

Original Research

The Effect of Mycorrhizal Fungal Strains on the Concentrations of Phosphorus and Selected Micronutrients in Tomato Fruit (*Lycopersicon esculentum* Mill.)

Joanna Majkowska-Gadomska^{1*}, Artur Dobrowolski¹,
Krzysztof K. Jadwisieńczyk², Anna Francke¹

¹Faculty of Agriculture and Forestry, Department of Agroecosystems and Horticulture,
University of Warmia and Mazury in Olsztyn, Prawocheńskiego 21, 10-957 Olsztyn, Poland

²Faculty of Technical Science, Department of Heavy Duty Machines and Research Methodology,
University of Warmia and Mazury in Olsztyn Poland

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Abstract

The research hypothesis postulates that mycorrhizal fungal strains can increase the content of phosphorus (P) and selected micronutrients in tomato fruit.

The aim of this study was to determine the suitability of three tomato cultivars for growing in a heated plastic tunnel, and to evaluate the influence of fungal strains applied to the rhizosphere of tomato plants on the concentrations of selected minerals, in particular P, iron (Fe), manganese (Mn), copper (Cu) and zinc (Zn), in the fruit. A two-factorial experiment was conducted in 2015-2016. Tomatoes were grown in coco coir in a heated plastic tunnel in the Experimental Garden of the University of Warmia and Mazury in Olsztyn. The first experimental factor were three tomato cultivars, 'Growdena F₁', 'Listell F₁' and 'Torero F₁', suitable for growing in a controlled environment. The second experimental factor was growing tomatoes in coco coir mats with and without fungal strains: control treatment (without fungal strains), growth medium inoculated with fungi of the genus *Glomus*, growth medium inoculated with a combination of fungi of the genus *Trichoderma*. The experiment had a split-block design, with 10 plants per replicate in each treatment. In order to determine the mineral content of tomato fruit, plant material was dried at a temperature of 65°C and ground in an electric mill. The samples were analyzed in the Chemical and Agricultural Research Laboratory in Olsztyn to determine their content of P, Fe, Cu, Mn and Zn. The analyzed fungal strains did contribute to a significant increase in P concentrations in tomato fruit, compared with the control treatment. Fungi of the genera *Glomus* and *Trichoderma* spp. increased the Fe content of tomato fruit by 12.5%

*e-mail: majkowska-gadomska@uwm.edu.pl

and 16.9%, respectively, compared with the control treatment, and *Glomus* fungi increased also Mn concentration in tomato fruit.

Keywords: mycorrhiza, fungi, *Glomus*, *Trichoderma* ssp., minerals

Introduction

In the past decade, coco coir, a promising and environmentally friendly growth medium, has become an increasingly popular substrate for growing tomatoes under cover. Cultivation in coco coir is similar to that in rockwool. Coir substrates have excellent physical properties (high water holding capacity and high porosity of approximately 30%) [1-5].

During cultivation on horticultural substrates, particular attention should be paid to protecting the root system against diseases as well as to strategies for increasing the yield and improving the nutritional value of tomatoes with the use of biostimulants based on e.g. marine algae extracts or live microorganisms. One of the options are mycorrhizal inocula based on association between the roots or other organs and seeds of vascular plants and fungi, referred to as a mycorrhiza. This symbiosis provides benefits to both species because they can share nutrient resources. Plants get easier access to water, mineral salts and growth-regulating substances. In turn, fungi can use glucose produced during photosynthesis. Moreover, mycorrhizas help protect plants against pathogens of the genera *Verticillium*, *Fusarium*, *Pythium* and *Rhizoctonia* [6-8]. Most studies conducted to date have investigated mycorrhizas present in ornamental, crops, trees and shrubs and orchard plants, whereas mycorrhizal associations in vegetable and food crops remain insufficiently researched [9-11]. Tomatoes (*Lycopersicon esculentum* Mill.) are widely grown and consumed in many countries around the world. The high quality of tomato fruit is determined by a high content of biologically active compounds, which are a rich source of organic substances and mineral nutrients [6, 12-15].

In the present study, the research hypothesis postulates that mycorrhizal fungal strains can increase the content of phosphorus (P) and selected micronutrients in tomato fruit. The aim of this study was to determine the suitability of three tomato cultivars for growing in a heated plastic tunnel, and to evaluate the influence of fungal strains applied to the rhizosphere of tomato plants on the concentrations of selected minerals, in particular P, iron (Fe), manganese (Mn), copper (Cu) and zinc (Zn), in the fruit.

Materials and Methods

Experimental

A two-factorial experiment was conducted in 2015-2016. Tomatoes were grown in coco coir in a

heated plastic tunnel in the Experimental Garden of the University of Warmia and Mazury in Olsztyn. The experimental facility was covered with inflated double-layered polyethylene film, and it was equipped with lateral and roof vents, and bottom, side, top and vegetation heating as well as thermal insulation and shade screen systems. Microclimate was controlled automatically with the use of a climate control computer based on readings from a weather station installed on the roof and from sensors located inside the tunnel. After emergence, the temperature was set at 21°C during the day and 19°C at night. Tomato plants were grown in rain gutters.

The first experimental factor were three tomato cultivars, 'Growdena F₁', 'Listell F₁' and 'Torero F₁', suitable for growing in a controlled environment. The second experimental factor was growing tomatoes in coco coir mats with and without fungal strains: control treatment (without fungal strains), growth medium inoculated with fungi of the genus *Glomus*, growth medium inoculated with a combination of fungi of the genus *Trichoderma*. The experiment, was carried out in 3 replications, had a split-block design, with 10 plants per replicate in each treatment.

The experimental materials comprised tomato plants grown in the greenhouse of the Department of Horticulture, University of Warmia and Mazury in Olsztyn. The seeds of the analyzed tomato cultivars were sown each year in mid-January. The applied nutrient solution was adjusted to the plants' growth stage and weather conditions. During seedling production: pH = 5.5, EC = 2.0 mS · cm⁻¹, N -205, P - 70, K- 230, Mg -60, Ca -210 (mg · dm⁻³), growth time in the tunnel pH = 5.5, EC = 3.0 mS · cm⁻¹, N -220, P - 40, K- 270, Mg -70, Ca -220 (mg · dm⁻³). It was dispensed into the coco coir mats at 10-minute intervals at a dose of 66 ml from each dripper to ensure consistent saturation of every mat. Immediately before transplanting (2.03. 2015 and 2016), the root system of each seedling was supplied with 3 g of mycorrhizal inoculum containing the spores of *Glomus* fungi. Mycorrhizal inoculum containing *Trichoderma* spp. spores was applied at 100 ml to the root system of each seedling three times during the growing season, in the form of an aqueous suspension. *Trichoderma* spp. spores were conditioned for 24 hours before use. Each time, the spore solution was prepared by dissolving 240 g of the spore formulation in 100 dm³ of water at 30°C. The solution was applied three times during the growing season to promote the colonization of the youngest roots by the fungal spores.

Plant Materials

Tomatoes were harvested when fully ripe. During the fully ripe stage, in the first decade of August, average samples (3 kg of fruit per replicate) were collected from the marketable yield to determine chemical composition of tomato fruit. In order to determine the mineral content of the fruit, plant material was dried at a temperature of 65°C and ground in an electric mill. The samples were analyzed in the Chemical and Agricultural Research Laboratory in Olsztyn to determine their content of P were extracted with concentrated sulphuric acid, and then the content of P was measured colorimetrically with the vanadate-molybdate method (Shimadzu UV 1201V [16] and Fe, Cu, Mn and Zn – by atomic absorption spectrometry (AAS) [17] .

Statistical Analysis

The mineral content of tomato fruit was analyzed in 2015-2016, and the results were presented as means for the years of the study since only minor differences were noted. The data on the yield and nutritional value of tomato fruit were processed statistically by analysis of variance (ANOVA). The significance of differences was determined by creating multiple confidence intervals in Tukey’s test at a significance level of $\alpha = 0.05$ [18].

Results and Discussion

It is highly important to use AMF (arbuscular mycorrhizal fungi) for the production of fruit, vegetable and medicinal crops as AMF increased the accumulation of macroelements (N, P) and microelements (Zn, S, Cu, Fe and Mn) [19]. Symbiotic mycorrhizal fungi colonize the roots of plants, thus increasing the bioavailability of nutrients, including P and other minerals. The root system colonized by mycorrhizal fungi facilitates nutrient uptake, particularly in soils characterized by relatively low concentrations and solubility of mineral compounds [20-25]. Uptake of different nutrients like phosphorous (P), nitrogen (N), potassium (K), sulfur (S), copper (Cu), Calcium (Ca) and Zinc (Zn) is facilitated by the incorporation of AMF in the soil-plant system [26]. According to Higo et al. [27, 28] phosphorous is highly immobile in plants. Due to the immobile nature of P in soil, application of AMF has the potential to increase agricultural production. P fertilizer application did not have an influence on root colonization and diversity of the AMF structures in tomato. Root abundance and AMF colonization are correlated negatively. Therefore an investigation is required whether AMF communities and P fertilization develop more effective management of P in tomato. Under P deficiency, the fungus supplies P taken from the soil to the plant. Under P deficiency, the fungus supplies P taken from the soil to the plant. The mycorrhizal mycelium secretes enzymes that are

able to solubilize P present in the soil in fixed form, thus making this nutrient available to plants [29-31]. Gnekow and Marschner [30] demonstrated that in P-poor soils, apple cuttings relied on mycorrhizal P uptake and the mycorrhizal fungus increased P utilization efficiency, whereas in P-rich soils, the fungal symbiont facilitated the uptake of Zn and Cu ions. According to Kyriacou and Rouphael [32], tomatoes are a rich source of minerals. In a study by Banuelos et al. [33], mycorrhizal inoculation of tomato and carrot plants increased their resistance to nematodes, and the weight of mycorrhizal plants was higher than that of non-mycorrhizal control plants. Schroeder and Janos [34] reported that at low P availability, root inoculation with mycorrhizal fungi had a beneficial influence on DM accumulation in tomato plants and cilantro. Sylvia et al. [35] also found that in the low-P soil environment, shoot dry weight was considerably higher in inoculated tomato plants than in non-inoculated control plants.

In the present study, the P content of tomato fruit was determined by an interaction between the experimental factors (Fig. 1) and the strain of mycorrhizal fungi (Fig. 2). The inoculation with mycorrhizal fungi increase

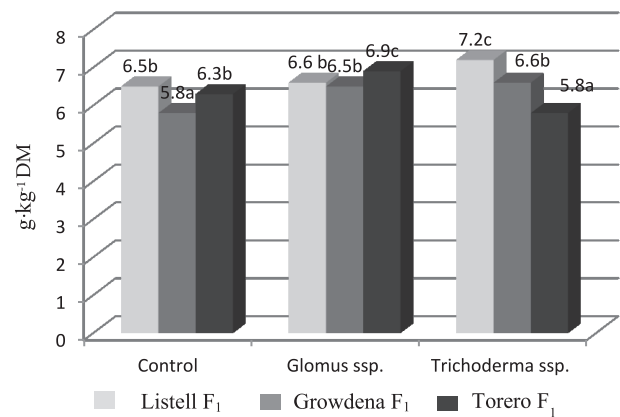


Fig. 1. Phosphorus content in the fruit of three tomato cultivars grown in coco coir mats inoculated with mycorrhizal fungal strains.

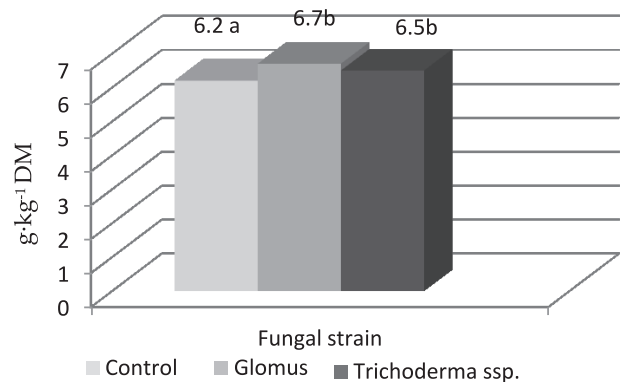


Fig. 2. Phosphorus content in the fruit of tomatoes grown in coco coir mats inoculated with mycorrhizal fungal strains.

P concentration, relative to the non-inoculated control treatment. An analysis of the interactions between the experimental factors revealed a similar trend. In tomato plants of cv. 'Listell F₁' inoculated with *Glomus* fungi, and in plants of cv. 'Torrero F₁' inoculated with *Trichoderma* ssp. fungi, the P content of fruit was 5.8 g·kg⁻¹ DM. Apart from control plants, plants of cv. 'Growdena F₁' inoculated with *Trichoderma* ssp. strains were most abundant in P. Their P concentration increased by 15.9%, relative to the treatment with the lowest P content. Tartanus [36] found that a mycorrhiza had no significant effect on the concentrations of macronutrients and micronutrients in pepper fruit, and it inconsiderably decreased the content of calcium (Ca), Fe and sodium (Na), relative to the control treatment. Mycorrhizal inoculation of the root system of peppers during seedling production and transplantation caused a minor increase in the concentrations of P, potassium (K), magnesium (Mg) and Zn in pepper fruit. Borowy et al. [37] demonstrated that a mycorrhiza had no significant effect on the content of nitrogen (N), P, K, Mg and Ca in tomato leaves, compared with non-mycorrhizal plants. In a study by Wu et al. [38], Ndung'u- Magiroi et al. [39], Thomson et al. [40] mycorrhizal fungi had a beneficial influence on the concentrations of K, Mg, Fe, Mn and Zn in the leaves and trunks, and Ca and Cu in the trunks of peach trees. In the study Fernandez et al. [41], the analyzed mycorrhizal fungi contributed to a significant increase in the Fe content of tomato fruit, compared with the control treatment. Ambroszczyk et

al. [42] investigated the effects of various biostimulants on tomato plants and found that the fruit contained 24.83 to 29.10 mg·100g⁻¹ DM P, 221.5 to 263.8 mg·100 g⁻¹ DM K, 11.63 to 20.63 mg·100g⁻¹ DM Ca, and 5.24 to 6.51 mg·100 g⁻¹ DM Mg. These values are lower than those determined in this study.

In the present experiment, the Fe content of fruit was higher in cv. 'Growdena F₁' (56.5 mg·kg⁻¹ DM) and lower in cv. 'Listell F₁' (43.5 mg·kg⁻¹ DM) (Table 1). The concentration of Zn was significantly highest in the fruit of cv. 'Growdena F₁' (13.4 mg·kg⁻¹ DM), and lowest in cv. Listell F₁' (7.6 mg·kg⁻¹ DM). In a study by Dyško et al. [43], the fruit of cv. 'Growdena F₁' had Fe content of 38.5 to 43.9 mg·kg⁻¹ DM, Cu content of 10.4 to 11.9 mg·kg⁻¹ DM, Mn content of 21.6 to 25.1 mg·kg⁻¹ DM, and Zn content of 28.3 to 34.6 mg·kg⁻¹ DM.

In the current study, the analyzed mycorrhizal fungi contributed to a significant increase in the Fe content of tomato fruit, compared with the control treatment. Fungi of the genus *Glomus* significantly increased the concentrations of Mn (21.5 mg·kg⁻¹ DM) and Zn (12.7 mg·kg⁻¹ DM) in the fruit. According to Al Mutairi et al. [44], Bhantana et al. [45] the uptake and use of Zn are improved by the application of AMF. An analysis of the interactions between the experimental factors revealed a significant increase in the Fe content of fruit in cv. 'Growdena F₁' in the control treatment and in the treatment with *Glomus* fungi, and in cv. 'Torero F₁' inoculated with *Trichoderma* ssp. fungi.

Table 1. Micronutrient content of tomato fruit depending on cultivar and the applied mycorrhizal fungal strains (mg·kg⁻¹ DM).

Cultivar (A)	Fungal strain(B)	Mean values for 2015-2016			
		Fe	Cu	Mn	Zn
'Listell F ₁ '	Control	37.8a	3.2	19.6b	9.6b
	<i>Glomus</i>	44.6a	2.8b	21.0c	7.2a
	<i>Trichoderma</i> ssp.	48.0b	2.9b	14.5a	6.0a
	Mean	43.5a	3.0a	18.4a	7.6a
'Growdena F ₁ '	Control	54.6b	1.8a	19.4b	10.8c
	<i>Glomus</i>	62.3c	2.8b	20.0b	12.8d
	<i>Trichoderma</i> ssp.	52.6b	3.8c	21.6c	16.6e
	Mean	56.5c	2.8a	20.3a	13.4c
'Torero F ₁ '	Control	40.4a	2.4a	15.4a	6.8a
	<i>Glomus</i>	44.8a	2.6a	23.6c	18.0f
	<i>Trichoderma</i> ssp.	59.4c	2.6a	17.4a	6.0a
	Mean	48.2b	2.5a	18.8a	10.3b
Mean	Control	44.3a	2.5a	18.1b	9.1a
	<i>Glomus</i>	50.6b	2.7a	21.5c	12.7b
	<i>Trichoderma</i> ssp.	53.3b	3.1a	17.8a	9.5a

Means with the same letter do not differ significantly at p<0.05 in Tukey's honest significant difference (HSD) test.

The concentration of Fe was significantly lower in the fruit of cvs. 'Listell F₁' and 'Torero F₁' in the control treatment and in the treatment with *Glomus* fungi. An interaction between the experimental factors led to a significant increase in the Cu content of fruit in cv. 'Listell F₁' in the control treatment (3.2 mg·kg⁻¹ DM) and in the treatment with *Trichoderma* ssp. fungi (2.9 mg·kg⁻¹ DM), and in the fruit of cv. 'Growdena F₁' in the treatment with *Trichoderma* ssp. fungi (3.8 mg·kg⁻¹ DM). The concentration of Mn was significantly higher in the fruit of cv. 'Torero F₁' in the treatment with *Glomus* fungi (23.6 mg·kg⁻¹ DM), cv. 'Growdena F₁' in the treatment with *Trichoderma* ssp. fungi (21.6 mg·kg⁻¹ DM) and cv. 'Listell F₁' in the treatment with *Glomus* fungi (21.0 mg·kg⁻¹ DM). According to Fernandez [41] regardless of the dose applied, inoculation with AMF led to significant increases in almost all leaf macro- and micro-nutrient concentrations compared to the control treatment. An interaction between the experimental factors induced a significant increase in the Zn content of fruit in cv. 'Torero F₁' F₁' in the treatment with *Glomus* fungi (18.0 mg·kg⁻¹ DM) and in cv. 'Growdena F₁' in the treatment with 'Listell F₁' *Trichoderma* ssp. strains (16.6 mg·kg⁻¹ DM). The Zn content of fruit was significantly lower in tomato plants of cv. 'Listell F₁' inoculated with mycorrhizal fungi, and in cv. 'Torero F₁' in the control treatment (6.8 mg·kg⁻¹ DM) and in the treatment with *Trichoderma* ssp. fungi (6.0 mg·kg⁻¹ DM).

Conclusions

The analyzed fungal strains did contribute to a significant increase in P concentrations in tomato fruit, compared with the control treatment.

Fungi of the genera *Glomus* and *Trichoderma* increased the Fe content of tomato fruit by 12.5% and 16.9%, respectively, compared with the control treatment, and *Glomus* fungi increased also Mn concentration in tomato fruit.

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Conflict of Interest

The authors declare no conflict of interest.

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