of temperature and storage time on Cantaloupe melons physicochemical quality. Ital. J. Food Sci., 25, 459-464.

Yadav, R.K., Kalia, P., Kumar, R., Jain, V. (2013). Antioxidant and nutritional activity studies of green leafy vegetables. Int. J. Agric. Food Sci. Technol., 4, 707–712.

# WARTOŚĆ BIOLOGICZNA LIŚCI *Eruca sativa* Mill. POD WPŁYWEM ZRÓŻNICOWANEGO ŻYWIENIA ROŚLIN AZOTEM I POTASEM

Streszczenie. Warzywa zajmują znaczące miejsce w piramidzie zdrowego żywienia. Zaleca się jak najczęstsze ich spożywanie w celu dostarczenia substancji odżywczych i biologicznie aktywnych. Celem prezentowanych badań było określenie wartości biologicznej liści rokietty siewnej pod wpływem zróżnicowanego żywienia mineralnego roślin azotem i potasem. Rośliny uprawiano w podłożu torfowym, w warunkach szklarniowych. Po zbiorze w liściach rokietty określono zawartość kwasu L-askorbinowego, chlorofilu, flawonoidów, glukozynolanów i olejku eterycznego. Liście rokietty okazały się cennym źródłem bioskładników i można je uznać za istotne składniki żywności funkcjonalnej. Średnia zawartość chlorofilu a + b w 100 g świeżej biomasy rokietty wynosiła 1,16 mg, a koncentracja kwasu L-askorbinowego - 92,66 mg. 100 g wysuszonych liści rokietty zawierało średnio 0,84 g flawonoidów i 0,15 ml olejku eterycznego, a 1 g suchego materiału roślinnego charakteryzował się średnio obecnością 10,56 µmol glukozynolanów. Interesującym źródłem potasu okazał się chlorek potasu, którego aplikacja istotnie podnosiła koncentrację glukozynolanów w liściach rokietty. Nie stwierdzono jednak istotnego wpływu postaci potasu na gromadzenie kwasu L-askorbinowego, chlorofilu, flawonoidów i olejku eterycznego. Podwyższona dawka azotu przyczyniła się do zmniejszenia zawartości kwasu L-askorbinowego i glukozynolanów. Przedstawione wyniki badań wskazują, że istnieje możliwość modyfikacji składu chemicznego liści rokietty przy zastosowaniu odpowiedniego schematu żywienia mineralnego roślin.

Słowa kluczowe: Brassicacea, warzywa, rokietta, substancje aktywne, kwas L-askorbinowy

Accepted for print: 12.05.2015

For citation: Nurzyńska-Wierdak, R. (2015). Biological value of *Eruca sativa* Mill. leaves under the different plant nutrition by nitrogen and potassium. Acta Sci. Pol. Hortorum Cultus, 14(5), 41–53.

53

Hortorum Cultus 14(5) 2015