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Analysis of chemical compounds' content in different varieties of carrots, including qualification and quantification of sugars, organic acids, minerals, and bioactive compounds by UPLC

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Abstract

Twelve carrot varieties in different colours and sizes were investigated for chemical properties (dry matter, ash, pectins, titratable acidity, and pH), contents of vitamin C, sugar, organic acids, mineral (sodium, potassium, calcium, iron, and magnesium), and anti-oxidant activities (ABTS, FRAP, and ORAC). Moreover, total polyphenolics and total tetraterpenoids of colourful carrot varieties were presented. According to the study, sucrose was the dominant sugar and isocitric acid was the most common organic acid in carrot samples. In the case of mineral content, potassium, sodium, calcium, magnesium, and iron were identified, while copper was not identified in carrots. Additionally, most of the analyzed carrots were a good source of pectins (average—1.3%), except for mini-orange carrot. Purple-coloured carrot samples demonstrated the highest results for total sugar (11.2 g/100 g fm), total organic acid (2.8 g/100 g fm), total polyphenolic contents (224.4 mg/100 g fm), and anti-oxidant activities (17.1 mmol Trolox equivalents/100 g dm). In turn, the lowest results were observed in normal yellow carrot for total polyphenols (7.3 mg/100 g fm), and anti-oxidant activities (2.5 mmol Trolox equivalents/100 g dm); besides, the lowest total tetraterpenoids were determined in micro-white carrot—0.2 mg/100 g fm.

Keywords Carrot roots · Chemical properties · Polyphenolic compounds · Carotenoids · Antioxidant activity

Introduction

Global dietary guidelines support boosting fruit and vegetables consumption and recommend five portions of fruit and vegetables per day [1]. It results from the fact that fruit and vegetables consumption prevent non-communicable diseases such as cardiovascular disorders, obesity, diabetes, and cancer [2]. However, many people do not consume fruit and vegetables for varied reasons such as price, eating habits, time, and concerns about fruit and vegetables production [3]. Thus, poor diet is the one reason to cause 80% of death from non-communicable diseases [4].

Childhood is a period when people can gain healthy eating habits from their parents [5]. Parental nutritional knowledge increases the consumption of more fruit, vegetables,

and fiber intakes of children [6]. Besides, the food choices of parents influence the eating habits of children as well [7]. For instance, to increase the fruit and vegetables consumption of children, parents may purchase more colourful fruit and vegetables. Moreover, family members can consume these raw materials together to gain more healthy eating habits. Hence, rainbow-healthy raw foods promote people to enhance their diet [8].

To clarify, colourful carrot varieties may apply with their elevated health benefits. Carrot breeding provides purple, orange, red, yellow, and white colours [9]. Besides, the chemical contents of carrots change depending on colours; carotenoids create orange and yellow colours [10, 11], purple colour is the result of anthocyanins [12], and white carrot contains no colour pigments [13]. Except for colour features, bioactive compounds of carrots are precious with activities against diabetes, cardiovascular diseases, obesity, cancers, and aging [14, 15].

Like other fruit and vegetables, nutrients in carrots change with genes, environmental factors, fertilizer, and storage periods [16]. Carrot roots are rich in different carotenoid types as well as polyphenols [17]. However, carrot



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varieties are not restricted to those compounds. Hence, carrots are valuable with vitamin and mineral contents as well.

The mineral content determines product quality which is an important parameter for raw food products [18]. Carrot roots are abundant in potassium (K), manganese (Mn), phosphorus (P), calcium (Ca), sodium (Na), iron (Fe), and magnesium (Mg) [19, 20]. These elements are essential minerals for human healthy body functions. Proof of these, K is necessary for functions of muscles, nerves, and cells; Mn is required in trace amounts for biosynthetic pathways; P shows functions for blood vessels and bones; Ca is essential for bones, teeth and blood cells; Na is necessary for functions of muscles and nerves; Fe is important for oxygen transportation in blood and muscle cells; besides, Mg adjusts blood sugar levels and blood pressure [21].

Vitamins in carrot varieties are another important content for product quality. Moreover, the main vitamin types in carrot varieties are the results of carotenoids. For instance, β -carotene is converted to vitamin A which is important against night blindness and increases immune system functions [22]. Second, α -tocopherol is the precursor of vitamin E that is crucial for cell signalling, gene expression, and cell membrane stabilities in the human body [23]. Except for these two vitamins, carrots are abundant with vitamin C which is necessary for controlling blood pressure, preventing iron deficiency and boosting immune system functions [20, 24]. Moreover, vitamin B derivatives (thiamine, riboflavin, cobalamin, and pyridoxine) are rich in carrot varieties as well [25]; and vitamin B is important for the functions of cell growth, brain, and digestion system.

Besides, carrots are good sources of carbohydrates such as simple sugars like fructose, glucose, and sucrose, a small amount of starch and fibers [26]. Sugar contents and quantities of carrot varieties may change with the impact of environmental and storage conditions [27]. Dietary fibers, which provide healthy bowel function, decreasing cholesterol level, and heart diseases, are grouped as soluble and insoluble. For instance, pectin and hemicellulose are soluble; cellulose is insoluble fiber [28]. Moreover, mono-, di-, or oligo-saccharides of fruit and vegetables are categorized as prebiotics by CODEX Alimentarius [29].

On the other hand, fruit and vegetables are rich in organic acids. The most known acid of raw materials is ascorbic acid (vitamin C) with its high anti-oxidant activities. However, many others boost healthy body functions, as well. For instance, benzoic acid demonstrates antibacterial activity; hydroxycinnamic acid is an anti-inflammatory agent; gallic acid fights with mutagenic factors. Moreover, acetic, succinic, citric, lactic, malic acids, and their salts help to iron absorption [30].

Therefore, the study aims to compare 12 coloured carrot varieties with their chemical properties (dry matter, ash, pectin, titratable acidity, and pH), and anti-oxidant

activities (ABTS, FRAP, and ORAC). Moreover, carrot varieties have been contrasted for total polyphenolic compounds, vitamin C, and total tetraterpenoid contents. Additionally, sugar, mineral, and organic acid profiles of carrot varieties have been presented. The study is the first to evaluate the chemical contents and anti-oxidant activities of colourful carrot varieties together.

Materials and methods

Chemicals

Standards of sugars, organic acids, carotenoids, chlorophylls, and polyphenolics were purchased from extrasynthese (Lyon, France). Acetonitrile, methanol, and formic acid for analyses of ultra-performance liquid chromatography (UPLC; Gradient grade) and ascorbic acid were purchased from Merck (Darmstadt, Germany). The additional reagents were purchased from Sigma-Aldrich (Taufkirchen, Germany).

Plant material and sample preparation

Normal and mini-sized carrots were purchased from Fusion Gusto (Dąbrowa, Poland). Micro carrots were purchased from Cato Produce (Johannesburg, South Africa) in June 2020. Normal-sized carrots were between 20 and 45 mm and weights from 50 to 150 g; mini-sized carrots were between 10 and 20 mm and weights from 8 to 50 g; micro-sized carrots were smaller than 10 mm and weights were smaller than 8 g.

The following varieties of carrot were investigated: micro-yellow carrot (MYC), mini-yellow carrot (MiYC), normal yellow carrot (NYC); micro-purple carrot (MPC), mini-purple carrot (MiPC), normal purple carrot (NPC); micro-orange carrot (MOC), mini-orange carrot (MiOC), normal orange carrot (NOC); micro-white carrot (MWC), mini-white carrot (MiWC), and normal-white carrot (NWC).

Approximately 2 kg of carrot roots were separated into two parts. One part of carrot varieties was analyzed for the contents of dry matter, ash, soluble solids, pH, titratable acidity, pectin, L-ascorbic acid, sugars, and organic acids. The second part of carrot roots was washed, dried, cut into slices, and then frozen at $-80\,^{\circ}$ C. The sliced carrots were then freeze-dried (24 h; Christ Alpha 1–4 LSC, Melsungen, Germany) and crushed by a laboratory mill (IKA A 11, Staufen, Germany) to obtain the homogeneous dry material for analysis.



Physicochemical analyses

Titratable acidity (TA) was evaluated by 0.1 N NaOH to an endpoint of pH 8.1 using an automatic pH titration system (pH-meter type IQ 150; Warsaw, Polska) and expressed as g malic acid/100 g FW (fresh weight). The pH of carrots was measured using the same equipment as that used for TA. The dry matter was estimated by mixing the sample with diatomaceous earth, pre-dried, and final drying under reduced pressure. The dry matter and TA were determined using the following PN norms: PN-EN 12,145:2001 and PN-EN 12,145:2000, respectively. Pectin content (mg/100 g FW) was evaluated according to Pijanowski et al. [31]. The contents of ash (%), L-ascorbic acid (mg/100 g FW), sugars, and organic acids (g/100 g FW) were determined according to Wojdyło et al. [32]. The content of sugars was estimated by HPLC-ELSD (Merck, Hitachi, Japan). The content of organic acids in carrots was quantified by Ultra Performance Liquid Chromatography with Photodiode Array Detector (Acquity UPLC System, Waters Corp., Milford, MA, USA). Organic acids were identified using reference standards (Merck KGaA, Darmstadt, Germany). The content of minerals in carrots was determined using Atomic Absorption Spectrophotometers (AA-7000F/AAC SHIMADZU, Shimadzu Corporation). All measurements were performed in triplicate.

Quantification of bioactive compounds

The quantification (UPLC/PDA/FL—ACQUITY UPLC BEH C18 column (1.7 μ m, 2.1 × 100 mm, Waters Corporation, Milford, USA) at 30 °C. The injection and elution of the samples (5 μ L) were concluded in 15 min with a sequence of linear gradients and a flow rate of 0.42 mL/min. The solvent A (2.0% formic acid, v/v) and solvent B (100% acetonitrile) comprised the mobile phase. The procedure operated through gradient elution with 99–65% solvent A (0–12 min), solvent A was later lowered to 0% for condition column (12.5–13.5 min), and the gradient returned to the initial composition (99% A) for 15 min to re-equilibrate the column) of polyphenolics was performed according to Wojdyło et al. [33]. The polyphenols' quantification was performed by external calibration curves and reference standards.

The quantifications (UPLC-PDA) of tetraterpenoids were performed according to Wojdyło et al. [34] by retention times and spectra to compare with authentic standards. The determination of tetraterpenoids was performed according to Kolniak-Ostek [35]. The total values of polyphenols and carotenoids were given as fresh weights (FW) after the identification and quantification of each bioactive compound. The results are expressed as mg/100 g FW.

Determination of anti-oxidant activity of carrot varieties

The ORAC (oxygen radical absorbance capacity), ABTS + (2,2'-azino-bis(3-ethylbenzothiazoline-6-sulphonic acid)), and FRAP (the ferric reducing ability of plasma) antioxidant activity assays were performed as described, respectively [36–38]. The results of anti-oxidant activity tests are shown as mmol of Trolox equivalents (TE) per 100 g of dry matter (dm). All tests were performed in triplicate using a microplate reader SynergyTM H1 (BioTek, Winooski, VT, USA).

Statistical analysis

The two-way analysis of variance (ANOVA, $p \le 0.05$) and Duncan's test were performed by Statistica version 13.3 (Stat-Soft, Cracow, Poland). The results are shown as the mean value $(n=3) \pm \text{standard deviation (SD)}$.

Results and discussion

Quantification of bioactive compounds in different varieties of carrot

The total L-ascorbic acid, total polyphenolic, and total tetraterpenoid contents of carrot roots are shown in Fig. 1.

Humans cannot synthesise L-ascorbic acid and should obtain the vitamin from other sources such as fruits and vegetables [39]. Besides, the recommended daily intake of ascorbic acid is 75 mg/day for an adult woman and 90 mg/ day for an adult man [40]. In the present study, the content of L-ascorbic acid ranged from 1.0 to 5.3 mg/100 g FW. The highest values were observed for MYC (5.3 ± 0.0) a mg/100 g FW), followed by MPC (5.0 ± 0.0 b mg/100 g FW) and MOC $(3.6 \pm 0.0 \text{ c mg/}100 \text{ g FW})$. Conversely, the lowest results were detected for NYC $(1.2 \pm 0.4 \text{ l mg}/100 \text{ g})$ FW) and NWC (1.0 \pm 0.1 m mg/100 g FW). According to Alasalvar et al. [26], white carrot (1.3 mg/100 g FW) had the lowest ascorbic acid content, while orange carrot (5.3 mg/100 g FW) showed the highest amount of L-ascorbic acid. However, the authors could not quantify the ascorbic acid content in the purple carrot. In the present study, darkcoloured carrot samples exhibited much higher L-ascorbic acid content than other carrots, and similar results have been reported in the literature [41]. Moreover, micro- and minisized carrot samples showed better results for L-ascorbic acid content than normal-sized carrot samples. In another study [42], the contents of L-ascorbic acid of six carrot varieties were compared during the storage periods. The result of the study demonstrated that after harvesting, L-ascorbic acid levels of carrot roots ranged from 54 to 132 mg kg⁻¹. However,



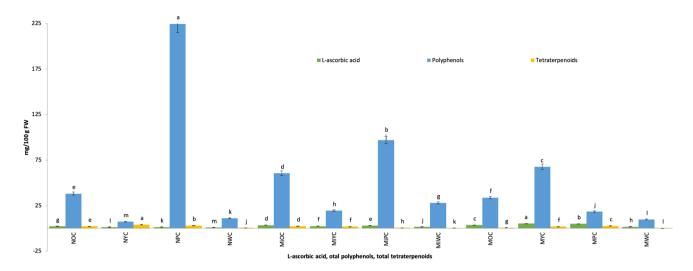


Fig. 1 Total L-ascorbic acid, total polyphenol, and total tetraterpenoid contents in colourful carrot varieties. The L-ascorbic acid, total polyphenol, and total tetraterpenoid contents (mg/100 g FW) in 12

carrot varieties; significant at $p \le 0.05$; ‡ values (mean of three replications) followed by the same letter were not significantly different (p > 0.05) according to Duncan's test

after 30 days of storage, L-ascorbic acid levels decreased by almost 50%. Therefore, the content of L-ascorbic acid changes depending on variety, carbon dioxide, temperature, storage, and age [43].

The total phenolic contents of carrot roots ranged from to 224.4 mg/100 g FW. The NPC (224.4 \pm 9.5 a mg/100 g FW) and MiPC (97.0 ± 4.1 b mg/100 g FW) had the highest total polyphenolic contents. However, NYC $(7.3 \pm 0.3 \text{ m mg}/100 \text{ g FW})$ and MWC $(9.8 \pm 0.41 \text{ mg}/100 \text{ g})$ FW) demonstrated the lowest contents of total phenolics. In the literature, the total phenolic content of beetroot is 257.2 mg/100 g FW, turnip 127.0 mg/100 g FW, and yam is 92.0 mg/100 g FW [44, 45]. High total phenolic contents of plants are related to antimicrobial and anti-oxidant activities of species, as well [46, 47]. Moreover, some phenolics activate anti-oxidant molecules in the cell [48]. Furthermore, phenolics are a part of chemicals that fight against diabetesrelated diseases [49]. Thus, the activities of phenolics can be influenced by many factors such as plant species, varieties, climate, storage conditions, and biotic and abiotic stress

On the other hand, carotenoids and chlorophylls are a part of tetraterpenoids and support healthy body functions against aging, cataract, and cancer [51, 52]. In the present study, total tetraterpenoids of 12 carrot varieties ranged from 0.2 to 4.1 mg/100 g FW. The highest tetraterpenoid contents were observed in NYC (4.1 \pm 0.2 a mg/100 g FW) and NPC (3.1 \pm 0.1 b mg/100 g FW); but the lowest tetraterpenoid contents were determined in MWC (0.2 \pm 0.0 l mg/100 g FW) and MiWC (0.4 \pm 0.0 k mg/100 g FW). Normal-sized and purple carrots were rich with total tetraterpenoid contents. In the literature, sweet potato and potato had 2.4 mg/100 g and 0.0 mg/100 g of total tetraterpenoid

contents, respectively [53]. Therefore, tetraterpenoid contents can be affected for varied reasons such as plant types, harvesting conditions, climate, and soil factors of plants [54, 55]. Moreover, tetraterpenoid contents are closely related to the colours of plant parts or vice versa.

To summarise, when L-ascorbic acid, total polyphenolic, and total tetraterpenoid contents were evaluated together, any correlation was not found. However, MYC had the highest L-ascorbic acid content with the third place of the highest total polyphenols (after the NPC and MiPC). Besides, NPC had the highest total polyphenolics and the second-highest value of tetraterpenoids, as well.

Sugar and organic acid profile of different carrot varieties

Sugars and organic acids are crucial for the quality and sensory attractiveness of raw materials [56]. These natural compounds comprise approximately 60% of dry matter, soluble solid content, and flavour of fruits and vegetables [57]. In the present study, sugars and organic acids were found to provide the taste and sweetness to the carrot varieties. As shown in Table 1, significant differences ($p \le 0.05$) in sugar and organic acid contents were observed in carrot roots. Fructose, sorbitol, glucose, and sucrose were determined as sugar components in the studied carrot varieties. Fructose and glucose were observed in all carrot samples; however, sorbitol was detected only in NOC, MiOC, MiYC, MiPC, MOC, MYC, and MPC. Moreover, sucrose was not identified in MiPC. The highest fructose levels were observed in NOC (3.8 g/100 g FW), MWC (1.9 g/100 g FW), and MiYC (1.9 g/100 g FW); the highest sorbitol levels were found in MiYC (0.5 g/100 g FW) and MYC (0.4 g/100 g FW); the



Table 1 Sugar and organic acid contents of carrot varieties

Compounds	NOC	NYC	NPC	NWC	MiOC	MiYC	MiPC	MiWC	MOC	MYC	MPC	MWC
Fructose	3.78*a	0.25 ^g	0.64 ^f	1.42 ^c	1.21 ^d	1.85 ^b	1.40 ^c	0.80 ^e	1.20 ^d	0.65 ^f	0.79 ^e	1.86 ^b
Sorbitol	0.01^{f}	0.00^{f}	0.00^{f}	0.00^{f}	0.23^{c}	0.53^{a}	0.23^{c}	0.00^{f}	0.10^{e}	0.39^{b}	0.18^{d}	0.00^{f}
Glucose	1.31 ^h	0.79^{m}	0.91^{1}	4.09^{a}	1.65^{f}	2.81 ^c	3.83^{b}	1.10^{j}	1.80 ^e	1.91 ^d	0.95^{k}	1.45 ^g
Sucrose	0.53^{g}	4.56 ^c	9.68^{a}	1.57 ^d	0.40^{h}	0.31^{j}	0.00^{k}	7.45^{b}	0.56^{f}	0.84^{e}	0.82^{e}	0.81^{e}
Total sugars	5.63 ^d	5.60^{d}	11.24 ^a	7.09^{c}	3.49^{k}	5.50 ^e	5.46^{f}	9.35 ^b	3.66^{j}	3.79^{h}	2.73^{1}	4.11 ^g
Oxalic acid	0.26^{f}	0.20^{k}	0.17^{l}	0.23^{h}	0.22^{j}	0.52^{b}	0.26^{g}	0.22^{j}	0.44 ^c	0.58^{a}	0.32^{e}	0.43^{d}
Maleic acid	0.00^{a}	0.00^{a}	0.00^{a}	0.00^{a}	0.00^{a}	0.00^{a}	0.00^{a}	0.00^{a}	0.00^{a}	0.01^{a}	0.01^{a}	0.00^{a}
Citric acid	0.10^{d}	0.04^{f}	0.06^{e}	0.00^{g}	0.00^{g}	0.00^{g}	0.00^{g}	0.00^{g}	0.00^{g}	0.14^{a}	0.11^{c}	0.12^{b}
Isocitric acid	0.63 ^h	0.70^{e}	0.68^{g}	0.48^{j}	0.30^{k}	0.49^{j}	0.28^{l}	1.78^{a}	0.70^{f}	1.14 ^b	0.89^{c}	0.78^{d}
Malic acid	0.34^{k}	0.60^{g}	0.86^{a}	0.67^{d}	0.52^{h}	0.69^{c}	0.66^{e}	0.22^{m}	0.64^{f}	0.50^{j}	0.70^{b}	0.26^{l}
Lactic acid	0.31^{b}	0.18^{f}	0.21^{e}	0.00^{g}	0.00^{g}	0.00^{g}	0.00^{g}	0.00^{g}	0.30^{d}	0.00^{g}	0.40^{a}	0.31 ^c
Fumaric acid	0.00^{h}	0.06^{c}	0.02^{f}	0.02^{ef}	0.03^{d}	0.02^{e}	0.02^{ef}	0.01^{g}	0.08^{a}	0.09^{a}	0.08^{b}	0.00^{h}
Adipic acid	0.05^{g}	0.14^{d}	0.18 ^c	0.05^{g}	0.00^{k}	0.04^{h}	0.01^{j}	0.00^{k}	0.10^{e}	0.33^{a}	0.26^{b}	0.06^{f}
Total organic acids	1.70^{f}	1.93 ^d	2.18 ^c	1.45 ^g	1.07^{j}	1.76 ^e	1.23 ^h	2.24^{b}	2.25^{b}	2.79^{a}	2.78^{a}	1.95 ^d

The sugar and organic acid contents (g/100 g FW) in 12 carrot varieties Significant at $p \le 0.05$

highest glucose levels were detected in NWC (4.1 g/100 g FW) and MiPC (3.8 g/100 g FW), and the highest sucrose contents were found in NPC (9.7 g/100 g FW) and MiWC (7.5 g/100 g FW). Besides, sucrose is the principal storage sugar and the level increases with maturity [58]. According to Alasalvar et al. [26], sucrose is the main sugar of purple carrot and is followed by lower contents of fructose and glucose. Similar results were also observed in the present study. Mini- and normal-sized carrot samples had the highest sugar contents, while white carrot samples of each size were rich in sugars. Sugar accumulation during the maturation of carrot varieties is related to the flavours of carrots [56, 59], and total sugar contents of carrot, turnip, and radish roots increase with high CO₂. Thus, this feature is important in terms of climate change.

Organic acids are significant compounds of plants for biological pathways as well as human well-being, because organic acids support moderate antibacterial activities [60]. Eight organic acids, namely oxalic, maleic, citric, isocitric, malic, lactic, fumaric, and adipic acids, were observed in the coloured carrot varieties. Among these organic acids, only oxalic, isocitric, malic, and fumaric acids were observed in all carrot varieties. Additionally, Bryant and Overell [61] identified malic, fumaric, and isocitric acids in carrot roots, as well. The present study showed that micro-sized carrots and purple samples were rich in organic acid contents. Thus, flavour compounds, sugars, organic acids, and pH are the factors that determine the specific aromas of carrot varieties. Moreover, organic acid composition changes with climate, growth conditions, and varieties [62].

Nutritional and chemical components of carrot varieties

Table 2 presents the basic chemical compositions of the 12 carrot varieties. The carrot root samples significantly differed in their chemical compositions ($p \le 0.05$).

Mean dry matter contents of the carrots ranged from 10.9 to 16.4%. Dry matter contents were the highest in NPC (16.4%) and NOC (15.1%), followed by MiWC (14.8%), MWC (14.7%), and MPC (14.4%). Gajewski et al. [63] studied different carrot varieties as well and confirmed the highest content of dry matter in purple carrots. Nevertheless, MiWC and MWC showed higher dry matter content than NWC. Thus, normal-sized white carrot holds more water than micro- and mini-white carrot samples.

In the present study, titratable acidity (TA) was determined as well. TA values ranged from 0.2 to 0.40 g/100 g FW. The highest TA was observed in MYC (0.4 g/100 g FW) and MPC (0.4 g/100 g FW), while the lowest results were observed for MiWC (0.2 g/100 g FW) and MiOC (0.2 g/100 g FW) samples. In terms of size and colour features of carrots, the micro-sized and purple-coloured carrot roots showed higher TA values. Moreover, TA influences flavour features of food products, and this parameter determines the pH and durability of carrot products. Therefore, high TA values in micro carrots make them much attractive for sensory characteristics and the food industry.

TA affects the pH of raw materials. The coloured carrot samples showed high pH values (> 5.3). Similar results were also noted in the literature [63]. In the present study, the pH



^{*}Values (mean of three replications) followed by the same letter within the same column were not significantly different ($p \le 0.05$) according to Duncan's test

Table 2 Chemical results of carrot varieties

Type of carrot	Dry matter	Ash	Pectin	TA	pН
NOC	15.06 ± 0.34 ^{‡b}	$0.78 \pm 0.00^{\text{hi}}$	1585.00 ± 0.18°	$0.27 \pm 0.01^{\rm f}$	$5.79 \pm 0.04^{\text{bcd}}$
NYC	11.81 ± 0.07^{de}	1.17 ± 0.04^{b}	1080.00 ± 0.01^{e}	$0.25\pm0.01^{\rm g}$	$5.54 \pm 0.01^{\rm f}$
NPC	16.39 ± 0.09^{a}	1.42 ± 0.03^{a}	935.00 ± 0.0^{ef}	$0.35 \pm 0.01^{\circ}$	5.35 ± 0.02^{h}
NWC	$10.92 \pm 0.14^{\rm f}$	1.07 ± 0.01^{c}	995.00 ± 0.06^{ef}	$0.24\pm0.0^{\rm gh}$	$5.42\pm0.0^{\rm gh}$
MiOC	11.14 ± 0.72^{ef}	0.74 ± 0.06^{i}	0.00 ± 0.00^{g}	$0.23\pm0.0^{\rm hi}$	$5.82\pm0.0^{\rm bc}$
MiYC	12.27 ± 0.25^{d}	0.82 ± 0.06^{gh}	$830.00 \pm 0.00^{\mathrm{f}}$	$0.27\pm0.00^{\rm f}$	$5.48\pm0.0^{\rm fg}$
MiPC	12.39 ± 0.24^{d}	0.99 ± 0.05^{d}	$140.00 \pm 0.00^{\rm g}$	$0.25\pm0.00^{\rm g}$	5.69 ± 0.00^{e}
MiWC	14.82 ± 0.14^{b}	1.12 ± 0.02^{bc}	1310.00 ± 0.10^{d}	$0.22\pm0.01^{\rm i}$	6.04 ± 0.08^{a}
MOC	$13.33 \pm 0.46^{\circ}$	0.87 ± 0.01^{fg}	1900.00 ± 0.00^{b}	$0.31\pm0.00^{\rm d}$	5.83 ± 0.00^{b}
MYC	$13.27 \pm 0.40^{\circ}$	0.90 ± 0.00^{ef}	2190.00 ± 0.00^{a}	0.40 ± 0.00^{a}	5.99 ± 0.00^{a}
MPC	14.42 ± 0.59^{b}	0.95 ± 0.00^{de}	1430.00 ± 0.00^{cd}	0.38 ± 0.00^{b}	$5.75 \pm 0.0^{\rm cde}$
MWC	14.68 ± 0.04^{b}	0.80 ± 0.00^{ghi}	$1380.00 \pm 0.23^{\rm d}$	$0.29 \pm .0.00^{\rm e}$	5.74 ± 0.05^{de}

Dry matter [%]; ash [%]; pectin (mg/100 g FW)

TA titratable acidity (g malic acid/100 g FW)

values of the carrots ranged from 5.4 to 6.0. The highest pH values were observed in MiWC (6.0), MYC (6.0), MOC (5.8), and MiOC (5.8). Moreover, the orange samples of different sizes showed much higher pH values (MOC: 5.8, MiOC: 5.8 and NOC: 5.8). According to Podsedek [64], the differences between samples are due to differences in varieties, soil pH, climate, growth, and storage conditions.

Pectins are subgroups of carbohydrates, provide functionalities to plant cell walls, and protect them against pathogens [65, 66]. However, enzymes in the human body cannot digest pectins, and gut bacteria help to reduce pectins which are important to prevent colon-related diseases [67]. In the present study, pectin levels ranged from 0 to 2190 mg/100 g FM in carrot samples. The highest content of pectin was seen in MYC (2190 mg/100 g FM) and MOC (1900 mg/100 g FM). In turn, MiOC showed no presence of pectin, and MiPC had a low amount of pectin content (140 mg/100 g FM). In the study, micro- and normal-carrot samples showed a high amount of pectins (from 1380 to 2190 mg/100 g FM and from 935 to 1585 mg/100 g FM, respectively); however, pectin contents were the lowest levels (from 0 to 1310 mg/100 g FM) in mini-carrot samples. According to Müller-Maatsch et al. [68], total pectin is divided into chelating agent soluble solids (CASS) and dilute alkaline soluble solids (DASS), and parsley has 47 mg/g of CASS. Besides, pectins of carrot, cabbage, and onion are sodium-carbonate soluble [69], and the solubility of pectin can alter with postharvest conditions of fruit and vegetables [70]. However, maturation is the main period of plants rising their water-soluble pectin content [71], because pectin fractions may eliminate reactive oxygen species (ROS) which are (non) free radicals of oxygen [72].

The other parameters analyzed in the study were minerals. They are essential for healthy body functions, and plants are good sources of minerals. Table 3 shows the contents of minerals in different varieties of carrots with significant differences. In the present study, it was observed that NYC and NOC were rich in Na; NPC and MYC showed the highest content of K; Ca was abundant in NWC and NYC; Fe was rich in MYC and MPC; and finally, the highest content of Mg was observed in MiYC and MYC. Normal-sized carrot samples showed higher mineral contents than micro- and mini-sized carrots; moreover, yellow carrots exhibited better results for mineral contents than carrots of other colours for each size. According to Nicolle et al. [41], K is the most abundant element in carrots, and a similar result was also observed in the present study. Additionally, obvious differences were noted when mineral values were compared with the ash contents of the carrots. NPC and NYC had the highest ash contents, while the lowest results were observed for MiOC and NOC. Thus, normal-sized carrots showed the highest results for ash content, and purple carrots exhibited the highest results for ash content in each size.

Antioxidant activity in carrot varieties

Table 4 presents the anti-oxidant activities of the 12 carrot varieties. Significant differences in anti-oxidant activities $(p \le 0.05)$ were observed in the carrot varieties.

Antioxidants inhibit reactive oxygen species that trigger the development of cancers, cardiovascular disorders, aging-related disorders, and other diseases [72]. Therefore, the anti-oxidant properties of plants make them a common food material for use in the food industry. Carrot is rich in



 $^{^{\}dagger}$ Significant at $p \le 0.05$

 $^{^{\}ddagger}$ Values (mean of three replications) followed by the same letter within the same column were not significantly different ($p \le 0.05$) according to Duncan's test

Table 3 Minerals of carrot varieties

Type of carrot	Macroelements Na	Macroelements K	Macroelements Ca	Macroelements Mg	Microelements Fe
NOC	378.0*b	1773 ^j	23.3 ^d	24.4 ^h	1.5 ¹
NYC	494.0 ^a	1954 ^c	32.7 ^b	11.7 ^k	1.9 ^k
NPC	95.9 ^k	2400 ^a	27.3°	3.8 ^m	2.2^{j}
NWC	372.0°	1718 ¹	37.6 ^a	61.4 ^e	2.3 ^h
MiOC	102.0 ^j	1479 ^m	22.2 ^e	37.9 ^g	$2.7^{\rm f}$
MiYC	355.0 ^d	1805 ^g	17.6 ^l	79.9 ^a	3.1 ^e
MiPC	274.0^{f}	1745 ^k	20.3^{j}	$40.5^{\rm f}$	5.1 ^d
MiWC	301.0 ^e	1922 ^d	18.6 ^k	72.1 ^c	2.5^{g}
MOC	136.0 ^h	1782 ^h	20.3 ^h	3.9^{l}	5.3°
MYC	86.1 ¹	2007 ^b	22.0^{f}	74.0 ^b	13.4 ^a
MPC	55.8 ^m	1820 ^f	20.3 ^h	14.2 ^j	12.9 ^b
MWC	169.0 ^g	1869 ^e	20.7 ^g	69.5 ^d	5.4 ^c

Minerals (mg/100 g dm)

Significant at $p \le 0.05$

Table 4 The anti-oxidant activities [mmol TE/100 g dm] of carrot varieties

Type of carrot	ABTS	FRAP	ORAC
NOC	$0.82 \pm 0.03 * fg$	0.71 ± 0.21^{e}	3.13 ± 0.19^{j}
NYC	0.48 ± 0.01^{g}	0.28 ± 0.01^{e}	2.52 ± 0.03^{1}
NPC	7.38 ± 0.39^{b}	4.60 ± 0.2^{b}	16.17 ± 1.03^{b}
NWC	0.46 ± 0.02^{g}	0.27 ± 0.03^{e}	2.84 ± 0.12^{k}
MiOC	2.42 ± 0.08^{d}	2.66 ± 0.76^{c}	4.28 ± 0.30^{g}
MiYC	0.96 ± 0.01^{f}	0.64 ± 0.02^{e}	3.86 ± 0.30^{h}
MiPC	7.93 ± 0.51^{a}	5.77 ± 0.17^{a}	17.06 ± 0.84^{a}
MiWC	$0.81 \pm 0.04^{\mathrm{fg}}$	0.58 ± 0.04^{e}	5.09 ± 0.20^{e}
MOC	$1.02 \pm 0.04^{\rm f}$	0.73 ± 0.02^{e}	5.93 ± 1.22^{d}
MYC	2.00 ± 0.14^{e}	1.62 ± 0.07^{d}	$4.92 \pm 0.11^{\rm f}$
MPC	3.98 ± 0.17^{c}	$2.21 \pm 0.63^{\circ}$	15.42 ± 1.14^{c}
MWC	$1.06 \pm 0.03^{\rm f}$	$0.82 \pm 0.06^{\rm e}$	3.42 ± 0.13^{i}

TE Trolox equivalents

Significant at $p \le 0.05$

phytochemicals such as phenolic compounds, carotenoids, and ascorbic acid that are essential as nutritional antioxidants in the human diet [73]. In the present study, in vitro anti-oxidant activity assays of all the carrot samples were quantified by ABTS, FRAP, and ORAC methods. Remarkable differences were observed in carrot varieties depending on colour, size, and/or the applied anti-oxidant activity techniques.

In the ABTS method, activities of carrots ranged from 0.5 to 7.9 mmol TE/100 g dm. The highest results were observed

for MiPC (7.9 mmol TE/100 g dm) >> NPC (7.4 mmol TE/100 g dm) >> MPC (4.0 mmol TE/100 g dm). The lowest results were observed in NWC (0.5 mmol TE/100 g dm) and NYC (0.5 mmol TE/100 g dm). According to Singh et al. [74], the ABTS test showed the highest anti-oxidant activity in purple carrot. A similar result was observed in the present study, as well. Therefore, anti-oxidant activity showed a positive correlation with anthocyanins. Sun et al. [75] reported the same relationship.

In the FRAP test, anti-oxidant activities ranged from 0.3 to 5.8 mmol TE/100 g dm. The MiPC (5.8 mmol TE/100 g dm) showed the highest results, followed by NPC (4.6 mmol TE/100 g dm) and MiOC (2.7 mmol TE/100 g dm). The lowest result was observed for NWC (0.3 mmol TE/100 g dm) and NYC (0.3 mmol TE/100 g dm). Hence, mini-sized and purple carrots exhibit the highest anti-oxidant activities by FRAP. Singh et al. [74] also showed the highest results for purple carrot in the FRAP assay.

The ORAC assay also showed similar results. The highest results were again observed for MiPC (17.1 mmol TE/100 g dm), followed by NPC (16.2 mmol TE/100 g dm) and MPC (15.4 mmol TE/100 g dm). Thus, purple carrots showed the highest anti-oxidant activity results in the ORAC assay for each size. The lowest results were observed for NYC (2.5 mmol TE/100 g dm) and NWC (2.8 mmol TE/100 g dm). According to Nicolle et al. [41], the dark-coloured carrot demonstrated the highest anti-oxidant activity, and the white carrot showed the lowest activity in the ORAC assay. Therefore, dark-coloured carrots are rich in anthocyanins and thus exhibit strong anti-oxidant activities.

In the present study, the total carotenoid content did not correlate with the total anti-oxidant activity. Mech-Nowak



^{*}Values (mean of three replications) followed by the same letter within the same column were not significantly different (p > 0.05) according to Duncan's test

^{*}Values (mean of three replications) followed by the same letter within the same column were not significantly different (p>0.05) according to Duncan's test

et al. [76] also reported similar results. According to Smeriglio et al. [77], the anti-oxidant activity of purple carrot results from anthocyanins and phenolic acids. Similar results were observed in the present study. Algarra et al. [78] compared the anti-oxidant activities of purple carrot and orange carrot growing in the same region and showed that purple carrot exhibited higher anti-oxidant activities than orange carrot depending on the content of anthocyanins.

Conclusions

The study evaluates the carrot varieties with different size and colour features for vitamin C as well as chemical contents (dry matter, ash, pectin, titratable acidity, and pH), sugar, organic acids, minerals (sodium, potassium, calcium, iron, and magnesium), and anti-oxidant activities. Besides, total polyphenolics and total tetraterpenoids of colourful carrot varieties were presented. According to study results, carrot samples were rich in fructose, sorbitol, glucose, and sucrose as sugars; citric, isocitric, maleic, malic, lactic, fumaric, oxalic and adipic acids were the organic acid compounds in coloured carrot samples. Moreover, sucrose was the dominant sugar and isocitric acid was the most common organic acid in carrot samples. Purple-coloured carrot samples demonstrated the highest results for total sugar, total organic acid, total polyphenolic contents, and anti-oxidant activities (Mini > Normal > Micro). In turn, the lowest results were observed in normal yellow carrot for total polyphenols, and anti-oxidant activities; besides, the lowest total tetraterpenoids were determined in micro-white carrot. However, normal-white carrot exhibited low anti-oxidant results for all three tests (ABTS, FRAP, and ORAC) and the lowest L-ascorbic acid content as well. The conducted study had shown that purple carrots are particularly interesting roots, they are sweet and colorful (they will arouse sensory interest), and additionally, they are rich in bioactive compounds (pectins, vitamin C, and polyphenolic compounds). Therefore, it is worth considering their use in the food industry, especially in the design of children's products.

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Declarations

Conflict of interest The authors declare no conflict of interest.



Compliance with ethics requirements This article does not contain any studies with human or animal subjects.

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