

## APPRECIATION OF QUALITY FOR SOME TOMATOES VARIETIES BY USING THE DETERMINATION OF PHYSICAL-CHEMICAL PARAMETERS

Daniela Giosanu\*, Mădălina Tudor-Radu\*\*, Loredana Elena Vijan\*\*\*

\*University of Pitesti, Faculty of Sciences, Targu din Vale Street no. 1, Pitesti, Arges, Romania  
E-mail: [giosanu@yahoo.com](mailto:giosanu@yahoo.com)

\*\*University of Pitesti, Faculty of Sciences, Targu din Vale Street no. 1, Pitesti, Arges, Romania,  
University of Craiova, Faculty of Agriculture and Horticulture, "A.I. Cuza" Street No. 13, Craiova, Dolj, Romania  
E-mail: [tudorradumadalina@yahoo.com](mailto:tudorradumadalina@yahoo.com)

\*\*\*University of Pitesti, Faculty of Sciences, Targu din Vale Street no. 1, Pitesti, Arges, Romania  
E-mail: [vloredana2005@yahoo.com](mailto:vloredana2005@yahoo.com)

### Abstract

*The studies were done on three varieties of tomatoes from Spain (red cherry, yellow cherry and kumato cherry). The following parameters: pH, soluble solids, content of water (moisture), content of minerals (ash), titratable acidity and content of some bioactive compounds (vitamin C, polyphenols, flavonoids, anthocyanins and carotenoids) were determinate in order to appreciate the quality of these products. The results showed the influence of the varieties on the values of bioactive compounds with antioxidant activity.*

*Keywords: tomatoes, vitamin C, polyphenols, flavonoids, anthocyanins, carotenoids*

### 1. INTRODUCTION

Food consumption is mainly aimed at maintaining the functionality of the body, the food giving important basic substances needed by the human body. The composition of a food product largely determines its safety, nutrition, quality attributes and sensory characteristics. Most foods are compositionally complex materials made up of a wide variety of different chemical constituents. These substances are classified into: macronutrients, micronutrients, dietary fibre, water, bioactive compounds.

The human body needs a large amount of macronutrients to provide the energy needed to maintain functionality to achieve daily activities. Macronutrients are divided into carbohydrates, proteins and lipids.

Micronutrients are comprised of vitamins and minerals. They are substances that cannot be synthesized to the body but are introduced into the body through food consumption. These substances have not energy and are needed in smaller amounts than macronutrients but hold positions of high importance in vital processes in the body.

Dietary fibres are the food part resistant to digestion and absorption in the small intestine, with the main role in the digestion stimulation.

Water is the component that is found in the largest amount in food and contributes to the total intake of water required of the human body, while bioactive compounds are compounds found in small

quantities in fruits and vegetables, but with an important role in maintaining a healthy body (because it helps in the fight against degenerative diseases).

The interest in bioactive compounds grows more and more, they having the applications in different branches: geo-medicine, plant science, agro-chemistry, food industry, etc. If in the beginning the people used the plants as food only for their nutritional value, over time they found that the plants have an important medical role due to bioactive compounds that contain them, compounds with strong antioxidant effect.

Tomatoes are the most widely consumed vegetables all year round, fresh and conservation in various ways; there are considered a food, a medicine and simultaneously a flavour. Tomatoes are available in a huge range of shapes, sizes and colours. There are more than 700 tomato varieties in cultivation today, being in about every colour from white to purple, pink, yellow, orange, mottled or striped, with different names. Regardless of its name, the tomato is known as *pomme d'amour* in French and "*pomodoro*" in Italy.

In our paper, the results obtained at the analysis of three varieties of cherry tomatoes (red cherry, yellow cherry and kumato cherry), grown in Spain are presented. For these cherry tomatoes, the water and minerals content, the soluble solids, pH, the titratable acidity and the content of some bioactive compounds (vitamin C, polyphenols, flavonoids, anthocyanins and carotenoids) were determined.

## 2. MATERIALS AND METHODS

### Plant material

Three varieties of cherry tomatoes (red cherry, yellow cherry and kumato cherry), with the indeterminate growth, grown in Almeria, Andalusia, Spain, were analysed. Vigorous plants, resistant to harsh conditions of development, these varieties of tomatoes produce the fruits in abundance throughout the summer, arranged in a cluster of about 18 to 22 grams (red cherry) or 25 to 30 grams (yellow cherry and kumato cherry), with a round shape, pointed at the base, very sweet and flavour, scented and juicy. It distinguishes the different colour: red at the red cherry variety, yellow at the yellow cherry variety, and reddish brown to dark red at the kumato cherry variety. Kumato cherry tomatoes are the research fruits, being the mixture of different varieties, using the traditional and natural methods. The tomatoes were washed, wiped with a paper towel, cut into pieces and converted into a homogeneous puree with a vertical mixer. The samples were stored in a refrigerator.

### Chemical substances

All reagents: standards of phenolic acids (gallic acid) and flavonoids (catechin), Folin-Ciocalteu's reagent, methanol, ethanol, acetone, hexane, sodium hydroxide, sodium carbonate, sodium nitrite, hydrochloric acid and aluminium chloride were purchased from Redox Bucharest - Sigma Aldrich, Romania.

### Methods

The water content (moisture) was determined gravimetrically by drying a 1 g tomato in an oven at 105-110 °C for 2 hours (AOAC, 1990).

The minerals content (ash) was determined by the calcination of residue after determining the water content at 550 °C for 4 hours (AOAC, 1990).

Soluble solids were determined using a Kruss DR201-95 refractometer and the results were reported as °Brix at 20 °C (AOAC, 1990).

pH was measured by the squeezing of tomatoes, using a multimeter C-561, after calibration of the apparatus with solutions of pH 7 and 4 (AOAC, 1990).

The titratable acidity was determined by titration of the cherry tomatoes samples with a solution of NaOH 0.1 M (AOAC, 1990), using the bromothymol blue (as indicator) and the results were expressed in g malic acid/ 100 g fruit.

Content of vitamin C expressed in mg/ 100 g of tomato, was determined using iodometric method (Takács-Hájos and Zsombik, 2015), by titration of the tomatoes samples with a solution of potassium iodate 0.0017 M freshly prepared, using starch 1 % (as indicator).

The polyphenols, flavonoids, anthocyanins and carotenoids content was determined by spectrophotometric method, with a UV-Vis spectrophotometer PerkinElmer Lambda25, using the methanolic extracts from the homogeneous puree of analysed cherry tomatoes.

The polyphenols were quantified by reaction with the Folin-Ciocalteu reagent, in accordance with the methodology proposed by Singleton and Rossi, 1965, based on forming a blue coloured compound between phosphotungstic acid and polyphenols, in an alkaline medium. The polyphenols content was expressed as mg gallic acid/ 100 g of extract.

The flavonoids content was determined by the methodology proposed by Zhishen et al., 1999, based on forming a yellow-orange-coloured compound by the reaction of flavonoids and aluminium chloride. The flavonoids content was expressed as mg catechin/ 100 g of extract.

The anthocyanins content was determined by extraction with the solution of hydrochloric acid 1%, in accordance with the methodology proposed by Di Stefano and Cravero (1989). The anthocyanins content was expressed as cyanidin 3-glucoside mg/ 100g fruit.

The carotenoids content was determined in accordance with the methodology proposed by Zechmeister and Polgar, 1943, by extraction with mixture of hexane, ethanol and acetone in 2:1:1 volume ratio. The content of carotenoids (lycopene and  $\beta$ -carotene) was calculated using its extinction molar coefficients of  $184900 \text{ M}^{-1}\text{cm}^{-1}$  at 470 nm and  $172000 \text{ M}^{-1}\text{cm}^{-1}$  at 503 nm for lycopene and  $108427 \text{ M}^{-1}\text{cm}^{-1}$  at 470 nm and  $24686 \text{ M}^{-1}\text{cm}^{-1}$  at 503 nm for  $\beta$ -carotene, in hexane (Zechmeister and Polgar, 1943; Rubio-Diaz et al., 2011).

### 3. RESULTS AND DISCUSSIONS

Food products are analysed for a variety of reasons, e.g. the assessment of product quality, the determination of nutritive value, the detection of forgery, research and development.

Table 1 presents the results regarding the water content, the minerals content, soluble solids, pH and titratable acidity from three tomato samples.

*Table 1. Moisture content, ash content, soluble solids, pH and acidity values*

Variety	Moisture, %	Ash, %	Soluble solids, °Brix	pH at 20 °C	Titratable acidity, g malic acid/100g fruit
red cherry	90.74	9.1	5.8	4.46	1.07
yellow cherry	91.25	8.5	5.4	4.41	1.47
kumato cherry	90.35	9.2	4.7	4.31	0.94

The water (moisture) content is one of the most commonly measured properties for the vegetables. It is important to determine this content because the texture, the taste, the appearance and the stability of foods depends on the amount of water they contain. The high water content gives the tenderness and the freshness of the vegetables, this state being required in during its processing. It is therefore important for the researchers to be able to measure the moisture contents of the foods. There are analytical techniques available that can provide some information about the relative fractions of water in different molecular environments (e.g. DSC, NMR, vapour pressure). The

moisture content is determined by measuring the mass of the food before and after the water was removed by evaporation. The basic principle of this technique is that water has a lower boiling point than the other major components within foods, e.g. lipids, proteins, carbohydrates and minerals. Sometimes a related parameter, known as the total solids, is reported as a measure of the moisture content. It is noted that all cherry tomatoes have water content between 90 and 92%, values closed to those reported by Gupta et al., 2011; Tudor Radu et al., 2016.

Ash is the inorganic residue remaining after the water and the organic matter have been removed by heating. The ash content is a measure of the total amount of minerals present within a food whereas the mineral content is a measure of the amount of specific inorganic components present within a food, such as calcium, potassium, sodium, etc. It is important to know the mineral content of foods because its quality depends on the concentration and type of minerals they contain. Some minerals are essential to a healthy diet (e.g. calcium, phosphorous, potassium and sodium) whereas others can be toxic (e.g. lead, mercury, cadmium and aluminium). High mineral contents are sometimes used to retard the growth of certain microorganisms. The analytical techniques for providing information about the mineral content are based on the fact that the minerals can be distinguished from all the other components within a food in some measurable way. The usual used methods are based on the fact that minerals are not destroyed by heating and they have a low volatility compared to other components from foods. The calcination of residue after determining the water content is used as the first step in preparing samples for analysis of specific minerals, by atomic spectroscopy or the various traditional methods. Ash content in cherry tomatoes ranged between 8.5% at yellow cherry and 9.2% in kumato cherry. These results are close to those presented by Pinela et al., 2012, which reported values between 6.3% and 9.37% for dry matter content in four tomato varieties grown in North-Eastern Portugal.

The dry substance from tomatoes consists of soluble solids in water (about 88%), such as sugar, proteins and water-soluble pectic substances. Cellulose, pectin and protopectin acid are water-insoluble components (Renquist and Reid, 1998). Thus, the soluble solids are a measure of the sugar content that is dissolved within a food. It is measured using a refractometer and is expressed in degrees Brix. At 20°C, the Brix scale measure the percentage of sucrose (sugar) in the solution reported at 100 grams sample (9° Brix is equivalent to a sugar content of 9%). In the analysed cherry tomato varieties an amount of soluble solids was determined between 4.4°Brix (kumato cherry) and 5.8°Brix (red cherry), these results being nearby to the values obtained Helyes et al., 2014, who reported values between 4.58 and 5.98 for the soluble solids from the different tomato varieties.

Other important parameter for assessing the food quality is the acidity. There are two forms to express the food acidity: the hydrogen ion concentration (or pH) and the titratable acidity. The optimum pH range (3.7 - 4.5), reported by Sulieman et al., 2011, is considered to be a limiting factor for the preservation and storage of tomatoes in the form of tomato pasta. pH for the analysed tomato varieties ranged between 4.31 and 4.46, the values being included in the optimum pH range, reported by Sulieman et al., 2011. The titratable acidity is a measure of the total amount of acid from the solution, being determined by the titration with a standard solution of sodium hydroxide, in presence an acid sensitive colour indicator, such as phenolphthalein or bromothymol blue. The acidity level in tomatoes is associated with the sensory attributes, such as flavour and astringency. The titratable acidity values for the three cherry tomato varieties ranged from 0.94 g malic acid/100 g fruit (kumato cherry) and 1.47 g malic acid/100 g fruit (yellow cherry). Similar results were reported by other authors who carried out studies of the different tomato genotypes (Mechlouch et al., 2012; Gupta et al., 2011; Tudor Radu et al., 2016).

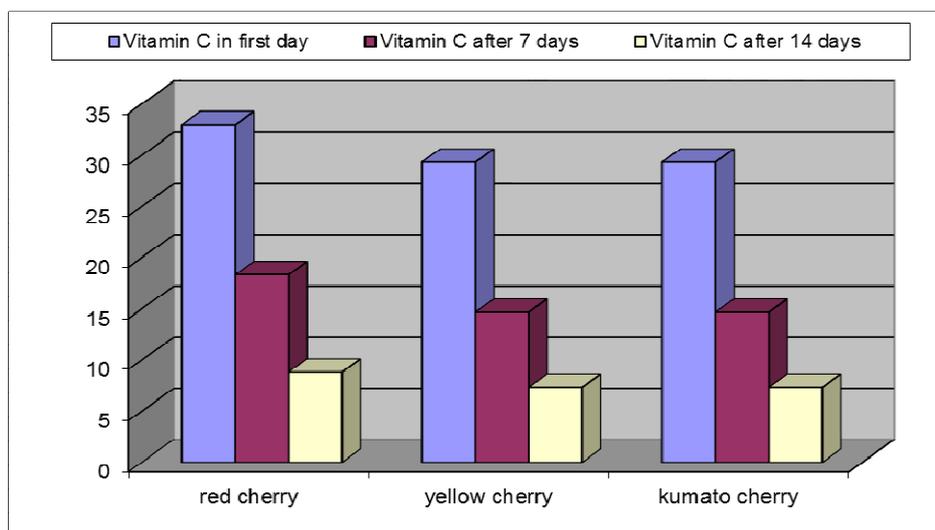
Tomatoes have antioxidant features due to the presence of bioactive compounds, such as vitamin C, polyphenols, flavonoids, anthocyanins, and carotenoids (Cernîşev and Şleagun, 2007). The level of these compounds in the body is an indicator of health, they operated as biomarkers for the food quality (Costin and Segal, 1999). Table 2 shows the content of vitamin C, polyphenols, flavonoids, anthocyanins and carotenoids (lycopene and  $\beta$ -carotene) from three tomato samples.

**Table 2. Content of some antioxidant compounds (mg/ 100g fruit) in cherry tomatoes analysed**

Variety	Vitamin C	Polyphenols	Flavonoids	Anthocyanins	Lycopene	$\beta$ -carotene
red cherry	32.99	565.4	25.59	14.25	10.57	1.04
yellow cherry	29.32	678.8	35.15	23.14	0.11	0.93
kumato cherry	29.32	46.8	31.79	11.53	6.02	3.18

Note: Polyphenols were expressed as mg gallic acid equivalent/ 100 g fruit; Flavonoids were expressed as mg catechin equivalent/ 100 g fruit; Anthocyanins were expressed as mg cyaniding-3-glucoside equivalents/ 100 g fruit

Vitamin C is probably best known as an antioxidant. Antioxidants are molecules that help keep the chemical reactions in our body in check. In particular, antioxidants help prevent the excessive activity on the free radicals. Vitamin C is required to produce collagen, a protein that plays a critical role in the structure of our bodies. In the analysed cherry tomatoes, the vitamin C content ranged between 29.32 mg/ 100 g fruit (yellow cherry and kumato cherry) and 32.99 mg/ 100 g fruit (red cherry). Similar results were obtained by Tudor Radu et al., 2016, who reported the vitamin C content between 22.61 and 32.21 mg/ 100 g, for the various genotypes of tomatoes. The evolution of the vitamin C content in time is shown in figure 1.



**Figure 1. Modification of vitamin C content in cherry tomatoes analysed**

The initial level of vitamin C corresponding to fresh cherry tomatoes and values after 7 and 14 days of harvest are presented as well. The observed decrease of the vitamin C content during storage is a result of the vegetables aging. These results correspond to the findings of studies carried out by Christakou *et al.*, 2005 and Tudor Radu et al., 2016, who found that an increase of the storage period causes the decrease of the vitamin C content in fruits and vegetables. The decomposition of

vitamin C during storage of vegetables is caused by the ascorbic acid degradation due to oxidation in the presence of oxidising enzymes.

Polyphenols are the most abundant antioxidants in our diet and are widespread constituents of the vegetables. These compounds prevent oxidative damage to lipids and thus increase the nutritional value of the food. Polyphenols are divided into several classes, i.e. phenolic acids, flavonoids, stilbenes and lignans. Flavonoids are molecules with a phenolic benzopyran structure and may themselves be divided into six subclasses as a function of the type of heterocycle involved: flavonols, flavones, isoflavones, flavanones, anthocyanidins and flavanols.

The concentration of polyphenols and flavonoids was calculated using the calibration curves, presented in figure 2, by the utilization of the absorbance values at 750 nm, respectively 510 nm. Finally, the polyphenols content was expressed as mg gallic acid/ 100 g fruit and the flavonoids content was expressed as mg catechin/ 100 g fruit.

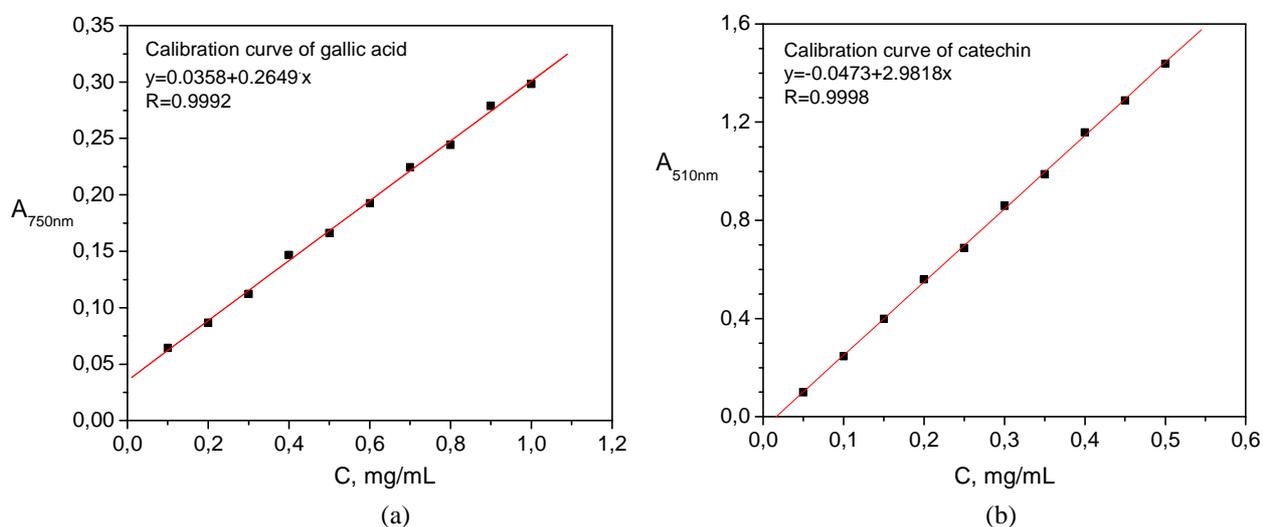


Figure 2. The calibration curves for polyphenols (a) and flavonoids (b)

The polyphenols content in analysed cherry tomatoes showed values ranging from 46.8 mg gallic acid/ 100 g fruit (kumato cherry) and 678.8 mg gallic acid/ 100 g fruit (yellow cherry). Similar results, except kumato cherry tomato, were obtained by Helyes et al., 2012; Martínez-Valverde et al., 2002 and Mechlouch et al., 2012.

The flavonoids content in analysed tomatoes showed values between 25.59 mg catechin/ 100 g fruit (red cherry) and 35.15 mg catechin/ 100 g fruit (yellow cherry). Similar results were obtained by Helyes et al., 2012; Martínez-Valverde et al., 2002 and Mechlouch et al., 2012.

The anthocyanins are members of the flavonoid group of phytochemicals, being pigments soluble, responsible for the red, purple and blue pigmentation of vegetables and also play important roles in plant physiology, such as attractants for insect pollinators and seed dispersal. They have been shown to play a beneficial role in: visual acuity, cancer, heart disease, neurodegenerative disorders. The investigated cherry tomatoes were found containing anthocyanins between 11.53 mg cyanidin-3-glucoside/ 100 g fruit (kumato cherry) and 23.14 mg cyanidin-3-glucoside/ 100 g fruit (yellow cherry). These results are comparable with those presented by Mes et al., 2008 and Tudor Radu et al., 2016, who reported the anthocyanin content between 7.79 mg cyanidin-3-glucoside/ 100 g fruit and 415 mg cyanidin-3-glucoside/ 100 g fruit to different tomato genotypes.

Carotenoids are derived from a 40 carbon polyene chain, which could be considered the backbone of the molecule. This chain may be terminated by cyclic end-groups and may be complemented

with oxygen-containing functional groups. They act as antioxidants, fighting against free radicals (works against premature ageing and cons the appearance of blemishes on the skin) and facilitate the cellular communication. Lycopene and  $\beta$ -carotene absorb light in the 350-550 nm region of the visible spectrum. The literature (Zang et al., 1997; Zechmeister and Polgar, 1943) show the characteristics of the UV-Vis absorption spectra of the two carotenoids dissolved in hexane. The UV-Vis spectra of the two carotenoids have three absorption peaks located at 443, 471 and 502 nm for lycopene and 425, 450 and 478 nm for  $\beta$ -carotene. It is noted that the absorption maximum at 360 nm indicates the presence of lycopene *cis*-isomers.

Figure 3 indicates the UV-Vis absorption spectra of the cherry tomatoes supernatant, obtained by extraction of carotenoids with mixture of hexane, ethanol and acetone.

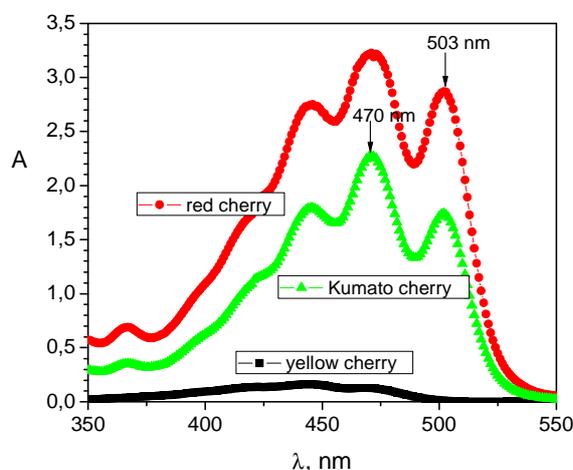


Figure 3. UV-Vis spectra of cherry tomatoes supernatant

By analysing the UV-Vis absorption spectra of the cherry tomatoes supernatant, the content of those two carotenoids was calculated. Thus, the content of lycopene in cherry tomatoes analysed ranged from 0.11 mg/ 100 g fruit (yellow cherry) to 10.57 mg/ 100 g fruit (red cherry) and the content of  $\beta$ -carotene ranged from 0.93 mg/ 100 g fruit (yellow cherry) to 3.18 mg/ 100g fruit (kumato cherry). Similar results, except yellow cherry tomato, were obtained by Helyes et al., 2012, 2014 and Martínez-Valverde et al., 2002. At yellow cherry tomatoes was found a very low content of lycopene (0.11 mg/ 100 g fruit) and a much higher content of  $\beta$ -carotene (0.93 mg/ 100 g fruit), the values which matches with the yellow colour of these tomatoes.

#### 4. CONCLUSIONS

Significant differences were detected between three varieties of tomatoes from Spain (red cherry, yellow cherry and kumato cherry) regarding the chemical composition, in particular the content of vitamin C, polyphenols, flavonoids, anthocyanins and carotenoids. A remarkable variety is red cherry tomato, which revealed an interesting nutritional composition, showing a high antioxidant status due to its high content of vitamin C, polyphenols, flavonoids and lycopene. Also, a remarkable variety is yellow cherry tomato, which showed a high antioxidant status due to its high content of vitamin C, polyphenols, flavonoids and anthocyanins, the only limitation being the reduced content of carotenoids. In conclusion, since the analysed tomatoes species were grown in the same conditions for agriculture, geography and climate (Almeria, Andalusia, Spain), the results showed variability in the bioactive components content due to the influence of the variety.

## 6. REFERENCES

- Association of Official Analytical Chemists (AOAC) (1990). *Official methods of analysis*. 15<sup>th</sup> ed., In Helrich, K. (ed.). Washington, DC, USA.
- Cernișev, S., Șleagun, G. (2007). Influence of dehydration technologies on dried tomato biological quality and value. *Cercetări agronomice în Moldova*, anul XXXX, 3(131), 63-68.
- Christakou, E.C., Arvanitoyannis, I.S., Khah, E.M., Bletsos, F. (2005). Effect of grafting and modified atmosphere packing (MAP) on melon quality parameters during storage. *Journal of Food Agriculture and Environment* 3(1), 145-152.
- Costin, G.M., Segal, R. (1999). *Alimente funcționale*. Editura Academica, Galați, 39-71.
- Di Stefano, R., Cravero, M.C. (1989). I composti fenolici e la natura del colore dei vini rossi. *L'enotecnico Ottobre*, 81-87.
- Gupta, A., Kawatra, A., Sehgal, S. (2011). Physical-chemical properties and nutritional evaluation of newly developed tomato genotypes. *African Journal of Food Science and Technology* 2(7), 167-172.
- Helyes, L., Lugasi, A., Pék, Z. (2012). Effect of irrigation on processing tomato yield and antioxidant components. *Turkish Journal of Agriculture and Forestry* 36, 702-709.
- Helyes, L., Lugasi, A., Daood, H.G., Pék, Z. (2014). The simultaneous effect of water supply and genotype on yield quantity, antioxidants content and composition of processing tomatoes. *Notulae Botanicae Horti Agrobotanici* 42(1), 143-149.
- Martínez-Valverde, I., Periago, M.J., Provan, G., Chesson, A. (2002). Phenolic compounds, lycopene and antioxidant activity in commercial varieties of tomato (*Lycopersicon esculentum*). *Journal of the Science of Food and Agriculture* 82(3), 323-330.
- Mechlouch, R.F., Elfalleh, W., Ziadi, M., Hannachi, H., Chwikhi, M., Ben Aoun, A., Elakesh, I., Cheour, F. (2012). Effect of different drying methods on the physico-chemical properties of tomato variety 'Rio Grande'. *International Journal of Food Engineering* 8(2), article 4.
- Mes, P.J., Boches, P., Myers, J.R. (2008). Characterization of tomatoes expressing anthocyanin in the fruit. *Journal of the American Society for Horticultural Science* 133(2), 262-269.
- Pinela, J., Barros, L., Carvalho, A.M., Ferreira, I.C. (2012). Nutritional composition and antioxidant activity of four tomato (*Lycopersicon esculentum* L.) farmer' varieties in Northeastern Portugal homegardens. *Food Chem. Toxicol.* 50(3-4), 829-834.
- Renquist, A.R., Reid, J.B. (1998). Quality of processing tomato (*Lycopersicon esculentum*) fruit from four bloom dates in relation to optimal harvest timing. *New Zealand Journal of Crop and Horticultural Science* 26(2), 161-168.
- Rubio-Díaz, D.E., Francis, D.M., Rodríguez-Saona, L.E. (2011). External calibration models for the measurement of tomato carotenoids by infrared spectroscopy. *Journal of Food Composition and Analysis* 24(1), 121-126.
- Singleton, V.L., Rossi, J.A.Jr. (1965). Colorimetry of total phenolics with phosphomolybdicphospho-tungstic acid reagents. *American Journal of Enology and Viticulture* 16, 144-158.
- Suliman, A.M.E., Awn, K.M.A., Yousif, M.T. (2011). Suitability of some tomato (*Lycopersicon esculentum* Mill.) genotypes for paste production. *Journal of Science and Technology* 12, 45-51.
- Takács-Hájos, M., Zsombik, L. (2015). Total polyphenol, flavonoid and other bioactive materials in different asparagus cultivars. *Notulae Botanicae Horti Agrobotanici* 43(1), 59-63.
- Tudor-Radu, M., Mitrea, R., Tudor-Radu, C.M., Tița, I. (2014). The characterization of some new varieties of tomatoes which have been grown under ecological condition certificated at I.N.C.D.B.H. Stefanesti, *Analele Universității din Craiova, seria biologie, horticultură, tehnologia prelucrării produselor agricole, ingineria mediului*, XIX (LV), 617-623.
- Tudor-Radu M., Vijan L.E., Tudor-Radu, C.M., Tița, I., Sima R., Mitrea, R. (2016). Assessment of Ascorbic Acid, Polyphenols, Flavonoids, Anthocyanins and Carotenoids Content in Tomato Fruits. *Notulae Botanicae Horti Agrobotanici* 44(2), 477-483.
- Zang, L.Y., Sommerburg, O., van Kuijk, F.J. (1997). Absorbance changes of carotenoids in different solvents. *Free Radical Biology & Medicine* 23(7), 1086-1089.
- Zechmeister, L., Polgar, A. (1943). *cis-trans* Isomerization and spectral characteristics of carotenoids and some related compounds. *Journal of the American Chemical Society* 65(8), 1522-1528.
- Zhishen, J., Mengcjheng, T., Jianming, W. (1999). The determination of flavonoid contents in mulberry and their scavenging effects on superoxide radicals. *Food Chemistry*, 64(4), 555-559.