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SPECTROPHOTOMETRIC CHARACTERIZATION AND CLASSIFICATION OF TRADITIONAL BULGARIAN RED PAPRIKA

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Abstract
This study focuses on the spectrophotometric characteristics and classification of samples of traditional Bulgarian red paprika. Five commercially available brands of paprika were analyzed. Ethanol was used to extract the coloring pigments. Spectral analysis was performed using a SpectroVis Plus spectrophotometer operating in the 380–950 nm range.
The results reveal characteristic absorption profiles with distinct maxima, reflecting variations in carotenoid pigment content, the maturity of the raw material. Significant variations in optical density and absorption intensity were observed among the different brands, enabling their grouping.

Keywords: paprika, spectrophotometry, carotenoids, chlorophyll, maturity.

INTRODUCTION

Red paprika (*Capsicum annuum* L.) is an annual agricultural crop cultivated worldwide and belonging to the family Solanaceae. Plants of this genus possess numerous beneficial properties that distinguish them from other widely cultivated vegetables [1]. Dried and ground red paprika is used not only as a culinary spice but also in traditional medicine and pharmacology [2]. It has demonstrated antioxidant and antibacterial properties and has been reported to have protective effects in relation to cancer, diabetes, gastrointestinal disorders, obesity, high cholesterol and blood glucose levels, anaemia, and other conditions. Paprika extracts from red paprika are used in the food, agricultural, and textile industries, as well as in cosmetics, where they are incorporated into anti-aging formulations [1–5].

The popularity of this spice is primarily attributable to its sensory characteristics, namely color, pungency and flavour. Color measurement is a fundamental aspect of the red paprika trade.

Spices such as red paprika are often subject to food fraud. Adulteration of this spice can pose a substantial risk to consumer safety. Commonly reported forms of paprika adulteration include the addition of waste materials or lower-quality products, as well as the use of unauthorised colorants [6–9].

The aim of the present study is to initiate an in-depth investigation into the quality of red paprika spice available on the retail market in the Republic of Bulgaria.

EXPOSITION

Sweet paprika (*Capsicum annuum* L.) is an excellent source of carotenoids and ascorbic acid. It is naturally low in saturated fat and sodium, and cholesterol-free [10-11].

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Carotenoids are plant pigments widely distributed in fruits and vegetables, which play important roles in human health. There are two main classes of carotenoids: carotenes and xanthophylls. Carotenes consist of carbon and hydrogen atoms, whereas xanthophylls contain one or more oxygen atoms. In higher plants, carotenoids are found in the chloroplasts of photosynthetic tissues, as well as in the chromoplasts of flowers and fruits. In leaves, they occur primarily in a free form, whereas in other tissues, they are present as esters. Their composition in fruits and vegetables varies depending on cultivar, stage of ripeness, agro-climatic conditions, and other factors. These plant sources are the primary providers of β-carotene [12-15].

The characteristic red color of the ripe fruits of *Capsicum annuum* L. is due to capsanthin and capsorubin, while the yellow-orange color is associated with β-carotene, zeaxanthin, lutein, β-cryptoxanthin, violaxanthin, and capsolutein [16, 17]. Red paprikas can contain 12 or more different capsaicinoids, the concentrations of which are influenced by post-harvest processes [18].

Carotenoids found in green, photosynthetically active plant tissues are classified as primary, while those in red fruits are classified as secondary. The total carotenoid content can be quantified alongside with the chlorophyll content, by measuring absorbance not only at the absorption maxima of chlorophyll a and b, but also at a wavelength where carotenoids exhibit significant absorption – for example, 470 nm [19–21].

Carotenoid analysis in plant samples can be performed using varius methods, with spectrophotometry remaining a practical, reliable, and cost-effective choise.

The experiments were conducted using five commercially available samples of sweet red paprika powder, purchased randomly from local retail markets. For the spectrophotometric analysis, 0.5 g of each dry paprika powder sample was mixed with 100 mL of ethanol (95%). Following centrifugation, 5 mL of the solution was transferred into standard spectrophotometric cuvettes for analysis.

The spectra were acquired using a SpectroVis Plus spectrophotometer operating over the wavelength range of 380-950 nm, with the photometric accuracy ±0.10 A.U. The resulting absorbance spectra are presented in Fig. 1.

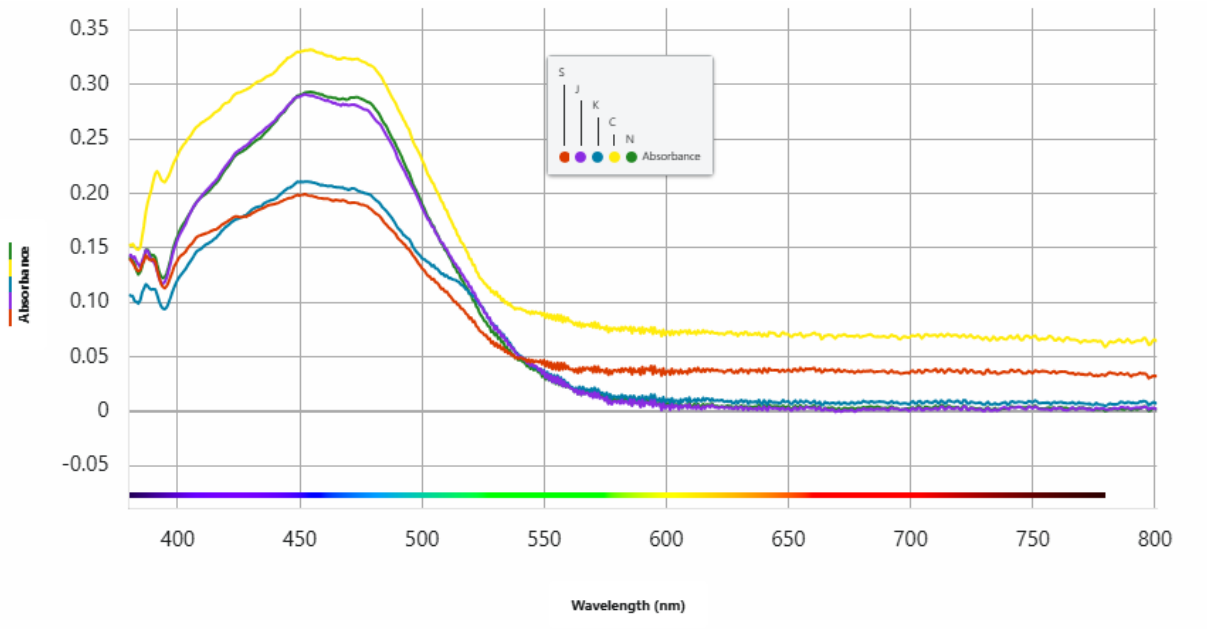


Fig. 1. *Absorbtion spectra of red paprika samples obtained from photometric measurements*

The descriptive statistics for the obtained absorption spectra are presented in Table 1.

Table 1. *Descriptive statistics of the absorbtion spectral data*

	S	J	K	C	N
Min	-0.038	-0.023	-0.027	0.041	-0.051
Max	0.199	0.291	0.211	0.332	0.293
Mean	0.064	0.062	0.052	0.121	0.059
Kurtosis	0.412	0.565	0.227	0.589	0.664
Skewness	1.365	1.434	1.299	1.461	1.478
SD	0.056	0.091	0.065	0.085	0.093
Median	0.037	0.009	0.014	0.074	0.008

Descriptive statistics of the obtained spectral data indicate that the data deviate significantly from a normal distribution. Skewness values ranging between 1.299 and 1.478 indicate a strong positive (right-sided) skew, while kurtosis values between 0.277 and 0.664 show a slightly leptokurtic distribution.

Second-derivative analysis of absorbance as a function of wavelength

Calculating the second derivative helps distinguish absorbance peaks. Second-derivetive analysis of absorbance as a function of wavelength was applied to better distinguish overlapping absorption peaks. For pure red paprika, a single principal minimum is observed in the second derivative at 460 nm. Figure 2 shows the calculated second-derivative spectra across the wavelength range function. In the 400 to 800 nm interval, the second derivative exhibits a principal minimum at 460 nm, with overall fluctuations around zero.

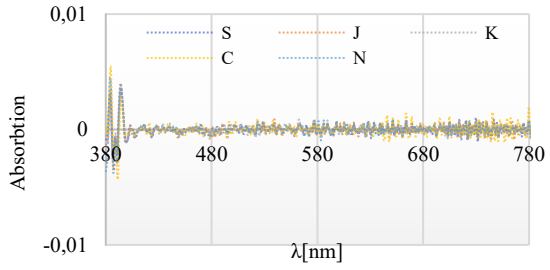


Fig. 2. *Second derivative absorption spectra of the analyzed paprika samples as a function of wavelength*

Assessment of total carotenoids

Based on the results of the absorption analysis, the concentrations of chlorophyll *a*, chlorophyll *b*, total chlorophyll, and total carotenoids were determined. The calculations utilized absorption data at 470 nm, as well as at 648.6 nm and 664.1 nm, which correspond to the absorption maxima of chlorophyll *a* and chlorophyll *b*. The calculations were performed using the following equations [9, 16-17]:

$$C_a = 13.36\, A_{664.1} - 5.19\, A_{648.6} \quad (1)$$

$$C_b = 27.43\, A_{648.6} - 5.19\, A_{664.1} \quad (2)$$

$$C_{a+b} = 5.24\, A_{664.1} + 22.24\, A_{648.6} \quad (3)$$

$$C_{X+C} = \frac{1000\, A_{470} - 2.13\, C_a - 97.64\, C_b}{209} \quad (4)$$

where C_a represents chlorophyll *a*, C_b represents chlorophyll *b*, C_{a+b} represents total chlorophyll, and C_{X+C} represents total carotenoids; all values are expressed in $\mu g \cdot mL^{-1}$. The ratio of chlorophyll *a* to chlorophyll *b* is used as an indicator of the functional pigment composition and functional activity of the photosynthetic apparatus [22]. The results are presented in Table 2.

Table 2. *Concentration of chlorophyll a (C_a), chlorophyll b (C_b), total chlorophyll (C_{a+b}) and total carotenoids (C_{X+C}), in the analyzied paprika samples ($\mu g \cdot mL^{-1}$), along with their respective ratios*

	S	J	K	C	N
C_a	0.285	0.0005	0.030	0.515	0.002
C_b	0.708	0.027	0.183	1.406	0.056
C_{a+b}	0.992	0.028	0.212	1.920	0.058
C_{x+c}	0.581	1.332	0.889	0.885	1.352
C_a/C_b	0.402	0.019	0.162	0.366	0.040
C_{a+b}/C_{x+c}	1.707	0.021	0.239	2.169	0.043

A key indicator of red paprika quality is the total carotenoid content, since carotenoids determine its coloring capacity.

The total carotenoid values (C_{x+c}) in samples N and J (1.338 and 1.332 μg·mL^{-1}, respectively) indicate high carotenoid content. Samples K and C show average content levels (0.890 and 0.889 μg·mL^{-1}, respectively), while sample S has the lowest pigment content (0.584 μg·mL^{-1}). The negative value obtained for chlorophyll *a* in sample N indicates that its chlorophyll *a* content is effectively zero; in other words, the paprika appears to have been produced without admixture of green plant material.

The ratio of chlorophyll *a* to chlorophyll *b* (Chl *a/b*) is used as an indicator of the functional pigment composition and activity of the photosynthetic apparatus [22]. The highest value was recorded for sample S (0.402), and the lowest for sample J (0.019) (Table 2).

The ratio of total chlorophylls to total amount of carotenoids (C_{a+b}/C_{x+c}) serves as an indicator of the degree of green coloration in plants. During the ripening process of fruits and vegetables that change color from green to yellow, orange, or red, this ratio continuously decreases, reaching values below 1.0. The values determined for samples J, K, and N are below 1.0, indicating that the paprikas used were fully ripe (Table 2). Samples C (2.169) and S (1.707) exhibits values greater than 1.0, suggesting either incomplete fruit ripening or the inclusion of green stems and seeds during milling.

Spectral-ratio criterion

The spectral-ratio criterion is determined using the following relationship:

$$Ratio = \frac{A_{520}}{A_{460}} \quad (5)$$

where A_{520} and A_{460} are the absorbance values measured at 520 and 460 nm, respectively. The absorption peak at 520 nm is used to evaluate the extract from green plant or fruit tissue.The resulting values are presented in Fig 3. The obtained results fall within the range of 0.3 to 0.6. The highest value was recorded for sample K, and the lowest for sample N.

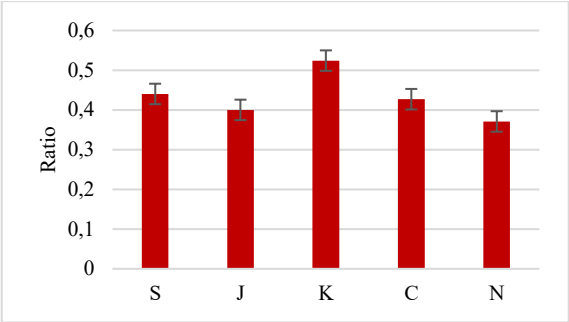


Fig. 3. *Values of the spectral-ratio criterion (Ratio) for the analyzed paprika samples.*

CONCLUSION

In this study, spectrophotometric analysis was carried out on paprika samples purchased at random from retail outlets in the Republic of Bulgaria. All examined commercial samples of sweet red paprika displayed a characteristic absorption maximum in the blue-green region at 460-480 nm. This confirms that their red-orange color is primarily due to natural carotenoid pigments (capsanthin, capsorubin and *β*-carotene). This conclusion is further supported by the calculated ratios of chlorophyll *a* to chlorophyll *b* and C_{a+b}/C_{x+c}.

Future research will include samples of homemade red paprika, as well as a comprehensive selection of samples from the national market.

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REFERENCE

[1] Lal, Bankey, Anshu Kamboj, Vipul Pratap Singh, Bhanu Prakash Singh, Anup Pratap Singh, and Sudhir Kumar Mishra. 2023. Phytochemical and Biological Properties of Domesticated Capsicum Species: A Review. International Journal of Plant & Soil Science 35 (18):427-37. https://doi.org/10.9734/ijpss/2023/v35i183307.

[2] Ananthan, R., Subhash, K., & Longvah, T. 2018. Capsaicinoids, amino acid and fatty acid profiles in different fruit components of

the world hottest Naga king chilli (Capsicum chinense Jacq). Food Chemistry, 238, 51–57.
[3] Dong, X., Li, X., Ding, L., Cui, F., Tang, Z., Liu, Z. 2014. Stage extraction of capsaicinoids and red pigments from fresh red paprika (Capsicum) fruits with ethanol as solvent. LWT - Food Sci. Technol. (Lebensmittel-Wissenschaft -Technol.) 59 (1), 396–402. https://doi.org/10.1016/j.lwt.2014.04.051.
[4] Baenas, N., Belovi´c, M., Ilic, N., Moreno, D.A., García-Viguera, C. 2019. Industrial use of paprika (Capsicum annum L.) derived products: technological benefits and biological advantages. Food Chem. 274 (September 2018), 872–885. https://doi.org/10.1016/j.foodchem.2018.09.047.
[5] Kim, J.S., Ha, T.Y., Kim, S., Lee, S.J., Ahn, J. 2017. Red paprika (Capsicum annuum L.) and its main carotenoid capsanthin ameliorate impaired lipid metabolism in the liver and adipose tissue of high-fat diet-induced obese mice. J. Funct.Foods 31, 131–140.https://doi.org/10.1016/j.jff.2017.01.044
[6] Galvin-King, P.; Haughey, S.A.; Elliott, C.T. Herb and spice fraud; the drivers, challenges and detection. Food Control 2018, 88, 85–97
[7] Horn, B.; Esslinger, S.; Pfister, M.; Fauhl-Hassek, C.; Riedl, J. Non-targeted detection of paprika adulteration using mid-infrared spectroscopy and one-class classification—Is it data preprocessing that makes the performance? Food Chem. 2018, 257, 112–119
[8] Hernández, A.; Martín, A.; Aranda, E.; Bartolomé, T.; Córdoba, J.J. Application of temperature-induced phase partition of proteins for the detection of smoked paprika adulteration by free zone capillary electrophoresis (FZCE). Food Chem. 2007, 105, 1219–1227.
[9]https://ec.europa.eu/food/safety/rasff/portal_en
[10] Damaso Hornero-Mendez, Ricardo Gomez-Ladron de Guevara, M.Isabel Minguez-Mosquera, 2000. Carotenoid biosynthesis changes in five red paprika (Capsicum annuum L.) cultivars during ripening. Cultivar selection for breeding. Journal of Agricultural and Food Chemistry, 48 :3857-3864
[11] Mara DUMA, Ina ALSINA. THE CONTENT OF PLANT PIGMENTS IN RED AND YELLOW BELL PAPRIKAS Scientific Papers, Series B, Horticulture, Vol. LVI, 2012 ISSN Online 2286-1580, ISSN-L 2285-5653
[12] Khoo H-E, Prasad KN, Kong K-W, Jiang Y, Ismail A. Carotenoids and their isomers: color pigments in fruits and vegetables. Molecules. 2011;16:1710–1738. doi: 10.3390/molecules16021710.
[13] Arimboor R, Natarajan RB, Menon KR, Chandrasekhar LP, Moorkoth V. Red paprika (Capsicum annuum) carotenoids as a source of natural food colors: analysis and stability-a review. J Food Sci Technol. 2015 Mar;52(3):1258-71. doi: 10.1007/s13197-014-1260-7
[14] Ong ASH, Tee ES. Natural sources of carotenoids from plants and oils. Methods Enzymol. 1992;213:142–167.
[15] Ranjith A, Kumar K, Venugopalan V, Arumughan C, Sawhney R, Singh V. Fatty acids, tocols, and carotenoids in pulp oil of three sea buckthorn species (Hippophae rhamnoides, H. salicifolia and H. tibetana) grown in the Indian Himalayas. J Am Oil Chem Soc. 2006;83(4):359–364.
[16] Topuz, Ayhan, Dincer, Cuneyt, Özdemir, Kubra Sultan, Feng, Hao, & Kushad, Mosbah (2011). Influence of different drying methods on carotenoids and capsaicinoids of paprika (Cv., Jalapeno). Food Chemistry, 129(3), 860–865.
[17] Hornero-Méndez D, Mínguez-Mosquera MI. Rapid spectrophotometric determination of red and yellow isochromic carotenoid fractions in paprika and red paprika oleoresins. J Agric Food Chem. 2001 Aug;49(8):3584-8. doi: 10.1021/jf010400l. PMID: 11513632.
[18] Yaldiz, Gulsum, Ozguven, Mensure, & Sekeroglu, Nazim (2010). Variation in capsaicin contents of different Capsicum species and lines by varying drying parameters. Industrial Crops and Products, 32(3), 434–438.
[19] Hartmut K. Lichtenthaler. 1987. Chlorophylls and Carotenoids: Pigments of Photosynthetic Biomembranes. Methods in enzymology, vol. 148, 350-382
[20] J. Muñoz-Postigo, E.M. Valero, M.A. Martínez-Domingo, F.J. Lara, J.L. Nieves, J. Romero, J. Hernández-Andrés, Band selection pipeline for maturity stage classification in bell paprikas: From full spectrum to simulated camera data, Journal of Food Engineering, Volume 365, 2024,

111824, ISSN 0260-8774, https://doi.org/10.1016/j.jfoodeng.2023.111824.

[21] Hartmut K. Lichtenthaler, Claus Buschmann. 2001. Chlorophylls and Carotenoids: Measurement and Characterization by UV-VIS Spectroscopy, Current Protocols in Food Analytical Chemistry, F4.3.1-F4.3.8

[22] H. K. Lichtenthaler, C. Buschmann, M. Doll, H-J. Fietz, T. Bach, U. Kozel, D. Meier, U. Rahmsdorf. 1981. Photosynthetic activity, chloroplast ultrastructure, and leaf characteristics of high-light and low-light plants and of sun and shade leaves, Photosynthesis Research 2: 115-141. 0166-8595/81/0022-0115