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IMPROVING THE QUANTITATIVE AND QUALITATIVE PRODUCTION OF PEPPERS UNDER THE INFLUENCE OF DIFFERENT FOLIAR FERTILIZATION TREATMENTS

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Abstract

The increasing nutritional properties of vegetable species as well as their use in disease prevention has become a growing concern among researchers. This paper aims to study the influence of foliar fertilizers on the nutritional properties of pepper. The biological material was represented by two varieties of Capsicum annuum, var. tetragonum ('Cornel') and Capsicum annuum var. longum ('Max'). The fertilization procedure of the experiment consisted in the application of N_{45}: P_{45}: K_{45} followed by foliar fertilization with Agroleaf Power 31:11:11, Agroleaf Total 20:20:20 and Agroleaf High K combined with Crop'Max'. For this purpose, production and quality elements (the content of soluble dry matter, titratable acidity, vitamin C content, reducing sugars, phenolic compounds and lycopene content and antioxidant activity) were determined. The results obtained in the fertilized varieties indicate an increase in the production and vitamin C and titratable acidity content, but a decrease in solid soluble dry matter and in reducing sugars content in both pepper varieties. Comparing the two varieties, the foliar fertilization increased the antioxidant activity caused by the vitamin C content (201.35 mg/100 g FW), the phenolic compounds (251.71 mg/100 g FW) and the lycopene content (126.91 mg/100 g FW) in the var. longum ('Max').

Key words: *pepper, vitamin C, lycopene, total polyphenols.*

INTRODUCTION

The pepper (*Capsicm annuum* L.) originates in Central America and South America. Christopher Columbus found the pepper in Haiti, from where it was brought to Europe and grown for the first time in Spain and Portugal, then in Germany, England, and Hungary in the middle of the 16th century. The pepper reached Romania much later, being brought by the Bulgarian gardeners in the 18th century, and being cultivated firstly in the south of the country, then in the other favourable areas.

The pepper fruits have high nutraceutical value due to their high content of sugars, vitamins and antioxidant capacity, but also due to the fact that the pepper is consumed fresher, a state in which these components are processed directly by the human body.

The proper fertilization of field crops is one of the most important factors affecting their growth and development as well as their production and quality (Rahman et al., 2014). Studies on the fertilizers effect on the production and quality of pepper fruits have been conducted and they are still being carried out but this mechanism has not yet been elucidated as there is a multiple complex of factors influencing the absorption of macro and microelements. Potassium is one of the main macronutrients contributing up to 6% to the dry weight of the plants (Shabala, 2003) and it is considered a key factor in the fruit quality. Flores et al., (2004) conducted some observations on the influence of K^+ on the quality of olive fruits and found that this is related to the nutritional quality of soil or to the applied foliar fertilization. Kaya and Higgs, (2003) found in peppers that the potassium ions influence on production yields is mainly related to the amelioration of the negative effects of salinity stress, and Xu et al., (2002) found that 15-30% of the total N from the nutrient solution could increase both the total fruit yield and the efficiency of K-fertilizer used in sweet pepper. The influence of the mineral nutrition on the quality characteristics, such as soluble solids, acidity, pH, and fruit shape index, have not been studied extensively in pepper plants, an issue which is important both for fresh and processed horticultural production.

In recent years, the research has focused on the use of different types of organic fertilizer in crops of vegetables, cabbage (Soare et al., 2017), eggplant (Becherescu et al., 2016), tomatoes (Dinu et al., 2013), melon (Dinu et Soare, 2017) or salad (Drăghici et al., 2016) in order to improve the quantity and quality of production. The use of chemical or organic products has a relevant effect not only in culture but also in shortening and stimulating the germination of pepper seeds (Drăghici et al. 2012, Bălan et al., 2014), tomatoes (Dinu et al., 2013; 2015) or floriculture crops (Manda et al., 2014).

The importance of the main nutrients that regulate the yield and quality components of pepper crops requires further studies. This is necessary to establish a rational fertilization both at the ground level and especially at the leaf level. In this respect, there is currently a lack of studies on pepper plants using nutrient concentrations similar to those used by growers. It is assumed that the current nutritional balances based on foliar solutions can be adapted to the pepper crops in order to improve the yield and the fruits quality.

This study focused on the effects of chemical foliar fertilizers, combined with the Crop'Max' organic product, on the yields and quality parameters of a field crop of bell and long pepper.

MATERIALS AND METHODS

The experiment was placed in the didactic field of the Faculty of Horticulture in Craiova between 2017-2018 period. The biological material was represented by two varieties of pepper, var. *longum* (''Max'') and var. *tetragonum* (''Cornel''). The experiment was placed in the field on an agrofond fertilized with Complex III (300 kg/ha) with 80 cm between the rows and 30 cm between the plants on the row. During the vegetation period, 3 foliar treatments were applied in the following combinations: Agroleaf Power 31:11:11, the first treatment, Agroleaf Total 20:20:20, the second treatment and Agroleaf High K combined with Crop'Max', the third treatment. During the vegetation period, the classical technology for cultivation was applied.

Analytical methods were applied, as fallows. Total soluble solids (TSS) was determined using a digital refractometer (Handheld Dr 301-95) at 20° C and expressed as %.

The titratable acid content (acidity) was determined by titration with 0.1 N sodium hydroxide (NaOH) using phenolphthalein as indicator and expressed as % citric acid.

The ascorbic acid was extracted from biological material in 2% HCl (1:10w/v). The ascorbic acid content was performed with iodometric titration from the supernatant in which iodine reacts with ascorbic acid, oxidizing it to dehydroascorbic acid. The redox titration endpoint is determined by the first iodine excess that is complexed with starch, giving a deep blue-violet color. The ascorbic acid content was expressed as mg/100 g fresh weight.

Reducing sugars (%) were extracted in distilled water (1:50 w/v), 60 minutes at 60 ˚C and assayed colorimetric at 540 nm with 3.5 dinitrosalicylic acid using glucose as standard (Soare et al., 2017).

Lycopene and β-carotene were extracted in 2:1:1 hexane:methanol:acetone (1g:25mL) 30 minutes in the dark. Further, 5 mL of distilled water are added and the solution is shaked for 5 minutes. After phase separation, the non-polar layer was collected and spectrophotometrically analysed versus a blank of hexane solvent. For analyzing the levels of total carotenoids, the absorbance was measured at 450 nm and results were calculated using a value of 2500 for the extinction coefficient (E1%). For the determination of lycopene content, absorbance was measured at 503 nm and the results were calculated using a value of 17.2 10^4/M/cm for the molar extinction coefficient for lycopene in hexane (Paraschivu et al., 2014). The results are expressed in mg / 100g fresh weight.

The extracts for the determination of total phenolic content and antioxidant activity were prepared into 80% aqueous methanol (1:10 v/v) at 24 °C for 16 h. The resulting slurries were centrifuged at 4000 rpm for 5 min and the supernatants were collected.

The total phenolics content was determined colorimetrically at 765 nm with the Folin Ciocalteu reagent method (Soare et al., 2015). The results were calculated with a standard curve prepared using gallic acid and expressed as mg gallic acid equivalents (GAE)/100g fresh weight.

Antioxidant activity was realised using DPPH (2,2-diphenyl-1-picrylhydrazyl) radical scavenging assay:

The capacity of extracts to reduce the radical 2,2-diphenyl-1-picrylhydrazyl has been evaluated colorimetrically (Soare et al., 2018).

2 mL of 0.075 mM DPPH solution in methanol was mixed with 0.1 mL sample methanolic extract and after 20 minutes the absorbance of the remaining DPPH radicals was measured at 515 nm. A blank reagent was used to study stability of DPPH over the test time. The Trolox calibration curve was plotted as a function of the percentage of DPPH radical scavenging activity. The results were expressed as μM Trolox equivalents (TE)/1g f.w.

ABTS radical cation scavenging activity was measured colorimetric at 734 nm (Paraschivu et al., 2014). ABTS radical cation was produced by reacting 7 mM ABTS solution with 2.45 mM potassium persulfate (final concentration) and allowing the mixture to stand in the dark at room temperature for 16 h before use. The ABTS radical cation solution was diluted with 80% methanol to an absorbance of 0.70 at 734 nm. 0.1 mL of the sample extract was mixed with 2.9 mL of diluted ABTS radical cation solution. After reaction at room temperature for 6 min, the absorbance at 734 nm was measured. The Trolox calibration curve was plotted as a function of the percentage of ABTS radical cation scavenging activity. The final results were expressed as μM Trolox equivalents (TE) per 1g.

All the spectrophotometric measurements were carried out using Evolution 600 UV-Vis spectrophotometer, Thermo Scientific, England, with VISION PRO software.

The statistical calculation was performed using Anova, and the LSD were calculated for P = 0.05 to assess the quantitative and qualitative characteristics of pepper fruits.

RESULTS AND DISCUSSIONS

The production capacity of a cultivar is determined by its physiological and morphological properties. From an economic point of view, the production potential is the 'Max'imum level of useful biomass that the plant can achieve under optimal growth and development conditions and free from diseases and pests. Thus, determinations were carried out that focused on the average fruit weight, the average number of fruits/plant and the average yield per plant (table 1).

The average fruit weight varied between the experimental variants and also between the varieties. The average weight of the fruit within the same variety (var. *tetragonum*) was 180.6 g for the unfertilized variant and 172.9 g for the fertilized one. It is noticed that the fertilization did not influence this indicator, a fact that is also similar for var. *longum* where the fertilized variant had lower weight fruits than those of the unfertilized variant (table 1).

The average number of fruits per plant ranged from 7.8 to 13.4 in 'Cornel' and from 8.0 to 11.4 in 'Max'. For this production indicator, the fertilization has led to an increase in the number of fruits in both varieties. The var. *tetragonum* reacted very well to fertilization and the average number of fruits was 13.4 in comparison to 11.4 fruits/plant in the var. *longum*. This increase in the number of fruits per plant is also reflected in the average yield/plant which was higher in the fertilized variants in comparison to the unfertilized variants. 'Cornel' recorded a production of 2316.9 g/plant, and 'Max' 1722.5 g/plant. The production differences between the experimental variants are distinctly significant in var. *tetragonum* and significant in var. *longum*.

The yields are similar with those reported by Singh I., and Kaur A., (2018) on an assortment of three cultivars of bell pepper with different types of green works. We can say that the production has been significantly influenced by fertilization and variety. Apahidean et al., (2010) argue that the pepper yield can also be influenced by the cultivation system (conventional or organic), and Hoza et al., (2016) asserts that the production of long pepper can also be influenced by the method of pests control (organic or chemical) or even the cultivar (Szafirowska and Elkner, 2008).

Table 1. The production characters of the bell and long peppers obtained from the experimental variants

Cultivars	Average fruit (g)	Average number of fruits/ plant	Average yield/plant (g)
'Cornel' controls	180.6a	7.8b	1408.7c
'Cornel' fertilized	172.9b	13.4a	2316.9a
'Max' controls	162.9c	8.0b	1303.2c
'Max' fertilized	151,1d	11,4ab	1722,5b
LSD 5%	7.36	4.17	201.02

The biochemical determinations were conducted on samples of fruit harvested at physiological maturity, when they were red and represent average values of repetitions within the studied variants (table 2 and 3). These determinations, which relate to the physico-chemical properties of the analysed products (e.g. tomato paste and juice) contribute to the determination of the nutritional value demanded by consumers (Căpruciu and Lascu, 2018).

The quality of the fruit taste is largely determined by the sugar content and the dry matter and soluble solids.

The dry matter ranged from 7.4 to 8.5% at 'Cornel' and from 8.8 to 11.8% at 'Max', and the one in reducing sugars from 4.65 to 5.24% at 'Cornel' and from 5.12 to 5.83%.

The soluble solids increased in the fertilized variants and the reducing sugars decreased compared to the unfertilized ones. We can state that the differences in value are given by the applied fertilization scheme and the variety of peppers. Bărcanu-Tudor et al., (2018) reported a soluble solids content ranging between 4.87 and 5.52% for two new pepper varieties obtained at S.C.D.L. BUZĂU. The previous studies conducted by our team have reported a reducing sugars content of bell pepper ranging between 3.01% and 5.76% for red bell pepper (Soare et al., 2017). The obtained results in this study are similar to those reported by Cebula et al., (2015) i.e. 3.20-4.92 mg/100g fw with an average value of 4.24 mg/100g fw.

The ratio between the soluble solids content and titratable acidity (SS/TA) determined a true flavour of fruit because the relationship between soluble solids and titratable acidity is affected by environmental factors, physiological factors and cultivation technology. In our study, the applied foliar fertilization determined an improvement in the organoleptic qualities of the pepper fruits in both varieties.

The titratable acidity recorded higher values for both 'Cornel' and 'Max' cultivars in both fertilization variants. In both cultivars, the applied treatment increases the citric acid content, the increase being more pronounced for 'Cornel' cultivar (25.78%), superior to those reported by Bărcanu-Tudor et al., (2018).

Previous studies referring to the nutritional value of some pepper cultivars cultivated in southern Romania reported a content of ascorbic acid between 132 mg/100 g and 204 mg/100 g for red bell pepper (Soare et al., 2017) and between 260.48 mg/100 g and 308.00 mg/100 g for autochthonous hot pepper cultivars (Dinu et al., 2013).

The carotenoids are another class of compounds present in peppers. The carotene among the carotenoids present in peppers has pro-vitamin A activity and the capsanthin, oxygenated carotenoids, capsorubin and cryptocapsin give the intense red colour characteristic of the fruit that reaches the physiological maturity. The total carotene content ranged from 9.9 mg/100 g (unfertilized 'Max') to 12.5 mg/100 g (unfertilized 'Cornel'). The fertilization in 'Cornel' cultivar decreased the total carotenoid content to 10.4 mg/100 g as compared to 12.5 mg/100 g for the unfertilized variant while the 'Max' cultivar recorded a variation which is insignificant.

The literature shows different values for the carotenoid content of pepper fruits: 11.4 – 132 mg/100 g dw in 10 genotypes of bell pepper (Deepa et al., 2007); 690-1320 mg / 100 g dw reported in 5 varieties of red pepper (Hornero-Mendez et al., 2000); 130.6-414.1 mg / 100g dw in four varieties of *Capsicum annuum* L. cultivated in Italy (Tundis et al., 2013).

These differences are explained by the fact that different values of carotenoid content vary in composition and concentration depending on genotype, crop technology and environmental conditions, as well as the maturity stage of the fruit.

The lycopene is a valuable component of pepper fruits because it is a powerful antioxidant. In this study, the lycopene content ranged from 118.29 mg/100 g in the unfertilized 'Max' cultivar and 126.91 mg/100 g in the fertilized 'Cornel' cultivar (table 2).

Table 2. The biochemical determinations conducted on bell and long pepper fruits

Cultivars	Dry matter (%)	Reducing sugars (%)	Taste index dry matter/titratable acidity (%)	Titratable acidity (% citric acid)	Total carotenes (mg/100g dw)	Lycopene (mg/100g dw)
'Cornel' controls	7.4c	5.24a	11.93c	0.620a	12.5 a	119.45a
'Cornel' fertilized	8.5bc	4.65a	12.35c	0.688a	10.4b	126.91a
'Max' controls	8.8b	5.83a	15.63b	0.563a	9.9b	118.29a
'Max' fertilized	11.8a	5.12a	19.53a	0.604a	10.0b	121.07a
LSD 5%	1.12	1.49	1.48	0.161	1.95	16.07

There is a significant increase in the lycopene content for both cultivars as a result of the applied fertilization.

The rich content of biologically active compounds of pepper fruits is also ensured by the presence of phenols that contribute to the sensory and nutritional quality and which have beneficial effects on health.

The content of phenolic compounds increased in both cultivars after the application of foliar fertilization (table 3).

Table 3. The antioxidant activity of the bell and long pepper fruits

Cultivars	Phenolic compounds (mg/100 g dw)	Vitamin C (mg/kg fw)	Antioxidant activity (µM Trolox/1 g dw)	
			ABTS	DPPH
'Cornel' controls	131.99d	119.60c	9.04c	8.73c
'Cornel' fertilized	147.50c	150.44b	15.20a	15.70a
'Max' controls	192.85b	198.56a	11.58b	12.24b
'Max' fertilized	251.71a	201.35a	14.84a	15.36a
LSD 5%	6.65	9.95	1.106	1.34

Regarding the 'Max' cultivar, higher values of phenolic compounds were recorded in both the unfertilized variant (192.85 mg/100 g) and the fertilized variant (251.71 mg/100 g) in comparison to the 'Cornel' cultivar, which recorded 131.99 mg/100 g in unfertilized one and 147.50 mg/100 g in the fertilized version.

The vitamin C content varied from 119.60 mg/kg fw to 201.35 mg/kg fw in the two pepper varieties, representing similar results to those obtained by our team in 2015 on a study of 6 cultivars of bell and long peppers in southern Romania (Soare et al., 2017). These results may be due to the role of potassium in plant metabolism and many important, regulatory processes in the plant, sustained statement by Bassiony et al., (2010).

Our results are similar to those obtained by other authors: 116.3-190.5 mg/100 g (Cebula et al., 2015); 48.23 -192.63 mg/100 g in 10 red bell pepper cultivars (Deepa et al., 2006); 102.4-202.4 mg/100 g (Howard, 2000); 101.19 -114.85 mg/100 g fw for sweet pepper in Poland (Peruka and Materska, 2007); 177-198 mg/100 g for sweet pepper hybrid with red fruit in Egipt (Shahein et al., 2015); 183.8 - 2246.7 mg/kg for 15 varieties of pepper grown in field conditions in southern Slovakia (Valsikova et al., 2006). Increasing K^+ concentration in plant fertilization improves the quality of paprika fruit by increasing the content of TSS, soluble sugars and ascorbic acid concentration (Botella et al., 2017).

The different content depends on the variety of peppers, the conditions of the crop, the maturity stage of the fruit when the determinations are made and, of course, the environmental factors correlated with the fertilization applied to the crop.

The antioxidant activity is especially determined by the ascorbic acid, carotenes, vitamins, phenols and flavonoids and it is of great importance for this species, whose fruits are consumed a lot and fresh.

The antioxidant activity of the pepper extracts was determined by two methods: the capacity of sample to reduce the radical DPPH and the radical cation ABTS.

The antioxidant activity determined by the DPPH method varied between 9.04 μM TE/1 g fw in unfertilized 'Cornel' and 15.20 μM TE/1 g fw in fertilized 'Cornel'. The ABTS method had values ranging from 8.73 μM TE/1 g fw in unfertilized 'Cornel' to 15.70 μM TE/1 g fw in fertilized 'Cornel'. It is observed that the fertilization scheme in both cultivars applied to the pepper culture in the field determined the increase of antioxidant activity measured by both methods.

The 'Cornel' cultivar shows a significant increase of the antioxidant activity in response to fertilization, 68.14% for the ABTS method and 79.83% for the DPPH method, while for the 'Max' cultivar the increase is 28.15% and 25.49% respectively.

The chemical foliar fertilization combined with the Crop'Max' organic product, applied during vegetation, resulted in an increase in the antioxidant activity of pepper fruits, with higher values for bell pepper (var. *tetragonum*).

Dinu et al., (2018) claim that each phyto-nutrient has a unique method of accumulation in pepper fruits that depends on the genotype, the maturity stage of the fruit and the crop area.

Caruso et al., (2019) claim that the antioxidant activity assessed by the ABTS method is significantly influenced by the interaction between the research year and the pepper cultivation system.

CONCLUSIONS

The study determined that the fertilization has influenced differently the production and nutritional quality of the pepper fruits.

The fertilization mode determined significant production differences between both varieties and between fertilization variants. The 'Cornel' cultivar (var. *tetragonum*) recorded the best production of 2316.9 g/plant, in the fertilized version.

With regard to the significant increase of quality attributes, also the 'Cornel' cultivar recorded the highest values. In terms of the significant increase in quality attributes has been highlighted, the 'Cornel' cultivar, for carbohydrate, carotene and antioxidant activity, and cultivar 'Max' (var. Longum) for increasing the content of phenolic compounds and vitamin C in response to fertilization.

Considering the increased consumer demand for healthy products and the current policies targeting environmentally sustainable crop systems or fewer chemical inputs, we can state that the fertilization scheme used by us has improved the nutritional composition of the bell and long pepper fruits.

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