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Research Article

Influence of Heat Treatments on the Content of Bioactive Substances and Antioxidant Properties of Sweet Potato (*Ipomoea batatas* L.) Tubers

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Heat treatment can affect the bioactive compounds in sweet potato (SP). In this work, we monitored the influence of heat treatment (boiling, steaming, microwaving, and baking) on the total polyphenols content (TPC), total antioxidant capacity (TAC), total anthocyanins content (TANTC), and phenolics acids (chlorogenic (CGA), neochlorogenic (neo-CGA), and *trans*-ferulic (tFA)) in two SP varieties grown in Slovakia and Croatia. TPC, TAC, and TANT were determined spectrophotometrically and phenolic acids by HPLC. TPC ranged from 576 (Beauregard, Croatia; Be/HR) to 3828 (414-purple, Slovakia; Pu/SK) mg/kg DW in the raw SP tubers. After heat treatment, TPC increased, most in steamed SP (8438 mg GAE/kg DW; Pu/SK), while only in boiled SP (Be/HR), TPC decreased (353 mg GAE/kg DW). TAC varied from 0.848 (Be/HR) to 8.67 (Pu/HR) (μ mol TE/g DW) in raw SP. The TAC increased by heat treatment (max. 14.2 μ mol TE/g DW; cooking Be/SK), except for Pu/HR. The TANT ranged from 151 (raw Pu/SK) to 1276 (microwaved Pu/SK) mg CyE/kg FW. Heat treatment had a negative effect on phenolic acid content; the largest reduction was after boiling: CGA by 29% (Pu/SK), neo-CGA by 69% (Pu/HR), and tFA by 29% (Be/HR). The influence of heat treatment on the monitored quantities is not definite.

1. Introduction

Sweet potatoes belong to the most economically important crops in the world with corn, wheat, rice, and potatoes. In 2019, their world production amounted to almost 92 million tons. The largest producer was Asia (75%), while the production of China was 53 mil. tons, which represented 57.6% of world production. In Europe, 93.4 thousand tons of SP were grown [1]. Sweet potatoes are tropical plants and do not tolerate frost. At temperatures below 1°C, their vegetative growth and productivity decline [2]. However, they quickly adapt to high temperatures and drought, making them an

attractive crop, especially in countries with limited agricultural resources [3]. In developing countries, they are the fifth most important food crop [4]. For human consumption, sweet potato tubers are preferably prepared in various ways: cooked, baked, fried, and dried in the form of sticks or slices. They are also used as a raw material in the production of pasta, in alcoholic beverages, as a source of natural pigments, or in the production of paper, cosmetics, and adhesives tissues [5, 6]. Sweet potatoes are a crop with high nutritional value; they contain large amounts of carbohydrates, fiber, vitamins, and minerals [7]. Carbohydrates are the predominant part of sweet potato tubers. The sweet taste

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of sweet potatoes is caused by the presence of glucose (2.7-4.7 mg/g DW), fructose (1.4-4.0 mg/g DW), sucrose (56.9-60.0 mg/g DW), and nondetectable maltose [8]. The major polysaccharide of SP is starch making up 80% of the dry matter [9]. Sweet potatoes are also a cheap and rich source of fiber. Its content depends on the variety; the amount of crude fiber is in the range of 0.6 to 1.5% [10]. The total dietary fiber content of sweet potatoes is enough to affect the glycemic index elicited by these roots, and we cannot rule out a bioactive effect of the protein components. It may prove beneficial for diabetic patients who consume sweet potatoes [11]. In general, sweet potato roots contain 1.73%-9.14% (dry basis) proteins. The main protein in sweet potato is sporamin, which can be divided into two subgroups: sporamin A (contains 219 amino acid residues) and sporamin B (contains 216 amino acid residues). The sweet potato protein contains 18 different amino acids, about 40.7% of the sweet potatoes protein consists of the essential amino acids Ile, Leu, Met, Phe, Thr, Val, Lys, and Trp. The last two of them are the primary limiting amino acids [12]. Sweet potato tubers contain a small amount of lipids (from 0.72% to 1.44%), while the crude fat content is significantly affected by a variety [13]. The primary fatty acids are palmitic acid (C16:0), linoleic acid (C18:2), and linolenic acid (C18: 3), while a smaller proportion is stearic acid (C18:0), oleic acid (C18:1), and arachidic acid (C20:0) [14]. Sweet potatoes are an excellent source of minerals. The most represented are Ca, K, and P and from trace elements Cu, Fe, Zn, Mn, Na, and Mg. In comparison with the other vegetables, sweet potatoes have more Mg, Na, P, and Fe than carrots or cabbage [5, 15]. According to [16], SP are also crucial in terms of the content of vitamins and other bioactive substances (polyphenolic compounds, phenolic acids, and anthocyanins and beta-carotene), which have antioxidant effects [4, 17, 18]. Sweet potato roots comprise hydrophilic vitamins (B1, B2, B6, niacin, pantothenic acid, biotin, and vitamin C) and lipophilic vitamins E and A [8, 15]. The precursor of vitamin A is beta-carotene with two beta-ionone rings, which also possesses antioxidant activity [19]. Beta-carotene is mainly present in orange varieties of SP. In 2016, because of the high content of vitamin A, the orange SP were recognized as a food able to improve nutrition in many households in sub-Saharan Africa [19-21]. Another antioxidant vitamin, ascorbic acid, also occurs in high amounts in the roots. The high content of vitamin B6 (pyridoxine) in the roots helps in reducing the blood levels of homocysteine, which is associated with the increased risk of cardiovascular diseases [19]. The characteristic color of purple SP is caused by the presence of anthocyanins. Together with phenols, they represent the main bioactive substances in purple SP. Anthocyanins create a group of water-soluble flavonoids; in sweet potato, they occur as mono- or diacylated forms of cyanidin and peonidin [22]. It is supposed that anthocyanins—natural pigment—can have many positive effects on human health, such as antioxidant, anti-inflammatory, anticarcinogenic, chemoprotective, and antihyperglycemic effects, and they can help avoid oxidation of LDL-cholesterol. They are present mainly in purple varieties of sweet potato—in peel and also in the flesh [23, 24].

Phenolic compounds are antioxidant molecules with at least one aromatic ring and one or more hydroxyl groups, including their functional derivatives. These slightly various substances are essential for the growth and reproduction of plants, and they act as antipathogenic agents [18]. The main components of the sweet potato phenolic compounds are chlorogenic acids, which belong to the group of ester compounds. They are formed by the condensation of quinic acid and trans-cinnamic acids, which include coffee acid, pcoumaric acid, and ferulic acid [25]. 3-O-Caffeoylquinic acid is the most common of chlorogenic acids [17, 26, 27]. For example, in sweet potato leaves, twenty CQA phenolic acids, such as 3-CQA, 3,4-diCQA, 3,5-diCQA, 4,5-diCQA, and tri-CQA, were identified by LC-MS2 analysis [25]. The phenolic hydroxyl of chlorogenic acids easily reacts with free radicals, making the free radicals lose activity. Chlorogenic acids in plants have high antioxidant activities, such as reducing power, metal chelating, and lipid peroxidation-inhibiting activity [27]. Thus, in addition to antimutagenic and anticarcinogenic effects in vitro and in vivo, chlorogenic acid can prevent the formation of hydroxyl radicals, remove free radicals, and eliminate oxidative activity [28]. However, there are only a few studies about the differences in antioxidant activities between different chlorogenic acids, which does not provide sufficient theoretical support for the application of antioxidant activities [27]. Sweet potatoes are commonly prepared before consumption in different ways: baking, boiling, microwaving, steaming, or frying. Heat treatment improves their digestibility [29], induces significant changes in the chemical composition, and thus influences the concentration and bioavailability of compounds [18, 30]. Generally, the total phenolic content of sweet potatoes increases; on the other hand, some phenolic derivatives such as caffeic acids are decreased by heat treatment. Concerning the variation of these constituents, the antioxidant activity of sweet potatoes also shows similar variation during thermal processes [30]. The aim of our study was to research changes in the content of bioactive substances and their antioxidant activity due to heat treatments which are commonly used in the preparation of sweet potato tubers.

2. Materials and Methods

2.1. Plant Material. Two varieties of sweet potatoes with different flesh colors were used for the analyses—Beauregard (orange) and 414-purple (purple). Both cultivars were grown in the cadastral area of Šoporňa in the Slovak Republic (Be/SK, Pu/SK) and the east part in Croatia (cadastral area of Vukovar) (Be/HR, Pu/HR). After plowing using the adjusted plow machine, sweet potatoes from Croatia were sorted and cured at 25°C for 4 to 5 days. After, they were stored at a temperature of 13–16°C and a maximum humidity of 70% (max. 10 days). After transport to Slovakia, they were immediately delivered to our workplace and used for sample preparation. Sweet potatoes from Slovakia were taken directly in the field and then brought to the workplace, where we cleaned them. The next day, they were used for sample preparation. About 3 kg of plant material was taken from the

given sampling sites for each cultivar. Raw and heat-treated sweet potatoes were used for analyses.

- Chemicals. MetOH (80%),MetOH (99.8%),Folin-Ciocalteu reagents, DPPH (2,2'-diphenyl-1-picrylhydrazyl), Trolox (2,5,7,8-tetramethylchroman-2-carboxylic acid), HCl (36%), buffer pH 3.5 (Na₂HPO₄, c = 0.2 mol/L; citric acid, c = 0.1 mol/L), EtOH (80%), authentic standards of chlorogenic acid (purity ≥ 95.0%), neochlorogenic acid (purity ≥ 95.0%), trans-ferulic acid (purity ≥ 95.0%), acetonitrile (gradient HPLC grade), and phosphoric acid (ACS grade) were purchased from Sigma-Aldrich (Sigma-Aldrich Chemie GmbH, Steinheim, Germany); gallic acid (p.a.) and Na₂CO₃ were purchased from Merck (Germany); gallic acid (p.a.) was provided by Merck (Germany); double deionized water (ddH₂O) was treated (0.054 µS/cm) in a Simplicity 185 purification system (Millipore, UK).
- 2.3. Preparation of Samples. All tubers were washed (first with drinking water and then distilled water) and peeled. The peeled tubers were rewashed (with distilled water) and cut into slices of the thickness of 3 mm. The slices from each tuber were divided into five approximately equal parts. One part of the tuber (crude) was mixed (Grindomix GM2000 Retsch, 2000 rpm, 30 sec) and then the sample was homogenized. Other sections were variously heat-treated: cooked in water (10 min), steamed (15 min, 97 \pm 2°C), and microwaved (5 min, 800 W) as well as baked (15 min, 200°C). After cooling, slices of sweet potatoes were mixed (Grindomix GM2000 Retsch, 2000 rpm, 15 sec) and then homogenized.
- 2.4. Preparation of Extracts. 25 g of homogenized sample (raw flesh and boiled, steamed, baked, and microwaved flesh, respectively) was taken and poured into 50 mL of 80% MetOH at laboratory temperature and extracted by horizontal shaker (Unimax 2010; Heidolph Instrument GmbH, Germany) for 12 hours. Sample was filtered through Munktell No. 390 paper (Munktell & Filtrac, Germany) and stored in closed 66 mL vial tubes. Determination of TPC, TAC, TANTC, and phenolics acids was performed eight times.
- 2.5. Determination of Total Polyphenols Content. The total polyphenols content (TPC) was determined using the colorimetric Folin–Ciocalteu method [31] by spectrophotometric analysis (spectrophotometer Shimadzu UV-1800). The aliquot portion of extract (0.1 mL) was pipetted into a 50 mL flask. Then 2.5 mL of Folin–Ciocalteu reagent was added; after 3 minutes, 5 mL of 20% sodium carbonate aqueous was added; and distilled water was added to mark. Also, blank with distilled water was prepared by the same procedure and the calibration curve was prepared with standard solutions of gallic acid. After mixing and leaving for 2 hours in the laboratory, the complex forming was ended and the absorbance of blue solutions was measured in

cuvettes of 1 cm width at a wavelength of 765 nm. The content of total polyphenols in the sample was expressed as the content of gallic acid in mg/kg of fresh matter and calculated to dry weight (mg GAE/kg DW).

2.6. Determination of Total Antioxidant Capacity. For the determination of total antioxidant capacity (TAC), the method based on the radical reaction of 2,2'-diphenyl-1picrylhydrazyl (DPPH) according to [32] was used. To obtain a stock solution, 0.025 g of DPPH was diluted to 100 mL with MetOH (99.8%) and kept in a cold and dark place. Immediately before the analysis, a 1:10 dilution of the stock was made with methanol. For the analysis, 3.9 mL of the DPPH working solution was added to a cuvette and the absorbance at a wavelength of 515.6 nm was measured (A_0) by UV-VIS 1800 spectrophotometer (Shimadzu, Japan). Subsequently, 0.1 mL of the extract was added to the cuvette with DPPH, and the absorbance was measured after 10 min (A_{10}) . An increasing amount of antioxidants present in the methanol extract of the sample reduced DPPH and faded the color of the solution in a correlation proportional to the antioxidant concentration. The percentage of DPPH inhibition was calculated according to the following equation:

% in h. DPPH =
$$\frac{(A_0 - A_{10})}{A_{10}} \times 100$$
, (1)

where A_0 is absorbance at time t = 0 min (solution of DPPH) and A_{10} is absorbance at time t = 10 min.

The antioxidant activity was calculated using a standard curve with known concentrations of Trolox and expressed in terms of μ mols of Trolox equivalents per Gram dry weight.

- 2.7. Determination of Total Anthocyanins Content. Total anthocyanin content (TANTC) was determined according to the pH differential spectroscopic method. Aliquot portions of extract (0.1 mL) were pipetted into two test tubes and 1 mL of 0.01% HCl in 80% EtOH was added to each tube. Subsequently, 10 mL of HCl (2%) was added to the first tube and 1 mL pH 3.5 buffer (Na₂HPO₄, c = 0.2 mol/L and citric acid, c = 0.1 mol/L) was added to the second tube. After 30 minutes of staining the solutions at room temperature, absorption was measured at 520 nm (Shimadzu UV-VIS 1800, Japan). Total anthocyanins content was calculated and expressed in mg CyE/kg (cyanidin eq/kg) FW.
- 2.8. Determination of Phenolic Acids Content (Chlorogenic, Neochlorogenic, and trans-Ferulic Acid). Aliquot volumes of supernatants from sweet potato extracts were filtered through a membrane filter of $0.22\,\mu\text{m}$. Separation of chlorogenic and neochlorogenic acids was carried out by reversed-phase HPLC with a column CORTECS C18 ($150\times4.6\,\text{mm}$; $2.7\,\mu\text{m}$). The column oven temperature was set at 30°C. A sample volume of $8\,\mu\text{L}$ was injected onto the column using a Waters 717 autosampler connected to a Waters pump (Waters Corp., Milford, MA). Phenolic acids were eluted using a mobile phase A 0.1% (v/v) phosphoric acid, B acetonitrile, and C methanol. Detected peaks were

identified and quantified by comparing to retention times and areas of peaks of known standards. The gradient program was set, as shown in Table 1. The results were quantified by using a Waters 2965 UV detector at wavelength 325 nm.

Peaks were identified and quantified by comparison of retention times and areas of peaks of known standards. Results were quantified by using Waters 2965 UV detector at wavelength 326 nm.

2.9. Statistical Analysis. All measurements were done in quadruplicate and presented as mean \pm SD (n=4). Results were statistically evaluated by one-way analysis of variance (ANOVA—Multiple Range Tests, method: 95.0 percent LSD) using statistical software STATGRAPHICS (Centurion XVI.I, USA) and a regression and correlation analysis (Microsoft Excel) was used. Differences at p < 0.05 were considered to be significant.

3. Results and Discussion

3.1. Total Polyphenols Content (TPC). Phenolic compounds are the primary antioxidants found in fruits, vegetables, and grains [28]. Sweet potatoes, which are also a source of many other bioactive substances, are also characterized by a high content of polyphenolic compounds [33]. Raw and heattreated sweet potatoes were analyzed for the total polyphenols content and other bioactive compounds. TPC ranged from 576 mg/kg DW (169 mg/kg FW, resp.) in variety Be/HR to 3828 mg/kg DW in variety Pu/SK (750 mg/kg FW Be/HR, resp.) (Table 2) in the raw sweet potato tubers. The wide range of TPC values in raw flesh of SP can be attributed to several factors. Many authors [17, 34-36] mention the variety as the most significant factor. In particular, purple sweet potatoes are rich in phenolic substances. White, yellow, and orange varieties have a lower content of this group of compounds compared to purple varieties [7].

Comparable polyphenol contents were determined in [30] (0.93–1.05 mg/g DW), [4] (28.4 mg/100 g FW), and [18] (1300-1930 mg/kg DW). Higher TPC were determined in [37] in 15 varieties from Tenerife Island and in 15 varieties of SP from La Palma Island (90-166, resp., 78.8-161 mg phenolic compounds/100 g FW), [17] in orange SP (0.130-0.472 mg/g FW) and purple SP (0.477-0.949 mg/g FW); [26] in orange SP (284.1 mg/100 g DW) and purple SP (757.4 mg/100 g DW). As in our case, the authors report lower TPC in orange sweet potatoes than in purple sweet potatoes. The variation can be attributable to phenolics extraction methods, sweet potato genotypes, and growing conditions [38]. The content of polyphenols in the flesh was significantly affected by the method of treatment of sweet potatoes. There are statistically significant differences between the TPC of raw and heat-treated SPs in all varieties. However, it can not be clearly stated which heat treatment had the greatest effect on the change of TPC in sweet potatoes. In all heat-treated SP except for Be/HR (boiled), TPC was higher than in raw flesh. Only in SP Be/HR cooked in

TABLE 1: Gradient profile.

Minutes	A (%)	B (%)	C (%)
0	90	0	10
3	80	10	10
5	40	25	5
8	50	30	20
10	10	80	10
12	0	90	10

water was TPC 39% lower compared to raw SP. In this variety, the effect of heat treatment was the least (maximum 24% in baked SP). In the Pu/SK variety, the TPC in microwaved sweet potatoes was up to 5.5 times higher than in raw flesh. The slight increase of phenolic content in cooked samples can be attributed to the release of bound phenolics and inactivation of polyphenol oxidase affected by heat treatment. Besides, some phenolics can be degraded by polyphenol oxidase during slicing of raw sweet potatoes samples [38]. C. Dincer et al. [30] report the highest TPC in the boiled samples for all cultivars (up to 1.98 mg GAE/g DW). The authors explained this definite increase of TPC as the release of phenolics by hydrolysis of glycoside bonds during treatment and the induction of TPC oxidation in fresh samples through the catalytic activity of the enzyme polyphenol oxidase. C. M. Donado-Pestana et al. [18] determined the content of total phenolic compounds in boiled SP from 1.33 to 2.05 mg/g DW and in steamed SP from 1.05 to 1.56 mg/g DW; while boiling increased TPC in 2 varieties and decreased TPC in 2 varieties, steaming decreased TPC in all varieties. Their results suggest that the methods of heat treatment sometimes led to a significant loss of the total phenolic content in sweet potato tubers. Y. Tang et al. [7] reported a decrease of TPC in orange SP of 60.6% (boiled SP) and 15.9% (steamed SP), resp., and in purple SP of 31.3% (boiled SP) and 6.0% (steamed SP). The study in [39] determined in raw pith 2.1 mg TPC/g DW and in microwaved (boiled, baked) potato root tissues 1.8 (1.6, 2.1) mg TPC/g DW. The study in [40] studied 12 different ways of preparation (freezing, cooking, baking, etc.) of 15 types of food. Most of the studied processes produced a wide range of retentional factors (ratio of concentration of polyphenol in processed food to centration of polyphenol in raw food). Home cooking methods of common plant foods caused significant losses of polyphenols (median RF = 0.45-0.70).

3.2. Total Antioxidant Capacity (TAC). Variety and locality are essential factors influencing an antioxidant activity. The purple SP showed higher TAC, whereas TAC in Pu/HR was 2.7 times higher than in Pu/SK. On the other hand, TAC was 2.2 times higher in the Beauregard variety grown in Slovakia than in Be/HR. There are statistically significant differences between TAC values (Table 3).

However, the impact of the technological treatment method on TAC can not be clearly stated. Based on the DPPH test, the raw SP flesh Be/HR showed the lowest antioxidant activity (0.848 μ mol TE/g DW, 212 μ g TE/g DW) and the highest TAC had boiled SP Be/SK (14.2 μ mol TE/g

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IABLE Z:	me totai	polyphenois (content (III	g GAE/kg DW).

	Raw	Boiled	Steamed	Microwaved	Baked
Be/SK	$1102 \pm 21.2^{a,B}$	$1762 \pm 1762^{c,B}$	$1770 \pm 34.4^{c,B}$	$1542 \pm 22.0^{b,B}$	$2065 \pm 38.2^{d,B}$
Pu/SK	$2056 \pm 114^{a,C}$	$3667 \pm 26.0^{b,C}$	$8438 \pm 839^{\rm d,D}$	$11338 \pm 371^{e,D}$	$7212 \pm 256^{c,D}$
Be/HR	$576 \pm 53.6^{b,A}$	$353 \pm 67.0^{a,A}$	$692 \pm 81.4^{c,A}$	$710 \pm 63.6^{c,A}$	$714 \pm 51.2^{c,A}$
Pu/HR	$2444 \pm 72.0^{a,D}$	$4748 \pm 40.5^{c,D}$	$5461 \pm 185^{e,C}$	$5070 \pm 109^{d,C}$	$3692 \pm 93.8^{b,C}$

GAE: gallic acid equivalent; Be/SK: cv. Beauregard from Slovakia; Be/HR: cv. Beauregard from Croatia; Pu/SK: cv. 414-purple from Slovakia; Be/SK: cv. 414-purple from Croatia.

Table 3: Total antioxidant capacity (μ mol TE/g DW).

	Raw	Boiled	Steamed	Microwaved	Baked
Be/SK	$1.81 \pm 0.239^{a,B}$	$14.2 \pm 0.178^{e,D}$	$10.9 \pm 0.195^{d,C}$	$5.50 \pm 0.073^{b,B}$	$5.80 \pm 0.062^{c,D}$
Pu/SK	$3.23 \pm 0.433^{a,C}$	$6.20 \pm 0.041^{c,B}$	$10.9 \pm 0.293^{e,C}$	$8.63 \pm 0.104^{d,C}$	$5.35 \pm 0.076^{b,C}$
Be/HR	$0.848 \pm 0.225^{a,A}$	$1.85 \pm 0.249^{b,A}$	$4.60 \pm 0.430^{c,A}$	$5.25 \pm 0.225^{\mathrm{d,A}}$	$5.17 \pm 0.186^{d,B}$
Pu/HR	$8.67 \pm 0.097^{\mathrm{d,D}}$	$11.3 \pm 0.120^{e,C}$	$7.14 \pm 0.735^{c,B}$	$5.19 \pm 0.039^{b,A}$	$4.74 \pm 0.051^{a,A}$

The values in the row marked with different lowercase letters are significantly different (p < 0.05); the values in the column marked with different uppercase letters are significantly different (p < 0.05). TE: Trolox equivalent.

DW, 3546 µg TE/g DW). Except for Pu/HR (steamed, microwaved, and baked), the highest TAC was in all heattreated SP. The study in [38] determined TAC in raw flesh of orange SP in the range from 1.1 to 2.0 µmol TE/g FW and in the boiled SP from 1.7 to $2.7 \mu \text{mol TE/g FW}$, which is an increase of 1.28-1.55 times of TAC after heat treatment. According to [41], there was no significant difference between the DPPH radical scavenging capacity of steamed and boiled sweet potato and untreated sweet potato. L. E. Steed and V.-D. Truong [22] determined the highest antioxidant activity (87.4 μ mol TE/g FW) in raw peels. There was no significant difference between DPPH values in raw $(75.5 \,\mu\text{mol TE/g FW})$ and steamed SP $(77.1 \,\mu\text{mol TE/g FW})$. In comparison with our result, [7] determined higher values of TAC in sweet potatoes (orange—raw: 25.07, purple—raw: $27.79 \,\mu\text{mol}\,\text{TE/g}\,\text{FW}$). The heat treatment resulted in a reduction of TAC (orange-boiled: 23.62, steamed: 15.22; purple—boiled: 22.11, steamed: 26.04 µmol TE/g FW). Individual heat treatments have different effects on the chemical composition. That is, after boiling and microwaving, 63.82% and 32.25% decreases of antioxidant activity were observed, while the antioxidant activity increased by 81.40%, 30.09%, respectively, and by 85.82% (frying). An increase of 9.44% in total polyphenol content was observed after steaming, while decreases of 30.51%, 25.70%, and 15.73% were observed after boiling, microwaving, and frying [25]. Heat treatment of SP possibly leads to a reduction of bioactive compounds content and, consequently, to a decrease of antioxidant capacity determined by the DPPH and ABTS tests. The antioxidant capacity can also be associated with the presence of phenolic acids in sweet potato roots. The processing of SP tubers (peeling, boiling, frying, and steaming) can also reduce the antioxidant capacity by the enzyme polyphenol oxidase, which catalyses the oxidative polymerization of phenolic acids [18]. Compared to raw SP (92.0--132.3), heat-treated SP showed a decrease of antioxidant capacity (DPPH, IC50) to 74.1-88.2 in boiled and to 62.2-86.3 mg/mL in baked SP [30]. The authors report that the differences between TAC values in different varieties

treated by the same method may be associated with various polyphenolic compounds that might have different sensitivity to heat treatment.

3.3. Total Anthocyanins Content (TANTC). The content of anthocyanins was determined only in purple varieties 414-purple grown in Slovakia and Croatia. The content of anthocyanins in PU/HR was almost 1.6 times higher than in Pu/SK, which indicates a significant effect of locality—the differences in TANTC in raw SP are statistically significant (Table 4). C. C. Teow et al. [17] determined anthocyanins in purple varieties SP from 0.246 to 0.531 mg/g FW, while the study in [42] reported the average content of anthocyanins in purple SP from 110 mg to 210 mg/100 g; the study in [26] reported 57.9 mg/100 g DW; and the study in [8] reported 32 to 1390 mg/100 g DW.

Anthocyanins are highly reactive molecules sensitive to degradation reactions. Their stability is affected by the structure and concentration of anthocyanins, the presence of enzymes, oxygen, pH, or temperature [43, 44]. Most authors report the reduction of anthocyanin content due to various heat treatments [7, 45]. Y. Tang et al. [7] determined a 34% lower content of anthocyanins in steamed SP (10.35 mg CyE/ g) and a 41% lower content of anthocyanins in boiled SP (9.24 mg CyE/g) compared to raw deep purple SP. On the contrary, the results of our analyses show an increase in the content of anthocyanins after heat treatment of SP. The average TANT in raw SP Pu/SK (Pu/HR) increased 3.7 (2.4) times by boiling in water, 4.3 (4.0) times by steaming, 6.6 (3.7) times by baking, and 8.5 (4.8) times by microwaving. Cooking in the microwave oven had the most significant effect. The content of anthocyanins in the microwaved SP was 1276 mg/kg FW in Pu/SK and 1194 mg/kg FW in Pu/ HR. Paper [23] reported a significant increase in the total anthocyanin content of boiled colored potatoes compared to fresh uncooked tubers of five studied cultivars. The highest increases were observed in cultivars with low TAC (11.1 times and 10.6 times), while in the cultivars with high TAC,

TABLE 4: Total anthocyanins content (mg CyE/kg FW).

	Raw	Boiled	Steamed	Microwaved	Baked
Pu/SK	$151 \pm 4.55^{a,A}$	$552 \pm 30.7^{b,A}$	$690 \pm 51.4^{c,A}$	$1276 \pm 9.16^{e,B}$	$1004 \pm 10.4^{\mathrm{d,B}}$
Pu/HR	$238 \pm 28.4^{a,B}$	$575 \pm 13.7^{b,A}$	$959 \pm 14.4^{d,B}$	$1147 \pm 10.1^{e,A}$	$874 \pm 16.0^{c,A}$

The values in the row marked with different lowercase letters are significantly different (p < 0.05); the values in the column marked with different uppercase letters are significantly different (p < 0.05). CyE: cyanidin equivalent.

an increase was relatively low (3.44 times and 3.20 times). Paradoxically, most anthocyanins have greater stability at higher temperatures used in the processing of fruit and vegetables. This phenomenon is explained by the protective effect of the various system components and the condensation of monomers. In these reactions, more stable oligomeric pigments are formed, the amount of which increases with temperature [46]. The thermal stability of anthocyanins is affected by other factors; one of the main factors in the processing of fruit and vegetables is pH value. Anthocyanins of purple SP achieved the highest stability at pH value from 3 to 4 [8]. The red cultivar of sweet potato grown in the Andean region has been reported to have higher antioxidant activity and phenolic content than blueberry cultivar, the fruit with a high level of antioxidants [47]. The antioxidant capacity of sweet potato roots can vary widely depending on the flesh color, which can differ among sweet potato cultivars from white and yellow to orange and even purple [17].

On the contrary, there are significantly fewer studies where the authors describe the preservation or increase of their content due to cooking, baking, steaming, or microwave heating. The stability of anthocyanins in berries is lower, such as the stability of anthocyanins in radish, red potatoes, red cabbage, and purple SP [48-50] because, in PSP, anthocyanins occur mainly in acylated forms [51]. Acylation with various phenolic (cinnamic, p-coumaric, caffeic, and ferulic) and aliphatic (acetic, malonic, and oxalic, as well as succinic) acids makes these anthocyanins more resistant to pH, sensitivity to light and heat [52, 53]. The study in [51] determined the stability of twelve individual anthocyanins in raw P40 and cooked P40 via various cooking conditions. Although some heat treatments caused the degradation of total anthocyanins, the content of individual anthocyanins increased several times, for example, by microwave cooking the content of cyanidin 3-p-hydroxybenzoyl sophoroside-5-glucoside 3.81-fold and peonidin 3p-hydroxybenzoyl sophoroside-5-glucoside 4.57-fold, or by baking the content of cyanidin 3-(6''-feruloyl sophoroside)-5-glucoside 2.75-fold. Monoacylated anthocyanins showed higher heat resistance than di- and nonacylated ones. Similarly, [54] report an almost 50% reduction in total PSP Shinzani anthocyanins after steaming, but only slightly after baking, and a tendency to increase some acylated anthocyanins (cyanidin 3-(6'-feruloyl sophoroside)-5-glucoside: 1.23-fold). The study in [55] investigated the effects of several representative home cooking methods on the content and composition of anthocyanins. Air-fried, fried, and stirfried cooking resulted in a reduction in anthocyanin content and the effect of steaming and microwaving on anthocyanin content was inconclusive, but cooking increased anthocyanin content by 6.55%. The application of different cooking

methods caused changes in the content of individual anthocyanins. For example, the content of cyanidin 3-dicaffeoyl sophoroside-5-glucoside (the main component of the analyzed SP GZ9) increased by 8.83% after cooking. Interestingly, nearly all monoacylated anthocyanins increased significantly after boiling and microwaving. On the other hand, total diacylated anthocyanins decreased significantly after frying, air-frying, and stir-frying. The effect of SP (steamed and kneaded sweet potato flour) treatment was manifested by a five-to-six-time increase in the content of anthocyanins in red peeled (RP) and white peeled (WP) SP, a 1.3-fold increase in the content of flavonoids in RP, and a 2-13-fold increase in the total phenols contents of all genotypes [35]. Effect of three cooking methods on the content of anthocyanins in potato tubers with red- and purplefleshed has been studied [56]. Cooking treatments resulted in a significant increase in total anthocyanin content in all cultivars in comparison with raw tubers. In the average of all cultivars, the greatest TAC increase was found as a result of the boiling of tubers (3.79 times against the value of raw tubers), followed by the microwave treatment (3.06 times), and the lowest increase in baked tubers (2.94 times) has been observed. While the most significant impact on TAC increase by boiling was observed in all cultivars except for Violette, higher TAC increase by microwaving was found only in three of the five cultivars. As the authors further state, there appears to be a consensus on the loss of anthocyanins upon exposure to heat (there appears to be a consensus on the loss of anthocyanins when exposed to heating). Therefore, it is necessary to continue doing the experiments, while the results will be the subject of our further research.

3.4. Phenolic Acids: Chlorogenic (CGA), Neochlorogenic (Neo-CGA), and trans-Ferulic (tFA) Acid. Phenolic acids show varied biological activity in the human body; among others, they take an active part in the removal of free radicals and metal ion chelation as well as affecting enzyme activity and protein availability [57]. Chlorogenic acid and its derivatives are esters of caffeic acid. These acids are the main component of sweet potato phenolic compounds. Chlorogenic acid and isochlorogenic acid are the primary acids [17, 26, 28]. Precision or repeatability of retention times and peak areas from five standards was calculated using percent relative standard deviation (% RSD) and is presented in Table 5.

The proportion of phenolic acids in the raw SP was different. The CGA content was the highest in the variety of Pu/HR (468 mg/kg) and decreased in order Be/HR > Be/SK > Pu/SK (147 mg/kg). The content of neo-CGA and *trans*-ferulic acid decreased in reverse order as follows: Pu/SK (7.26 and 10.4 mg/kg) > Be/SK > Be/HR > Pu/HR (0.768

Table 5: Validation parameters HPLC-DAD, analytical characteristics, and method validation data for HPLC using a Cortex $2.7 \,\mu\text{m}$, $4.6 \times 150 \,\text{mm}$ column.

Compound	Accuracy (% recovery)	Precision (% RSD)	Rt (min)	LOD (µg/mL)	LOQ (µg/mL)
tFA	98	2.897	5.2	0.0251	0.0753
CGA	115	5.564	2.3	0.0149	0.0447
Neo-CGA	113	4.326	2.8	0.0072	0.0216

RSD: relative standard deviation; RT: retention time; LOD: limit of detection; LOQ: limit of quantification.

TABLE 6: Content of phenolics acid (mg/kg FW).

	Raw	Boiled	Steamed	Microwaved	Baked			
Chlorogenic	Chlorogenic acid (CGA)							
Be/SK	$336 \pm 21.9^{b,B}$	$234 \pm 28.7^{a,B}$	$347 \pm 75.5^{b,B}$	$212 \pm 13.1^{a,B}$	$238 \pm 33.0^{a,B}$			
Pu/SK	$147 \pm 16.2^{c,A}$	$104 \pm 5.26^{a,A}$	$114 \pm 5.89^{ab,A}$	$109 \pm 6.15^{ab,A}$	$118 \pm 10.8^{b,A}$			
Be/HR	$362 \pm 22.4^{c,C}$	$281 \pm 52.7^{a,C}$	$344 \pm 7.55^{bc,B}$	$302 \pm 5.97^{a,C}$	$330 \pm 4.93^{b,C}$			
Pu/HR	$468 \pm 26.1^{c,D}$	$441 \pm 17.0^{\mathrm{b,D}}$	$414 \pm 8.55^{a,C}$	$414 \pm 6.26^{a,D}$	$470 \pm 11.1^{c,D}$			
Neochloroge	enic acid (neo-CGA)							
Be/SK	$6.23 \pm 0.268^{d,C}$	$3.29 \pm 0.107^{a,D}$	$5.38 \pm 0.143^{c,C}$	$4.51 \pm 0.152^{b,C}$	$3.29 \pm 0.098^{a,C}$			
Pu/SK	$7.26 \pm 0.164^{d,D}$	$2.60 \pm 0.326^{a,C}$	$5.35 \pm 0.099^{c,C}$	$6.31 \pm 0.150^{d,D}$	$4.31 \pm 0.076^{\mathrm{b,D}}$			
Be/HR	$3.30 \pm 0.103^{\mathrm{d,B}}$	$1.25 \pm 0.069^{a,B}$	$2.13 \pm 0.060^{b,B}$	$2.27 \pm 0.065^{c,B}$	$2.17 \pm 0.092^{b,B}$			
Pu/HR	$0.768 \pm 0.055^{e,A}$	$0.235 \pm 0.033^{a,A}$	$0.710 \pm 0.050^{d,A}$	$0.488 \pm 0.048^{c,A}$	$0.413 \pm 0.015^{b,A}$			
trans-Ferulic acid (tFA)								
Be/SK	$8.58 \pm 0.120^{d,C}$	$7.28 \pm 0.021^{a,C}$	$8.33 \pm 0.022^{c,C}$	$8.18 \pm 0.066^{b,C}$	$7.31 \pm 0.061^{a,C}$			
Pu/SK	$10.4 \pm 0.155^{d,D}$	$7.86 \pm 0.111^{a,D}$	$9.68 \pm 0.129^{c,D}$	$9.55 \pm 0.221^{c,D}$	$8.13 \pm 0.087^{\mathrm{b,D}}$			
Be/HR	$5.85 \pm 0.078^{e,B}$	$4.14 \pm 0.066^{a,B}$	$4.91 \pm 0.026^{c,B}$	$5.63 \pm 0.083^{\mathrm{d,B}}$	$4.64 \pm 0.144^{\mathrm{b,B}}$			
Pu/HR	$3.30 \pm 0.044^{e,A}$	$4.14 \pm 0.066^{a,B}$	$3.24 \pm 0.023^{d,A}$	$3.17 \pm 0.025^{c,A}$	$2.96 \pm 0.040^{b,A}$			

The values in the row marked with different lowercase letters are significantly different (p < 0.05); the values in the column marked with different uppercase letters are significantly different (p < 0.05).

and 3.30 mg/kg). These differences confirm not only the influence of variety but also the influence of growing conditions (Table 6).

The heat treatment of SP had a negative effect on the content of phenolic acids. The most considerable decrease of CGA content was recorded in the variety of Pu/SK (boiled SP: -29%). In the variety Pu/HR, the CGA content in the microwaved SP was comparable to raw SP and in the variety Be/SK (steamed SP), the CGA content was 3% higher than in raw SP. However, these differences are not statistically significant and it can not be clearly stated which method of heat treatment has the least/most significant effect on the CGA content. In the case of the other two phenolic acids, cooking in the water had the greatest effect on their content. The neo-CGA content in boiled SP decreased by 47% (64, 62, and 69) in the variety Be/SK (Pu/SK, Be/HR, and Pu/HR, respectively). The largest reduction of tFA acid content was in the Be/HR variety (-29%). In general, its content in SP decreased in the order: raw > steamed > microwaved > baked > boiled. A similar effect of heat treatment is reported by [57]. Home cooking methods of SP caused a significant loss of CGA and ferulic acid. The greatest decrease of CGA content compared to raw SP (463.7 mg/100 g DW) was observed after baking (38.52%), followed by boiling and cooking in the microwave oven (21.04% and 20.01%, resp.) while cooking in steam caused only a slight increase (p < 0.05). Compared to ferulic acid content determined in raw SP (24.36 mg/100 g DW), steaming and microwaving caused only a slight change in its content, while other

cooking methods induced a significant decrease (p < 0.05). M. S. Padda [39] reported a significant decrease in CGA content after processed sweet potato pith tissue. Its content (mg/g DW) decreased as a result of heat treatment in the order: raw (0.46) > microwaved (0.34) > baked (0.26)> boiled (0.20). The authors of [28] evaluated the effect of heat treatment with distilled water and methanol A on several phenolic acids, e.g., vanillic acid, protocatechuic acid, gallic acid, chlorogenic acid, and caffeic acid. After heat treatment with distilled water, as protocatechuic acid and chlorogenic acid content decreased, on the contrary, the content of minor phenolic acids of SP—vanillic acid, gallic acid, and caffeic acid-increased. Paper [58] reported the most significant reduction of CGA content in boiled SP and the smallest in microwaved SP (raw 45.7, microwaved 33.9, boiled 19.7, and baked 26.5 mg/100 g DW). In contrast, [41] reported a significant increase in CGA content in SP after steaming (7.99 \pm 0.45 mg/g DW) and boiling (5.16 \pm 0.18 mg/ g DW) compared to untreated SP $(2.34 \pm 0.09 \text{ mg/g DW})$. F. Rautenbach et al. [59] determined 28.7 and 47.1% higher CGA content in boiled SP of variety Resisto and W-119 compared to raw SP (69.6 \pm 4.8, 89.3 \pm 5.4 μ g/g FW). Increased contents of CGA and neo-CGA after different cooking methods were determined in three potato cultivars (Bintje, Piccolo, and Purple Majesty) by [60]. The higher CGA content was in baked potatoes and decreased in the order steamed > boiled > microwaved > raw. The neo-CGA content decreased in the order boiled > steamed > baked > microwaved > raw. L. E. Steed and V.-D. Truong [22]

determined 5.1–9.3 (raw flesh) and 10.6–16.2 (cooked flesh) mg CGA/100 g in three SP varieties. They also report higher contents of other phenolic acids (CQA, 4,5-di-CQA, 3,5-di-CQA, and 3,4-di-CQA) due to cooking. They hypothesize that these changes in the content of individual phenolic acids in SP affect the antioxidant properties, including ABTS and DPPH radical scavenging activities. It can be assumed that these changes in the content of individual phenolic acids in sweet potatoes according to cultivar and heat treatment conditions affect the antioxidant properties, including DPPH radical scavenging activities [28].

4. Conclusions

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Based on obtained results, it can be stated that specific methods of SP treatment differently affected the content of bioactive compounds such as polyphenols, phenolic acids, and anthocyanins and their antioxidant effects. Due to boiling, steaming, microwaving, and baking, the content of polyphenolic and anthocyanins and also antioxidant activity increased almost in all samples compared to raw sweet potatoes. In contrast, the content of CGA, neo-CGA, and tFA was lower in heat-treated than in raw sweet potatoes. In addition to degradation, their losses are caused by transport from tissue to the surrounding water and also by the type of phenolic acid. It is not possible to determine which method of heat treatment is the gentlest for the preservation of bioactive substances in sweet potatoes.

Data Availability

All basic data supporting the results of this study are available from the corresponding author.

Conflicts of Interest

The authors declare that there are no conflicts of interest regarding the publication of this paper.

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