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ASSESSMENT OF ROMANIAN PEPPERS CULTIVARS FOR FRUIT QUALITY TRAITS

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Abstract

In recent years, there has been a growing trend among people to be more conscious of the food they consume, seeking products with high nutritional value, bioactive compounds, and antioxidant capacity to promote good health and maintain an excellent body figure. Among the most popular and versatile fresh vegetables worldwide are peppers (Capsicum annuum L.), known for their vibrant colours, delightful flavours, and abundant nutritional benefits. In this study, we aimed to assess the phenotypic diversity in terms of fruit quality traits and yield for five Romanian pepper cultivars bred at VRDS Buzau: two bell peppers (Cantemir, Regal), two hot peppers (Roial, Decebal), and one accession (L-50). To evaluate fruit quality, we measured various parameters including dry matter content, fruit firmness, total soluble solids, titratable acidity, ascorbic acid, lycopene, and β-carotene content at different ripening stages. The results showed that fully ripe fruits exhibited significantly higher levels of bioactive compounds compared to unripe fruits. Specifically, the L-50 variety demonstrated the highest content of total soluble solids and β-carotene, making it a standout choice for its nutritional benefits. On the other hand, the Roial variety stood out with its rich vitamin C content. Moreover, the Cantemir cultivar displayed the highest fruit firmness (10.1 N), indicating an extended shelf-life capacity, which could be advantageous for storage and transportation. Importantly, all studied genotypes exhibited promising antioxidant properties, positioning them as potential natural ingredients for functional foods. In conclusion, our findings underscore the remarkable diversity and nutritional potential of these Romanian pepper cultivars. Their abundance in bioactive compounds and antioxidant capacity makes them excellent choices for health-conscious consumers seeking fresh and nutritious ingredients to support their well-being and dietary goals.

Keywords: *antioxidants, Capsicum annuum L., fruit quality, Romanian peppers*

1. INTRODUCTION

Capsicum peppers, members of the Solanaceae family, comprise a diverse genus with 44 described species (Barboza et al., 2019; Barboza et al., 2020; Muñoz-Ramírez et al., 2020). Among these, five are domesticated: *C. annuum* L., *C. frutescens* L., *C. chinense* Jacq., *C. baccatum* L. (var. *pendulum*), and *C. pubescens* Ruiz & Pav. (Hernández-Pérez et al., 2020). Notably, *C. annuum* L. stands out as the most important species cultivated and consumed in Romania due to its unique flavor and pungency.

Peppers are among the most significant vegetable crops worldwide, valued for their versatility and innovative culinary uses. They can be enjoyed raw, cooked, pickled, used as pasta ingredients, spices, or food colorants. Abundant in phytochemicals such as ascorbic acid, carotenoids, capsaicinoids, vitamins, minerals, and phenolic compounds (Bogusz et al., 2018; Ribes-Moya et al., 2018; Gamela et al., 2020), peppers are renowned for their high content of bioactive compounds and potent antioxidant capacity (Blanco-Ríos et al., 2013; Chávez-Mendoza et al., 2015). Regular pepper consumption provides various health benefits, including antioxidant properties that protect against oxidative damage, reducing the risk of common degenerative diseases like cardiovascular issues, diabetes, cancer, cataracts, and neurodegenerative conditions (Arimbor et al., 2015). These chemical compounds also promote optimal brain function by preventing oxidation of essential fats within brain cells (Obocha and Rocha, 2007).

According to the latest data from FAO (Food and Agriculture Organization), global Capsicum production in 2021 reached approximately 3.6 million tons of fresh fruit and 4.8 million tons of dry fruit (FAO, 2023). In Romania, pepper crops are primarily cultivated by small farmers seeking sustainable livelihoods, ranking as the fourth most important vegetable crop after cabbage, tomatoes, and onions.

The harvested area of green chilies and peppers in Romania, as per FAO 2022, comprises 10,370 hectares with a production of 154,790 tons (FAO, 2023).

The Vegetable Research Development Station (VRDS) Buzau plays a significant role in germplasm conservation and vegetable breeding in Romania. Currently, VRDS Buzau has registered 11 cultivars of peppers in the Official Catalogue of Species and Varieties of Cultivated Crops from Romania, including 4 cultivars of bell pepper (‘Buzau 10’, ‘Galben Superior’, Arum, and ‘Cantemir’), 2 cultivars of long pepper (‘Cosmin’ and ‘Regal’), 1 cultivar of pimiento pepper (‘Splendens’), and 5 cultivars of hot peppers (‘Decebal’, ‘Jovial’, ‘Roial’, ‘Vladimir’, and ‘Mitu’).

In recent years, plant breeders have prioritized research on genotypes with elevated antioxidant content, driven by the direct link between diet and health (Brezeanu et al., 2022). This study aims to evaluate phenotypic diversity concerning fruit quality traits and yield for five Romanian pepper cultivars, with the goal of understanding their potential applications and increasing consumer awareness of their health benefits.

2. MATERIALS AND METHODS

2.1. Materials

To achieve the goal of this research, we studied five pepper varieties bred at VRDS Buzau: Cantemir, Regal, Roial, Decebal, and accession L50 (Figure 1). Table 1 presents some morphological characteristics of the studied genotypes.

Fruits harvested at the green (HM) stage were noted as follows: Cantemir I, Regal I, Roial I, Decebal I, L50 I, while fully ripe fruits at physiological maturity (PM) were noted as: Cantemir M, Regal M, Roial M, Decebal M, L50 M.

No	Cultivar	Variety	Time of maturity	Fruit shape	Fruit colour
1	Cantemir	Sweet	very early	Blochy	Red
2	Regal	Sweet	early	Conical	Red
3	Roial	Hot	early	Elongated	Red
4	Decebal	Hot	early	Hornsahped	Orange
5	L50	Sweet	late	Pimiento	Red

Table 1. Morphological characteristics of the studied genotypes

2.2. Methods

2.2.1. Experimental Site

The experiment was conducted in the year 2022 at the experimental plot of the Vegetable Research and Development Station Buzău, located in Buzău County, Romania (latitude: 5.13711°N, longitude: 26.81711°E, and elevation: 95 m above sea level).

Seeds from each genotype were sown in alveolar pallets with 70 cubes, each having a volume of 50 cm3, using peat as the substrate. To prevent the spread and build-up of seed-borne viruses, we applied a treatment to the seeds following the Protocol for Regeneration and Characterization of Capsicum, as described by The World Vegetable Center (AVRDC, 2018). Germination and seedling development were monitored daily, and irrigation was performed once a day. The seedlings were transplanted to the polytunnels at the age of 55 days, and the plants were arranged in a randomized complete block design with inter-row and intra-row spacing of 70 cm and 30-35 cm, respectively. The crop technology applied for pepper cultivation followed the recommendations from the specialized literature (Barcanu et al., 2021).

2.2.2 Fruit Material

Harvesting of pepper fruits to determine fruit quality traits was performed at two different ripening stages: harvest maturity and physiological maturity. From each genotype, a total of 15 fruits were randomly chosen and analysed following the protocol described by Silva et al. (2011).

For the analyses of dry matter, total soluble contents, acidity, lycopene, β-carotene, and vitamin C content, randomly mixed samples were prepared for each genotype, and each analysis was carried out in triplicate, as described by Brezeanu et al. (2022). All determinations were conducted on the day of harvest. The studied genotypes are represented in Figure 1.

Fig. 1. Pepper fruit cultivars: a) Regal, b) Cantemir, c) Decebal, d) Roial, and e) L 50

2.2.3 The Analytical Methodology Used

2.2.3.1. Determination of Dry Matter Content (TDM, %)

The Dry Matter Content of the fresh peppers was determined according to Lannes et al., 2007 and do Rêgo et al., 2011, methods adapted according to AOAC 930.04, Loss on Drying (Moisture) in Plants. (2005).

2.2.3.2. Determination of Total Soluble Solids (TSS, °Brix,)

The homogenized juice obtained by pressing the fresh fruit was used to estimate the total soluble solids (TSS) via portable digital refractometer (KERN OPTICS ORF 1RS) and the results were express in °Brix, according to AOAC 932.12 methods.

2.2.3.3 Titratable Acidity Analysis

Total titratable acidity was determined using 5 g of ground pepper diluted in 25 mL of double distilled water. The titratable acidity of peppers was determined by titration with 0.1 N NaOH until pH reach 8.1 (Bozokalfa and Esiyok., 2011). Titration was made with TitroLine easy (Saad et al., 2014; Official method AOAC 942.15). The results were expressed in g acid citric per 100 g of pulp.

2.2.3.4 Determination of β–Carotene and Lycopene

The quantification of lycopene and β-carotene was conducted according to the method developed by Nagata and Yamashita (1992) with slight modifications. One gram of tomato sample was homogenized with 10 ml of hexane: acetone (4:6 v/v). The absorbance readings were performed at the following wavelengths: 663 nm, 645 nm, 505 nm and 453 nm, and pigments determination were calculated using the referred equations:

β-carotene mg $(mL)^{-1} = 0.216A_{663} - 1.220A_{645} - 0.304A_{505} + 0.452A_{453}$

Lycopene mg $(mL)^{-1} = -0.0458\ A_{663} + 0.204A_{645} + 0.372\ A_{505} – 0.0806\ A_{453}$

The results were expressed mg $\cdot 100mL^{-1}$.

2.2.3.5 Determination of Acid L-ascorbic

Vitamin C was determined using a relatively simple titration method using iodine 0,01 M as described by Sheeba and Shalini 2021 with slight modification. Pepper juice were prepared like this, 10 g of each sample was cut into small pieces, blend together with 50 ml of distilled water using an electronic blender and then flirted. The filtered was transferred to a 100 ml Standard Measuring flask and the flask was filled up to the mark with distilled water. 20ml flirted was transferred in a 200 ml Erlenmeyer flask and mixed together with 150 ml distilled water. 20 ml of solution of the juice is titrated against iodine solution using an automatic TitroLine easy. Results were expressed as $mg100g^{-1}$ on FW.

2.2.3.6 Fruit firmness

The fruit firmness was tested using the electronic penetrometer TR Turoni 53205 equipped with a 3 mm piston and the results were expressed in Newton (N).

2.2.3.7 Statistical Analysis

Statistical analyses were performed using analysis of variance (ANOVA). In the case of a significant F-value, the means were compared using Duncan test.

3. RESULTS

3.1. Physical properties and dry matter content

As shown in Table 2, the influence of the cultivar on the dry matter content of pepper fruits at harvest maturity varied from 7.76% for the Decebal variety to 8.29% for the Cantemir variety. During ripening, the dry matter content (DM) of peppers showed an upward trend and exhibited a significant increase at physiological maturity. These changes were consistent with the findings reported by Ionică et al. in 2017. Notably, the Roial variety exhibited the highest dry matter content at physiological maturity (14.08%), while the Regal variety registered the lowest content in dry matter, with 7.88%.

As expected, hot peppers generally have lower weight than sweet peppers. In the current research, the average weight of pepper fruits varied widely, ranging from 27.4 g at harvest maturity (HM) and 33.11 g at physiological maturity (PM) for the Decebal variety, to 136.45 g at HM and 168.6 g at PM for the L50 accession. According to Carusso et al. in 2018, these differences between varieties are often genetically influenced.

The quantitative expression of the structural characteristics of the cell wall in harvested products constitutes one of the attributes of quality most appreciated by the consumer, making this parameter one of the main selection criteria in plant breeding programs (Perez-Grajales et al., 2019). Fruit firmness of the studied genotypes changed during ripening, with the highest firmness noted at physiological maturity for the Cantemir variety (10N). According to Barcanu et al. in 2021, pepper fruits with high firmness are suitable for fresh consumption and have a longer shelf life.

Genotype	Firmness (N)	Dry matter (%)	Yield/plant (g)
Cantemir I	9.25±0.92^{cd}	6.64±0.18^{a}	102.48± 3.57^{b}
Cantemir M	**10.00±1.24^{d}**	7.80±0.07^{a}	119.38± 9.73^{c}
Regal I	6.13± 0.19^{ab}	5.49±0.11^{a}	90.57± 1.72^{b}
Regal M	8.88± 0.52^{bcd}	7.88±0.13^{a}	134.57± 10.30^{c}
Roial I	**5.78±0.37^{a}**	9.23±0.30^{a}	16.07± 1.44^{a}
Roial M	6.87±0.29 ^{abc}	**14.72**±1.41^{b}	**21.99± 3.67^{a}**
Decebal I	7.52± 3.10^{abcd}	**5.25**±0.17^{a}	27.40± 3.14^{a}
Decebal M	7.88± 0.90^{abcd}	11.68±0.43^{a}	33.11±7.52 ^{a}
L-50 I	8.97±0.18^{bcd}	7.30±0,12^{a}	136.45± 3.75^{c}
L-50 M	8.05±0.15^{abcd}	9.62±0.17^{a}	**168.60 ±17.80^{d}**

Table 2. Physical properties and dry matter content of studied genotypes.

Note: Values are mean ± SD. Means within the same row carrying different superscript letter were significantly different at $p < 0.01$ according to a Duncan`s multiple range

3.2. Biochemical components

Table 3 presents the results of fruit quality parameters. Significant differences were observed in total soluble solids, lycopene, β-carotene, and vitamin C content.

Genotype	TSS (°Brix)	TA (%)	Lycopene (mg/ml)	β-carotene (mg/ml)	Vitamin C (mg/100g FW)
Cantemir I	**2.84±0.05^{a}**	**0.41±0.43^{a}**	0.002± 0.01^{d}	0.01±0.01^{a}	93.91± 10.77^{ab}
Cantemir M	3.84±0.07^{ab}	0.26±0.02^{a}	0.22±0.12^{ef}	0.47±0.01^{c}	127.04±3.31^{c}
Regal I	3.84±0.17^{ab}	0.19±0.01^{a}	0.002±0.01^{d}	0.02±0.01^{a}	87.54±1.77^{a}
Regal M	5.93±0.12^{cd}	0.30±0.03^{a}	0.27±0.01^{f}	0.62±0.01^{c}	95.56±3.18^{ab}
Roial I	6.87± 1.49^{d}	0.14±0.01^{a}	**-0.004±0.01^{c}**	0.01± 0.04^{a}	154.53±5.48^{d}
Roial M	**9.33±0.58^{e}**	0.28±0.02^{a}	**0.28±0.01^{f}**	0.50±0.35^{c}	**209.70±1.71^{f}**
Decebal I	3.49±0.37^{ab}	**0.09±0.01^{a}**	**-0.004±0.01^{c}**	**0.04±0.01^{a}**	94.09±7.72^{ab}
Decebal M	8.15±0.16^{e}	0.27±0.02^{a}	-0.04±0.01^{b}	0.45±0.02^{bc}	182.52±11.16^{e}
L-50 I	4.75±0.45^{bc}	0.12±0.01^{a}	-0.27±0.02^{a}	0.19±0.03^{ab}	**81.42±2.88^{a}**
L-50 M	6.00±0.20^{cd}	0.21± 0.02^{a}	0.09± 0.01^{e}	**0.98±0.06^{d}**	106.55±2.79^{b}

Table 3. Physical properties and dry matter content of studied genotypes.

Note: Values are mean ± SD. Means within the same row carrying different superscript letter were significantly different at $p < 0.01$ according to a Duncan`s multiple range

In this study, TSS values showed variations in both maturity stages. However, analyzing the average content of each parameter by statistical comparisons according to genotype, it was observed that the TSS content did not show statistical differences. The highest value of TSS in both maturity stages was recorded for the L50 variety with a value of 4.75°Brix at harvest maturity and 6°Brix at physiological maturity. Similar to the DM content during ripening, the TSS content increased for all studied cultivars, which is consistent with the results of Rahman et al. (2017). These changes in TSS contribute to the sweetness of the fruit due to increased accumulation of hexose sugar during fruit ripening (Nielsen et al., 1991).

The titratable acidity of the pepper samples significantly increased during fruit ripening ($p<0.05$). At harvest maturity, the highest value was registered for the Regal cultivar with 0.18 g citric acid·100g-1 FW, and the lowest value was registered for the Decebal cultivar with 0.04 g citric acid·100g-1 FW. However, at physiological maturity, the Decebal variety showed the highest increase in TA, being the genotype with the highest content in TA with a value of 0.27 g citric acid·100g-1 FW.

Values of lycopene and β-carotene content were statistically different between cultivars. A higher concentration of lycopene was found in the fruits of the Regal variety (0.21 $mg \cdot ml^{-1}$), while the L50 accession had higher average values of β-carotene (0.98 $mg \cdot ml^{-1}$). In our study, the Decebal cultivar had orange fruit, and the level of lycopene was below the limits of detection (-0.01 $mg \cdot ml^{-1}$). Isaacson et al. (2002), in studies on orange tomatoes, found that they have a recessive mutation (*tangerine mutant*) that prevents the completion of the catalysis of prolycopene into lycopene.

Regarding the vitamin C content of the five genotypes of peppers studied, it varied significantly depending on the genotype, with higher values at physiological maturity ranging from 209.70 mg/100g FW for the Roial variety to the lowest values between 95.56 mg/100g FW and 106.55 mg/100g FW for the Regal and L50 genotypes, respectively. Also, an increase in vitamin C content was observed during fruit ripening for all studied cultivars. Considerable variation in ascorbic acid content of peppers was reported by Dinu et al. (2013) and Soare et al. (2017). Variation in ascorbic acid content can be attributed to multiple factors such as cultivar response, sampling variation, and temperature fluctuations (Kouassi et al., 2012).

4. DISCUSSION

Peppers contain bioactive compounds that can be used for animal feed or as nutraceutical, cosmeceutical, and functional food ingredients (Razola-Diaz et al., 2022). Based on these bioactive compounds, epidemiological studies have consistently demonstrated a positive relation between the consumption of fruits and vegetables and a reduction in the mortality rate due to cancer, heart disease, degenerative diseases, as well as aging (Chávez-Mendoza et al., 2015).

One bioactive compound with multiple health benefits is Vitamin C. It is an essential dietary nutrient for a variety of biological functions, fundamental in the biosynthesis of collagen and serves as a co-factor in several important hydroxylation reactions in humans. Additionally, positive relationships have been shown between ascorbate supplement use and reduced incidence of Alzheimer's disease. Furthermore, Vitamin C plays a role as an antioxidant, providing a buffer against cell damage that occurs during routine cell functions (Grosso et al., 2013). Since humans cannot synthesize vitamin C due to a series of mutations of the gene encoding gulonolactone oxidase, the identification of dietary sources of vitamin C is very important.

All the pepper varieties analyzed in the present study are an excellent source of vitamin C and exceeded the recommended daily dosage (RDD) of 60 mg/100 g. The presented data showed that the concentration of this vitamin increases with fruit ripeness, as reported by Shaha et al. (2013). On average, Regal contributed with 158% of the RDD and Roial with 348%. These results are consistent with other studies reporting that bell peppers contribute from 25% to 218% of the RDD of vitamin C (Chávez-Mendoza et al., 2015).

The interest in carotenoids has grown exponentially due to studies suggesting their impact on health and human diseases. Peppers are an important source of carotenoids such as lycopene and β-carotene (Brezeanu et al., 2022; Igbokwe et al., 2013). Lycopene is not toxic and has anti-inflammatory, antioxidant, and chemotherapeutic effects in heart and brain diseases and on some types of cancer (Cruz et al., 2013), while β-carotene, the precursor of vitamin A, possesses pronounced free radical scavenging properties (Deepa et al., 2006).

The concentration of carotenoids is related to the ripening of fruit due to the increase in this carotenoid and the consequent decrease in chlorophyll when the fruit changes from green to red (Flores-Velazquez et al., 2022). For this reason, at harvest maturity, lycopene and β-carotene content of studied varieties was relatively low.

The high content of dry matter is an important quality descriptor in the food industry for obtaining chili powder (Ribes-Moya et al. 2018). The total dry matter values in previous research ranged between 4.83% - 17.78% (Brezeanu et al., 2022) and 11.9% to 16.5% (Stan et al., 2021). The values obtained in this study fall within the mentioned interval, ranging between 7.41% and 14.08%.

Shelf-life of fruits is characterized for its value as a quality and postharvest parameter, and high values of firmness are desirable for those products that must be transported over long distances before reaching a consumer's market (Falik et al., 2019; Flores-Velazquez et al., 2022). A much higher value of firmness of bell pepper, between 12.64N and 21.60N, was reported by Figueroa et al. (2015), while Palacio and Sánchez (2015) reported lower values (6.39N and 7.0N) of firmness for the same crops.

According to Martínez et al. (2007), the total soluble solids (TSS) comprise total sugars and organic acids, among other constituents, and these components can be used along with color change to determine the maturity of peppers and are often evaluated in analyses of vegetables as they are critical to the overall sensory quality. In the current research, the sugar values varied between 3.83 and 6.00 °Brix. In the literature, the values of TSS in peppers vary between 3.5 and 7°Brix (Soare et al., 2017), 5.20 and 8.85 (Flores-Velazquez et al., 2022).

The most reliable guide to ripening is titratable acidity (TA) or the ratio of TSS (°Brix) to TA. These values represent the amount of organic acids within the fruit (Martínez et al., 2007). In this study, similar to TSS, the titratable acidity showed an increase during ripening. A similar result was reported by Ghasemnezhad et al. (2011).

5. CONCLUSIONS

The present research has provided valuable insights into the diverse characteristics of the five pepper varieties studied, including physical properties, bioactive compounds, and yield potential.

Among the varieties, L-50 exhibited the highest content in total soluble solids and β-carotene, making it a promising choice for its nutritional benefits. On the other hand, Roial demonstrated the highest content in vitamin C, adding to its appeal as a source of essential nutrients. Additionally, Cantemir stood out with its exceptional fruit firmness, indicating a long shelf-life capacity, which could be advantageous for transportation and storage.

This study has contributed to a deeper understanding of the phytochemical potential of local Romanian pepper cultivars, shedding light on their health benefits. Based on our findings, we highly recommend these varieties for direct usage in marketplaces, as they offer a diverse range of flavours, nutrients, and unique characteristics that can cater to consumer preferences and contribute to a healthy diet.

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