American Journal of Experimental Agriculture
4(4): 376-383, 2014

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Effect of Type and Method of Fertilization on Marrows (*Cucurbita pepo* L.) Yield and Fruit Quality

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Authors' contributions

Authors DK and DH designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Authors DK and DM managed the analyses of the study. Author DH managed the literature searches. All authors read and approved the final manuscript.

Original Research Article

Received 9th March 2013
Accepted 15th November 2013
Published 17th December 2013

ABSTRACT

Aims: The main aim of this study is to determine the influence of foliar and soil fertilization upon the nutritious value of fruit of marrows and to clarify the opportunity for using Blue tetrazolium chloride (BTC) as a reagent for determination of micro quantities of manganese in plant samples.
Study Design: The investigation was conducted in the period 2007–2009 in an open field condition with the marrows (*Cucurbita pepo* L.) cultivar Izobilna F_1 on the experimental field of the Agricultural University of Plovdiv, Bulgaria.
Methodology: Treatments of the experiment: 1. Control - non fertilized; 2. Foliar fertilization with 0.3% Fitona; 3. Foliar fertilization with 0.3% Hortigrow; 4. Foliar fertilization with 0.3% Humustim; 5. Soil fertilization with $N_{160}P_{160}K_{160}$; 6. Soil fertilization with $N_{160}P_{160}K_{160}$ + 0.3% Fitona; 7. Soil fertilization with $N_{160}P_{160}K_{160}$ + 0.3% Hortigrow; 8. Soil fertilization with $N_{160}P_{160}K_{160}$ + 0.3% Humustim. Total carbohydrates, dry matter, content of nitrates and manganese were determined in the fruits of marrows. Materials were collected in the phase of mass fruit production.
Results: The results show that application of foliar fertilizers Fitona, Hortigrow and Humustim increased yields of marrows in preserved and improved product quality. The

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effect occurred more strongly when foliar fertilizers were applied on a background of soil fertilization. The BTC method can be successfully applied to the determination of manganese in plant samples.
Conclusion: The use of foliar fertilizers containing different macro- and micronutrients in the period of growing marrows influenced on the content of mn in their fruit. The highest content of mn in the fruits of marrows was found after combined soil and foliar fertilization with 0.3% hortigrow the decrease in yield induces a concentration in manganese in the fruit of marrows. The trend is more clearly expressed in the mixed application.

Keywords: Blue tetrazolium chloride; fertilization; manganese; Cucurbita pepo L.

1. INTRODUCTION

Optimization of technologies for growing plants is a method to increase plant productivity and to obtain quality yields. Foliar application is considered as part of the agronomic techniques carried out during the period of cultivation as an opportunity to increase plant productivity [1-6]. In previous our publications there are data about the influence of foliar fertilizers upon the quality of fruit of marrows [7], as well as about the content of Mo in marrows [8]. Marrows are a traditional in Bulgaria early crop grown throughout the country. Interest is due to their excellent flavor and dietary qualities. In this connection it is important to trace the influence of foliar application on some components of the biological value of the fruit. In previous studies it was found correlation between foliar application and qualitative indicators of the yield [9-11].

The main aim of this study was to investigate effect of type and method of fertilization on marrows yield and fruit quality. Furthermore, we aimed at clarifying the possibility for using Blue tetrazolium chloride [12] as a reagent for determination of micro quantities of manganese in plant samples.

2. MATERIAL AND METHODS

The investigation was conducted in the period 2007–2009 under open field conditions with marrows (*Cucurbita pepo* L.) cultivar Izobilna F_1 on the experimental field of the Agricultural University of Plovdiv, Bulgaria. The soil of the field is classified as Molic Fluvisols [13]. The depth of the humus horizon is 28-30 cm. The soil is loamy (clay content from 30% to 41%). Chemically, the soil is characterized by a low content in organic matter (1.46%), pH neutral to slightly alkaline (7.17-7.37) and by the presence of large amounts of $CaCO_3$, which gives more favorable physical-chemical water and soil properties, despite the heavy mechanical composition. Nitrogen content was low (32-46 mg kg^{-1}), while there was a good stock of soluble phosphorus (P_2O_5 - 16.7-18 mg kg^{-1}) and potassium (K_2O - 67-96 mg kg^{-1}).

For the purpose of the experiment three different commercially available complete foliar fertilizers with distinctly different nutrient composition were used: Fitona (7.20% N, 5.20% K_2O, 1.5% Ca, 0.9% Mg, 0.1% Fe, 0.1% B, 0.01% Cu, 0.007% Zn, 0.01% Mn, 0.001% Mo; Fitotech Ltd., Bulgaria), Hortigrow® (20% N, 20% P_2O_5, 20% K_2O, 0.06% Fe, 0.02% Zn, 0.01% Mn, 0.01% Cu, 0.02% B, 0.001% Mo and 1% amino acids; Hortiland Ltd., Nederland), Humustim (on base of potassium humates-3% N, 1.14% P_2O_5, 7.83% K_2O, 3.92% Ca, 1.1% Mg, 0,001% Cu, 0.01% Zn, 0.001% Mo, 0.004% Mn 0.001% Co, B and S; Agrospeis Ltd., Bulgaria).

Soil fertilization was carried out with NPK using a ratio $N_{160}P_{160}K_{160}$. Quantity of fertilizers was 160 kg ha^{-1} like active substance. Phosphorus ($Ca(H_2PO_4)_2$ - 46% P_2O_5) and potassium (K_2SO_4 - 50% K_2O). Fertilizers were applied with last tillage of soil before planting. Nitrogen fertilizer, introduced as NH_4NO_3 (34% N), was applied twice during the growing season. First application was after formation of new leaves of plants and the second - 20 days after the first.

Water solution of foliar fertilizers was prepared. For the purpose of the experiment were used by the manufacturer recommended concentrations of use. Foliar fertilizers were applied in the below given concentrations three times in the following phases - beginning of flowering, beginning of fruit production and mass fruit production. Control plants were treated with pure water. The consumption of working solution in the first spraying was 600 l ha^{-1} and in the second and third was 800 l ha^{-1}.

2.1 Treatments of the Experiment

1. Control - non fertilized; 2. Foliar fertilization with 0.3% Fitona; 3. Foliar fertilization with 0.3% Hortigrow; 4. Foliar fertilization with 0.3% Humustim; 5. Soil fertilization with $N_{160}P_{160}K_{160}$; 6. Soil fertilization with $N_{160}P_{160}K_{160}$ + 0.3% Fitona; 7. Soil fertilization with $N_{160}P_{160}K_{160}$ + 0.3% Hortigrow; 8. Soil fertilization with $N_{160}P_{160}K_{160}$ +0.3% Humustim.

Plants were cultivated according to the conventional technology for early field production of marrows, using previously produced seedlings [14]. The seedling was planted after thirty days cultivation in non-heated polythene tunnel. Plants were planted on bed-furrow surface, according to scheme 100+60/50 cm and density of plantation 25000 plants per ha in beginning the May. Growth period was 45 days after planting.

Total carbohydrates were determined using the method of Haygedorn-Jensen [15]. Dry matter was determined according to the method of Stanchev et al. [15]. The content of nitrates in fruits was determined by test method GOST 29270-95 [16]. Manganese was determined in the fruits of marrows according to method described by Kostova and Mehandjiev [12]. Materials were collected in the phase of mass fruit production.

2.2 Determination of Manganese (VII) with BTC in Plant Samples

An analysis for manganese content was carried out using a new extraction-spectrophotometric method with Blue tetrazolium chloride [12]. Manganese (VII) forms an ion-association complex BTC. The molar absorptivity of the ion-association complex is $4.97x10^4$ l mol^{-1} cm^{-1} at 255 nm. Beer's law is obeyed in the range of 0.1 – 1.6 μg ml^{-1}. Limit of detection (DL) is 6.86 ng ml^{-1} Mn (VII) and limit of quantitation (QL) is 22.8 ng ml^{-1} Mn (VII). The determination was carried out without preliminary separation of manganese. One gram of the plant material was reduced to ashes in an oven in 450 – 500°C. The dry residue was dissolved in a dilute hydrochloric acid (1:1) solution with water. It was transferred into a volumetric flask of 50 ml and diluted to the mark with distilled water. Aliquot parts of this solution were taken for analysis. 5 ml of a sample solution was transferred into 100 ml separatory funnel. It was added 2 ml of Blue tetrazolium chloride $1x10^{-4}$ mol l^{-1}, 0.5 ml of sulphuric acid 2 mol l^{-1}. The resulting solution was diluted with distilled water to a total volume of 10 ml. Then 3 ml of 1, 2-dichloroethane was added and the funnels were shaken for 15 s. The organic layer was transferred through a dry filter paper into a 1 cm cuvette and the absorbance was measured at 255 nm against a reagent blank using a

Spectrophotometer UV-VIS (Germany) [12]. The obtained results were compared with those of the AAS (Atomic absorption spectrometry) method using a Flame Atomic Absorption Spectrophotometer “Perkin Elmer” (Germany).

2.3 Statistical Analysis

The results from the determination of total carbohydrates and dry matter were elaborated using the dispersion analysis method for one factor field trial [17], using the program BIOSTAT (ANOVA). Relative Standard Deviation about Mn was determined according to the BTC method.

3. RESULTS AND DISCUSSION

The dry matter in the fruit of marrows is almost the same after fertilization with Fitona, Hortigrow, (Hortigrow + soil fertilization), Humustim, (Humustim + soil fertilization) and control (Table 1). An exception can be discerned in fertilization with (Fitona + soil fertilization) and soil fertilization $N_{160}P_{160}K_{160}$.

Table 1. Content of manganese, dry matter and yield of the fruit of marrows in foliar and mixed fertilization

No	Variants	Mn, mg kg^{-1} BTC method	*RSD BTC method	**Dry matter, %	Mn mg kg^{-1} AAS method	***Yield kg ha^{-1}
1	Control - non fertilized	9.67	1.9	6.02	9.80	26210
2	0.3 % Fitona	8.32	1.5	6.97	8.00	30840
3	*0.3 % Hortigrow*	9.70	1.2	6.18	9.55	32430
4	0.3 % Humustim	8.69	1.7	6.45	8.80	39170
5	$N_{160}P_{160}K_{160}$	8.48	2.0	4.80	8.65	38270
6	$N_{160}P_{160}K_{160}$ + 0.3% Fitona	8.63	1.4	4.41	8.55	39270
7	$N_{160}P_{160}K_{160}$ + 0.3% Hortigrow	10.10	1.9	5.94	10.30	40840
8	$N_{160}P_{160}K_{160}$ + 0.3% Humustim	8.85	1.7	5.70	8.67	49450

** Relative Standard Deviation for BTC method (n = 5)*
*** Dry matter, % - LSD 0.064, P=5%, LSD 0.085, P=1%; LSD 0.110, P=0.1%; ***Yield kg ha^{-1}- LSD 121.378, P=5%, LSD 163.059, P=1%; LSD 214.425, P=0.1%; AAS - Atomic absorption spectrometry, BTC - Blue tetrazolium chloride*

The change of the dry matter content in the fruits may be due to the increased activity in the production of biomass and their priority allocation to the product portion and in direct connection with the increase in the weight of the fruit. These results are in line with studies of the effect of foliar fertilization on the dynamics of accumulation of dry matter by Irannejad et al. [18] in fruits of marrows.

Manganese is an important nutritious element for plants that has an influence on the quality of yield and production. In this respect the influence of manganese upon the yield of marrows in the two kinds of fertilization (foliar fertilization and mixed fertilization) is represented on Table 1. The experimental data show that the highest content of Mn can be seen in foliar fertilization with Hortigrow and mixed fertilization $N_{160}P_{160}K_{160}$ + Hortigrow. The researches showed that in the control (non fertilized), the content of manganese is lower as in foliar fertilization and also in mixed fertilization.

The content of manganese in the fruit of marrows is highest after fertilization with Hortigrow - foliar fertilization and mixed fertilization (Table 1.) Hortigrow comprises amino acids that stimulate growth, development and assimilation of nutritious substances including microelements. Content of amino acids of is the main difference between Hortigrow and Fitona responsible for the differences detected. The distinction in the content of manganese in the two treatments of fertilization is 0.40 mg kg^{-1}. Hence, the additional introduction of N, P and K in soil has small influence on the accumulation of manganese in the fruit of marrows.

The interaction between soil and foliar fertilization increased plant productivity. Early and total yield was the highest after simultaneous soil fertilization with $N_{160}P_{160}K_{160}$ and foliar application with 0.3% Humustim. Therefore appropriate from agronomic and economic point of view is combination of soil fertilization and foliar application of 0.3% Hortigrow and 0.3% Fitona (Table 1). The highest yield is obtained in fertilization with Humustim (mixed fertilization and foliar fertilization). This may be due to the fact that in the composition of the organic foliar fertilizer Humustim the basic nutritious elements are humic acids that exhibit hormone activity which stimulates growth and development. In independent foliar fertilization the highest yield 39170 kg ha^{-1} is obtained in the content of Mn 8.69 mg kg^{-1} in the fruit of marrows. In mixed fertilization the highest yield 49450 kg ha^{-1} is obtained in the content of Mn 8.85 mg kg^{-1} in the fruit of marrows. The experimental data show that in mixed fertilization, yield is with 10000 kg ha^{-1} higher than that in foliar fertilization. It makes an impression that the content of manganese is almost equal for the two ways of fertilization in which the highest yield is obtained. Consequently the main factor for higher yield is the different fertilization and not the content of manganese.

In a recent review, [6] it was stated that the foliar uptake is due to the ability of leaves and stems to absorb the nutritious elements taken in the form of certain solutions. The effect of the foliar nutrition takes place much promptly in comparison to the soil fertilization, which reduces the risks of physiological diseases of plants caused by deficiency or scarcity of a given element [5].

In the same treatments of fertilization the content of total carbohydrates in the fruit of marrows has been determined. The content of total carbohydrates is the highest in plants receiving soil and foliar fertilization ($N_{160}P_{160}K_{160}$ + Hortigrow). The experimental data (Fig. 1) show that the content of total carbohydrates is the highest (2.39 %) after mixed fertilization. After only foliar fertilization, the content of total carbohydrates is 0.78% or three times less than in mixed fertilization. The content of total carbohydrates in the control (non fertilized) is even lower (0.67%). The content of carbohydrates is the highest when we used the foliar fertilizer Hortigrow together with the soil fertilization ($N_{160}P_{160}K_{160}$), a result that paralleled that observed for the manganese content in fruits. This may be due to the higher percentage content of nitrogen, phosphorus and potassium in the leaf fertilizer Hortigrow. Results are in line with established specificities in the content of total sugars in the fruits of marrows from the research of Irannnejad et al. [18]. They were established that nitrogen and potassium fertilization influence on the content of total sugars. These conclusions are also supported by research of Sedano-Castro et al. [19]. This all indicates that the nutritious elements N, P and K that have been provided additionally have positive influence on the accumulation of carbohydrates in the fruit of marrows.

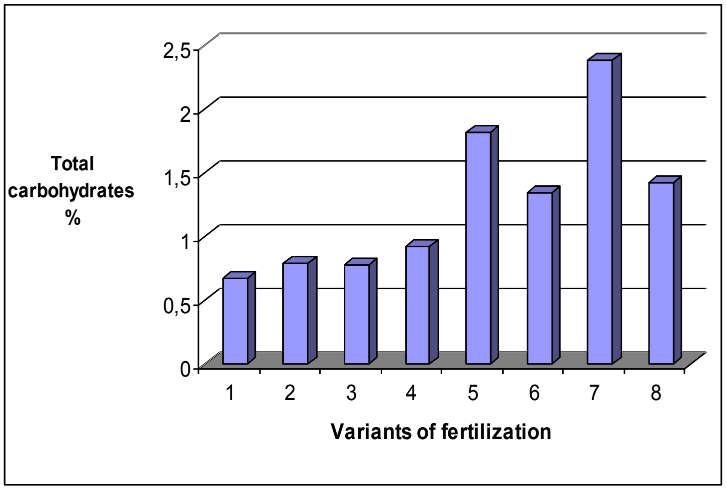

Fig. 1. Total carbohydrates in the fruit of marrows (% of dray matter). 1- Control, 2- Fitona, 3- Hortigrow, 4- Humustim, 5- Fertilization with $N_{160}P_{160}K_{160}$, 6 - $N_{160}P_{160}K_{160}$ + Fitona, 7- $N_{160}P_{160}K_{160}$ + Hortigrow, 8- $N_{160}P_{160}K_{160}$ + Humustim. Total carbohydrates % - LSD 0.079, P=5%, LSD 0.107, P=1%; LSD 0.140, P=0.1%

The influence of fertilization upon nitrate content in the fruit of marrows is presented on Fig. 2. The highest nitrate content (250 mg kg^{-1}) in the fruit can be seen in the leaf fertilizer Hortigrow, as well as in mixed fertilization $N_{160}P_{160}K_{160}$ + Hortigrow. This can be explained with higher percentage content of nitrogen in the leaf fertilizer Hortigrow.

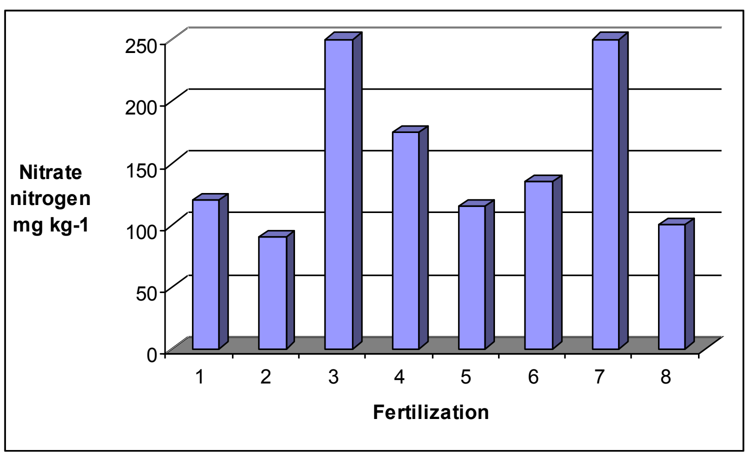

Fig. 2. Content of nitrate ($mg.kg^{-1}$) in the fruit of marrows (FW). 1- Control, 2- Fitona, 3- Hortigrow, 4- Humustim; 5- $N_{160}P_{160}K_{160}$, 6- $N_{160}P_{160}K_{160}$ + Fitona, 7- $N_{160}P_{160}K_{160}$ + Hortigrow, 8- $N_{160}P_{160}K_{160}$ + Humustim

4. CONCLUSION

The results show that application of foliar fertilizers Fitona, Hortigrow and Humustim increased yields of marrows in preserved and improved product quality. More strongly when it was applied on a background of soil fertilization.

The use of foliar fertilizers containing different macro- and micronutrients in the period of growing marrows influenced on the content of Mn in their fruit. The highest content of Mn in the fruits of marrows was found after combined soil ($N_{160}P_{160}K_{160}$) and foliar (0.3% Hortigrow) fertilization. The decrease in yield induces a concentration in manganese in the fruit of marrows. The trend is more clearly expressed in the mixed application. The BTC method can be successfully applied to the determination of manganese in plant samples.

ACKNOWLEDGEMENTS

The authors would like to thank the Research Fund of the Agricultural University for financial support.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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