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THE EFFECT OF MYCORRHIZAL INOCULUM ON THE LEAF GREENNESS INDEX AND YIELD OF TOMATO (*LYCOPERSICON ESCULENTUM* MILL.) PLANTS GROWN IN A HEATED PLASTIC TUNNEL

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Abstract. In an experiment conducted in 2012-2013, tomatoes were grown in coco coir inoculated with mycorrhizal fungi, in a heated plastic tunnel in the garden of the Research and Experimental Station of the University of Warmia and Mazury in Olsztyn. The experimental materials comprised seedlings of three tomato cultivars, 'Torero F₁', 'Growdena F₁' and 'Listell F₁', supplied by a horticultural farm. The second experimental factor was inoculation of the plant growth medium with endomycorrhizal fungi from the genus Glomus. The aim of this study was to determine the chlorophyll content of leaves of three tomato cultivars grown in coco coir inoculated with mycorrhizal fungi. Each treatment consisted of seven coco coir mats (100 x 20 x 7.5 cm) with four tomato plants grown in each mat, pruned to produce 23 clusters. The experiment was carried out in triplicate. Seedlings, planted in rockwool cubes, were initially placed next to the openings made in the coir mats. Immediately before planting, each seedling was supplied with 10 ml of a standard working solution containing mycorrhizal fungi. The mycorrhizal inoculum was applied directly to the coco coir. Mycorrhizal fungi were not used in the control treatment. The results showed that, in most treatments, SPAD readings recorded in the fall were lower than in the summer, and the noted values were similar to those determined at the beginning of the growing season. The mycorrhizal inoculum improved the nutritional status of tomato plants determined based on the leaf greenness index. The mycorrhizal inoculum increased the yields of all analysed tomato cultivars.

Keywords: chlorophyll content, coco coir, fertigation, heated tunnel, tomato leaves, mycorrhiza

INTRODUCTION

Tomatoes (*Lycoperscum esculentum* Mill.) are widely produced and consumed in many countries around the world. The popularity of tomatoes can be attributed to their high biological value, unique flavour and dietary versatility (Kmiecik and Kobus 2005, Winiarska and Kołota 2007, Hasanuzzaman *et al.* 2015). The highest yields can be achieved when tomatoes are grown in a controlled environment in rockwool mats. Coco coir provides a viable alternative to rockwool (Grillas *et al.* 2001, Carrijo *et al.* 2004, Kobryń *et al.* 2007, Chohura 2007, Wysocka-Owczarek 2010, Kleiber 2014).

The root system of tomatoes grown in horticultural substrates has to be protected against pathogens such as *Verticillium*, *Fusarium*, *Phytium* and *Rhizoctonia*. Attempts are also made at increasing the yield potential of plants. Inocula containing spores of mycorrhizal fungi can be introduced into the substrate to develop, under controlled conditions, a symbiotic association known as mycorrhiza (Badura 2006, Ghazi and Al-Karaki 2006, Rumbos *et al.* 2006, Sumorok *et al.* 2011). Hyphae supply mineral nutrients (mostly phosphorus and nitrogen, but also zinc and copper) to plants, and protect them against pathogens (Fritz *et al.* 2006, Kubiak 2007).

The nutritional status of plants, including nitrogen supply, is an important consideration in crop production. The nitrogen content of the indicator parts of plants is usually estimated with the use of the following methods: N-tester, NNI, total nitrogen test and SPAD test. Their advantages include non-invasive measurements, rapid analysis taking a few seconds, and the use of portable analytical instruments. The SPAD (Soil-Plant Analysis Development) chlorophyll meter is a portable non-destructive device, widely used for measuring leaf greenness, i.e. the chlorophyll content of leaves (Spanner *et al.* 2005, Samborski *et al.* 2006, Pacewicz, Gregorczyk 2009, Borowski and Nurzyński 2012).

The objective of this study was to determine the effect of a mycorrhizal inoculum on the leaf greenness index and on the yield of tomato plants grown in a heated plastic tunnel.

MATERIALS AND METHODS

In an experiment conducted in 2012-2013, tomatoes were grown in coco coir inoculated with mycorrhizal fungi, in a heated plastic tunnel in the garden of the Research and Experimental Station of the University of Warmia and Mazury in Olsztyn. The experimental materials comprised seedlings of three tomato cultivars, 'Torero F₁', 'Growdena F₁' and 'Listell F₁', supplied by a horticultural farm. The second experimental factor was inoculation of the plant growth medium with endomycorrhizal fungi from the genus *Glomus*, to increase the surface area of the roots available for absorption. Each year, towards the end of February, treatment temperature was adjusted to the requirements of tomato plants and coco coir mats were saturated with a nutrient solution whose chemical composition was determined based on the chemical composition of water and the nutrient requirements of tomato plants. The nutrient solution contained (per dm³): N – 210 mg, P – 40 mg,

K - 320 mg, Mg - 60 mg, Ca - 240 mg, Fe - 1.8 mg, Mn - 0.55 mg, B - 0.33 mg, Cu -0.05 mg, Zn -0.38 mg and Mo -0.05 mg. During the experiment, the composition of the nutrient solution was modified to account for changing weather conditions, the growth stages and nutrient requirements of tomato plants. Each treatment consisted of seven coco coir mats (100 x 20 x 7.5 cm) with four tomato plants grown in each mat, pruned to produce 23 clusters. The experiment was carried out in triplicate. Seedlings, planted in rockwool cubes, were initially placed next to the openings made in the coir mats. Immediately before planting, each seedling was supplied with 10 ml of a standard working solution containing mycorrhizal fungi. The mycorrhizal inoculum was applied directly to the coco coir. Mycorrhizal fungi were not used in the control treatment. Seedlings were inserted into the mats when flowers emerged on 50% of the plants. In the first 4-6 weeks, tomatoes were fertilised with a starter solution with electricity connectivity (EC) of 3.2-3.4 mS cm⁻¹. During that time, the EC of the mat reached approximately 4.5 mS cm⁻¹. In successive weeks, plants were supplied with a standard nutrient solution whose EC was adapted to the weather conditions (2.7-2.9 mS cm⁻¹ on sunny days and 3.0-3.3 mS cm⁻¹ on overcast days). Runoff was maintained at 10-30%, depending on weather conditions.

Throughout the growing season, the leaf greenness index was determined with the Konica Minolta SPAD 502 Plus chlorophyll meter, twice in every month at three heights of the plant (in each treatment, 10 lower, middle and upper leaves were selected randomly from three plants). The results of the measurements were averaged.

The results of the study were analysed statistically. The significance of differences was determined by creating confidence intervals in Tukey's test at a significance level of $\alpha = 0.05$. All calculations were performed in the STATISTICA 12 program.

RESULTS AND DISCUSSION

Tomatoes are grown in greenhouses or heated plastic tunnels until 20-23 clusters reach typical form and size, usually from the beginning of March to the end of October. The tomato growing season is characterised by different sunlight conditions. Tomato plants suffer from insufficient light exposure until mid-April, the highest photosynthetic efficiency is observed until the end of August, and in the last two months days become shorter and plants receive less sunlight (Wysocka-Owczarek 2010). Mycorrhizal fungi are applied to the roots of tomato plants to improve growing conditions and to stimulate plant growth. Such a symbiotic relationship between a fungus and the host plant's roots provides benefits to both species which can exchange nutrients and other substances. The hyphae of endomycorrhizal fungi penetrate the root tissue without suppressing root hair development (Gianinazzi *et al.* 2010, Sumorok *et al.* 2011).

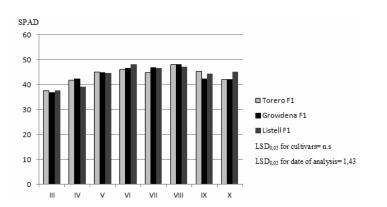


Fig. 1. SPAD readings in the leaves of the analysed tomato cultivars regardless of the mycorrhizal inoculums

Analyzing the SPAD, regardless of the cultivars in different months it was stated its lowest level in March, and the highest in August. An analysis of leaves of the three tomato cultivars revealed that cv. 'Torero F_1 ' was characterised by SPAD values of 37.75 to 49.56 units. In cv. 'Growdena F_1 ', SPAD values were determined at 36.91 units in March to 48.21 units in August. The leaf greenness index of tomato plants of cv. 'Listell F_1 ' varied from 37.21 to 48.94 SPAD units in the growing season (Fig. 1). In the leaves of cv. 'Torero F_1 ' higher values were noted in mycorrhizal inoculation treatments, as compared with the control treatment where those values ranged from 36.91 SPAD units in March to 47.28 SPAD units in July (Fig. 2). The interaction between cv. 'Torero F_1 ' and the mycorrhizal inoculum was not statistically significant, but a tendency towards an increase in chlorophyll content was observed in May and June, followed by a decrease in July and an increase in August.

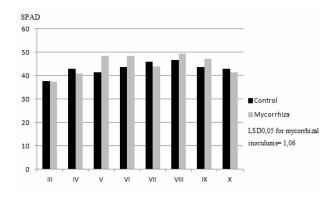


Fig. 2. The effect of the mycorrhizal inoculum on SPAD readings in the leaves of cv. 'Torero F_1 '

An analysis of the effect exerted by the mycorrhizal inoculum indicated that the leaf greenness index of tomato plants of cv. 'Growdena F_1 ' was greater in May, June, August and September ranged from 36.91 to 48.21 SPAD units in both control and mycorrhizal inoculum treatments (Fig. 3).

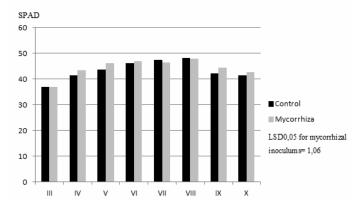


Fig. 3. The effect of the mycorrhizal inoculum on SPAD readings in the leaves of cv. 'Growdena F_1 '

Statistical analysis revealed that cv. 'Listell F_1 ' was characterised by the highest SPAD value in June (48.26 units). In the group of the analysed tomato cultivars, plants of cv. 'Listell F_1 ' had the highest average SPAD value (44.14 units), but the differences between mean values in the treatments were not statistically significant.

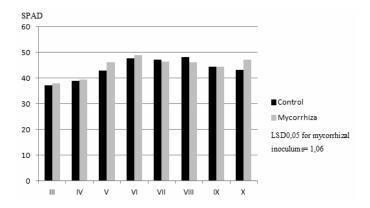


Fig. 4. The effect of the mycorrhizal inoculum on SPAD readings in the leaves of cv. 'Listell F₁'

Chlorophyll content of leaves of cv. 'Listell F_1 ' varied depending on the presence or absence of the mycorrhizal inoculum (Fig. 4). In the control treatment, the

highest SPAD value was noted in August (48.28 units) and the lowest – in March (37.21 units). In the mycorrhizal inoculum treatment, the lowest SPAD value was observed in March (37.95 units) and the highest – in June (48.94 units). Tomato plants of cv. 'Listell F_1 ' inoculated with mycorrhizal fungi were characterised by significantly higher SPAD values in the fall, relative to the other cultivars. In October, the measurement reached 47.02 SPAD units in cv. 'Listell F_1 ', whereas in the remaining cultivars the SPAD value did not exceed 43 units.

Chlorophyll content of leaves varied depending on the date of analysis. The leaf greenness index increased from the moment when tomato seedlings were planted in mat openings to reach the highest level in the summer, at the end of July and at the beginning of August. A similar trend was observed by Olszewska (2003), and Pacewicz and Gregorczyk (2009) who found that SPAD values were also affected by other factors such as the age of plants, the height at which the measurement was taken and nitrogen fertilisation. The mycorrhizal inoculum contributed to an increase in SPAD values in cv. 'Torero F₁' and 'Growdena F₁', which could result from the fact that hyphae formed a mantle covering the roots, thus facilitating nutrient uptake by tomato plants (Kubiak 2007).

According to Kołota (2004) and Ortuzar-Iragorri *et al.* (2005), the measurement of the chlorophyll content of leaves with the SPAD meter is a relatively stable and reliable method. In the present experiment, SPAD readings recorded in the fall (Figs 1-4) were lower than in the summer in most treatments, and the noted values were similar to those determined at the beginning of the growing season. A similar distribution of SPAD values was reported by other authors (Olszewska 2003, Liszewski 2008). This is due to the fact that in the spring, plants utilise nutrients more effectively for metabolic processes and tissue growth. Nitrogen is essential for yield formation, and it is taken up in the largest amounts by plants, particularly in periods with high temperatures and high solar radiation (Wysocka-Owczarek 2010).

An evaluation of the chlorophyll content of leaves contributes to optimising the nutritional status of plants through fertilisation, and to improving crop yield. Attention should also be paid to the response of plants to mycorrhizal inocula. Tomato yields in all the treatments are presented in Table 1. It was found that the mycorrhizal inoculum had a significant effect on the yields of all analysed tomato cultivars. In cv. 'Listell F₁', the yield attained in the mycorrhizal inoculum treatment was higher by 8.70 kg m⁻² than in the control treatment. The total yield of tomato plants of cv. 'Growdena F₁' was higher by 1.12 kg m⁻² in the mycorrhizal inoculum treatment than in the control treatment.

Table 1. The effect of the mycorrhizal inoculum on the total yield of three tomato cultivars grown in a heated plastic tunnel (mean values of 2012-2013)

Cultivar	Treatment	Total yield (kg m ⁻²)
'Listel F ₁ '	Control	23.10
	Mycorrhiza	31.80
	Mean	27.45
'Growdena F ₁ '	Control	27.42
	Mycorrhiza	28.54
	Mean	27.98
'Torero F1'	Control	33.90
	Mycorrhiza	35.68
	Mean	34.79
Mean treatment	Control	28.14
	Mycorrhiza	32.01
LSD $\alpha = 0.05$		
Cultivar (a)		3.49
Treatment (b)		n.s.
Interaction (axb)		0.18

CONCLUSIONS

1. In most treatments, SPAD readings recorded in the fall were lower than in the summer, and the noted values were similar to those determined at the beginning of the growing season.

2. The mycorrhizal inoculum improved the nutritional status of tomato plants determined based on the leaf greenness index.

3. The mycorrhizal inoculum increased the yields of all analysed tomato cultivars.

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WPŁYW STOSOWANIA SZCZEPIONKI MIKORYZOWEJ NA INDEKS ZAZIELENIENIA LIŚCI ORAZ PLON ROŚLIN POMIDORA (*LYCOPERSICON ESCULENTUM* MILL.) UPRAWIANEGO W OGRZEWANYM TUNELU FOLIOWYM

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Streszczenie. W latach 2012-2013 w ogrzewanym tunelu foliowym zlokalizowanym na terenie Zakładu Dydaktyczno-Doświadczalnego Uniwersytetu Warmińsko-Mazurskiego w Olsztynie zostało przeprowadzone doświadczenie z uprawą pomidora. Prowadzona była ona na matach z włókna kokosowego z zastosowaniem preparatu posiadającego w swoim składzie szczep grzybów mikoryzowych. Materiał badawczy stanowiły rośliny trzech odmian pomidora, 'Torero F1', 'Growdena F1' oraz 'Listell F1', pozyskane z gospodarstwa ogrodniczego specjalizującego się w produkcji rozsady warzyw. Drugi czynnik badań stanowiła zastosowana szczepionka mikoryzowa zawierająca szczepy grzybów endomikoryzowych z rodzaju Glomus. Celem badań była ocena poziomu chlorofilu w liściach trzech odmian pomidora uprawianego na matach kokosowych z dodatkiem szczepionki mikoryzowej. Jeden obiekt badawczy stanowiło 7 mat z włókna kokosowego o wymiarach 100 x 20 x 7,5 cm z czterema roślinami prowadzonymi na 23 grona. Doświadczenie przeprowadzone zostało w trzech powtórzeniach. Rozsada przygotowana w kostkach z welny mineralnej początkowo ustawiana była obok otworów przygotowanych do uprawy w matach. Bezpośrednio przed wstawieniem rozsady na miejsce stałe zastosowano preparat mikoryzowy w dawce 10 ml roztworu roboczego pod każdą roślinę. Preparat użyto bezpośrednio na podłoże kokosowe, na które ustawiano rośliny. W obiekcie kontrolnym szczepionki mikoryzowej nie zastosowano. W wyniku przeprowadzonych badań stwierdzono, że jesienne wyniki pomiarów w badanych wariantach uprawy były niższe, aniżeli w okresie letnim, i osiągały wartości zbliżone do wyników otrzymywanych w początkowych okresach uprawy. Stosowanie preparatu mikoryzowego wpłynęło na poprawę stanu odżywienia roślin określanego za pomocą indeksu zazielenienia liści. Stosowanie szczepionki mikoryzowej wpłynęło korzystnie na plonowanie wszystkich badanych odmian pomidora.

Słowa kluczowe: zawartość chlorofilu, włókno kokosowe, fertygacja, tunel ogrzewany, liście, mikoryza