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EFFECT OF THE PHOSPHORUS CONTENT IN A NUTRIENT SOLUTION ON THE EXPRESSION OF GENES ENCODING PHOSPHORUS TRANSPORTERS IN TOMATO GROWN ON DIFFERENT SUBSTRATES*

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Abstract

Effects of the phosphorus content in a nutrient solution (15 or 50 mg P dm^{-3}), growing substrate (rockwool or coconut fiber) and the plant growth stage (for roots: 71 or 113 days after transplanting DAT; for leaves: 71 or 92 DAT) on the chemical composition of roots, the phosphorus content in leaves and the expression of genes encoding proteins involved in the transport of phosphorus from the medium to the plant were investigated in tomato cv. Admiro F_1 grown in a foil tunnel. A fertigation system without recirculation was used. Regardless of the plant age and growing substrate, tomatoes fertilized with a nutrient solution containing 50 mg P dm^{-3} had more phosphorus, iron, boron and copper in roots and more phosphorus in leaves. Irrespective of the stage of plant growth and phosphorus level in the medium, the content of almost all macro- and microelements was higher in roots of plants grown in rockwool than in coconut fiber. The stage of plant growth significantly affected the mineral composition of roots as well as the P content in tomato leaves. More phosphorus was stored in roots of younger plants, whereas the phosphorus content was lower in younger than in older leaves. Our analysis of the gene expression showed that transporters encoded by *LePT1-LePT4* were involved in phosphate nutrition. Expression of the genes was generally (except *LePT4*) higher in plants treated by the solution containing 15 mg P dm^{-3} than in plants treated by 50 mg P dm^{-3}. The expression of genes *LePT2*, *LePT3* in roots of older plants (113 DAT) was generally higher than in young plants.

Key words: fertigation, gene expression, growing medium, stage of growth, transport of phosphorus in plant.

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INTRODUCTION

The greenhouse production of many vegetables is mostly based on soilless cultivation systems, where the space available for the development of roots is limited to a small volume of the substrate. Therefore, the plants need a continuous supply of a nutrient solution. An increase in the efficiency of the uptake of nutrients may allow for their lower concentrations in nutrient solution, which will reduce both costs of plant production and environmental pollution. Reduction of fertilizers can be achieved by the induction of mycorrhiza, where symbiotic fungi stimulate the development of roots, and by the creation of new cultivars with an increased ability to take up and transport macroelements (Karandshov, Bucher 2005, Al-Karaki 2006, Karagiannidis et al. 2007).

Plants require relatively large amounts of inorganic phosphate, which is often a limiting nutrient in the plant growth (Marschner 1995). Following its uptake, inorganic phosphorus (P_i) is transported to plant organs by transporters of low or high affinity. High-affinity transport is predominant at low concentrations of P, and low-affinity transport prevails at higher concentrations. The low-affinity system is apparently expressed constitutively, whereas the high-affinity system is induced under P_i deficiency (Furihata et al. 1992, Daram et al. 1998, Liu et al. 1998*a*,*b*). Three members of the subfamily of *Medicago truncatula* genes encoding MtPT1, MtPT2 and MtPT3 show low affinity for P_i, while MtPT5 shows high affinity for P_i (Liu et al. 1998*b*, Javot et al. 2007). Moreover, there are eight genes *LePT1-LePT8* known in tomato (Liu et al. 1998*a*, Balestrini et al. 2007, Chen et al. 2014).

The type of substrate may have a significant impact on the tomato yield and content of phosphorus in leaves. Chohura and Komosa (2003) showed a higher content of phosphorus in leaves of the greenhouse tomato cv. Maeva F_1 grown in expanded clay than in rockwool and polyurethane foam. Moreover, the uptake of water and nutrients depends on the stage of plant growth, light, temperature, medium pH and EC, nutrient proportion, substrate and air moisture (Marschner 1995, Heinen et al. 2003). In the cultivar Cunero F_1 tomatoes, the lowest phosphorus uptake was observed from transplanting until the flowering of 1st cluster, increasing afterwards, from the ripening of fruits on 1st cluster until the ripening of fruits on 4th and 5th cluster (Kowalska 2004).

Results of studies on the expression of P transporters in tomato grown in soil, organic substrates or hydroponics under controlled conditions are available (Balestrini et al. 2007, Daram et al. 1998), although this paper is the first attempt to evaluate effects of the phosphorus content in a nutrient solution, the substrate type and stage of plant growth as well as interactions of these factors on the expression of *LePT1-LePT5* genes in roots of tomato plants. We hypothesize that the uptake of P by plants depends on the P content in a nutrient solution as well as the type of substrate and plant growth stage, and it may affect the expression of genes encoding P transporting proteins.

The aim of this study was to determine the effect of the phosphorus content in a nutrient solution on the expression of genes encoding proteins involved in the transport of phosphorus in a tomato plant as well as on the phosphorus content in roots and leaves of tomatoes grown on different substrates. Additionally, the effect of the growth stage of a plant was considered.

MATERIAL AND METHODS

Plant culture

Tomato plants of the cultivar Admiro F_1 were hydroponically grown in cultivation rows set under a foil tunnel, in the spring to summer season (March to July 2011). A three-factorial experiment was carried out with sub-blocks differentiated by the phosphorus content (15 or 50 mg dm^{-3}, supplied as KH_2PO_4) in the medium as the first factor and two substrates (rockwool Grotop Master Dry or coconut fiber Profit Power) within each sub-block as the second factor. The samples of roots and leaves (fully developed leaf, the forth from the top) were taken for analyses in two growth stages of plants (for roots – fruit setting on the 6th cluster – 71 days after transplanting DAT, or the end of cultivation – 113 DAT; for leaves – fruit setting on the 6th cluster – 71 DAT or the harvest of fruits from the 3rd and 4th cluster – 92 DAT) and the growth stage was the third factor. The leaf samples were fully developed leaves, the forth from the top of a plant. Three whole root balls were collected from each replication of a particular variant of the experiment. The aerial part of a plant was cut down, after which one-third of the mat occupied by the plant was cut out and the roots from this part were isolated, washed in distilled water, immediately frozen in liquid nitrogen and kept at -80°C.

There were three replicates, each with 15 plants, in every treatment. The total number of plants in the experiment was 180. The density was 2.5 plants per m^2. The plants were fertigated without recirculation of the medium. Commercially available seedlings (1st stage of flowering) were planted into rockwool or coconut mats and supplied with a nutrient solution containing 15 or 50 mg P dm^{-3}, and (mg dm^{-3}): N180, K 300, Ca 180, Mg 50, Fe 1.5, Mn 0.6, Zn 0.5, B 0.33, Cu 0.1 and Mo 0.05. The same levels of macro- and microelements (except P) were in both nutrient solutions. Plants were grown in the one shoot system until the 7th cluster.

The pH of the medium was maintained at pH of 5.5-5.8. During the plant growing season, the composition of the nutrient solutions, except for P, was adjusted to the plant growth stage. The frequency of dosing a nutrient solution was adjusted to the phase of the plant growth and environmental conditions (light, temperature). An overflow of 20% was maintained.

Plant analysis

The content of macro-, microelements and Na in roots as well as the expression of genes (*LePT1-LePT4*) encoding P transporters were determined in two periods (71 or 113 DAT). Leaf samples (with stalks) were collected from the fourth fully grown leaves from the top on 71 or 92 DAT.

For determination of macro- and microelements, leaves and roots were dried at 70°C and ground in a laboratory mill (Fritsch Pulverisette 14, Germany) with a 0.5mm mesh sieve. Samples were then mineralized in 65% super-pure HNO_3 in a microwave oven (CEMMARS-5 Xpress), according to Paslawski and Migaszewski (2006). The content of macro- and microelements was determined in an ICP-OES (Prodigy TeledyneLeemanLabs, USA). All concentrations of nutrients were expressed on a dry mass basis.

For molecular analysis, samples were lyophilized (Alpha 1-2/LD, Christ, Germany) and stored at -80°C until RNA isolation. RNA was isolated from 70 mg of tomato roots by the method of Gasic et al. (2004). In all samples, DNase-treated extracts were made. RNA quantification was obtained using an Eppendorf BioPhotometer.

RT-PCR was performed with primers designed for amplifications of genes *LePT1-LePT4* (LePT1 For CATTGTTTCTGCAGCATTCAAGG, LePT1 Rev GGCTCCTTTTGCTTCAGAAATAGCTG, LePT2 For GATTCGATCACGCGTATAGATCC, LePT2 Rev GAAATTTGTTCGATTTTGGCTTCC, LePT3 For TCAATCTACTGATCCATCTAAAGTC, LePT3 Rev TAGTTTGTGCATTTTCCCCTTTAG, LePT4 For GAAGGGGAGCCATTTAATGTGG, LePT4 Rev ATCGCGGCTTGTTTAGCATTTCC) and reference gene *18S rRNA* (Le18S For AAAAGGTCGACGCGGGCT, Le18S Rev CGACAGAAGGGACGAGAC) in tomato (Balestrini et al. 2007). RT-PCR analyses, with primers for a specific tomato ribosomal gene (*18S rRNA*), were performed to check the presence of the plant's RNA. The synthesis of cDNA and RT-PCR reactions was carried out using a Reverse Transcription System Kit (Promega, USA), according to the provided protocols, in a GeneAmp PCR System 2400 (Applied Biosystems, Life Technologies Corporation, USA). In all samples, the amount of RNA was the same (1 μg). Transcripts were amplified under the following conditions: 94°C for 10 min (1 cycle), 94°C for 30 s, 30 s of annealing at the temperature of 45°C, 70°C for 30 s (35 cycles), and 70°C for 5 min (1 cycle). The reaction products were visualized on agarose gels to confirm the presence of a single band of the expected size and archived using the Molecular Imager® Gel Doc™ XR+ Imaging System (Bio Rad, USA).

The sequence of the amplifiable RT-PCR products was determined on both strands by the dideoxy method (Poland, Genomed). The Basic Local Alignment Search Tool (http://blast.ncbi.nlm.nih.gov) and CLUSTALW (http://www.genome.jp/tools/clustalw) software package were used for sequence analyses and database searches. The neighbour-joining method (Saitou, Nei 1987) and MEGA6 (Tamura et al. 2004, 2013) were used for constructing phylogenetic trees.

Data statistical analysis

The results were subjected to a three-way analysis of variance using the Anova module of Statistica 10.0 PL. All data are presented as means for main effects. The significance was declared at $P<0.05$.

RESULTS

The level of P in the medium, regardless of the substrate type and stage of plant growth, had no effect on the content of macroelements in the roots of tomato plants, with exception of phosphorus (Table 1). The reduction of the phosphorus content in the medium to 15 mg dm^{-3} caused a significant, nearly 2-fold, decrease in the P content in roots (Table 1) and leaves (Table 2). A low P content in the medium lowered the levels of most of the essential trace elements in roots of plants, except molybdenum, where an opposite relationship was observed. A decrease in the content of Mn and Zn in roots resulting from the lower P content in the medium was not significant.

Table 1

Effects of the phosphorus content in a nutrient solution, growth stage and medium on the chemical composition (macroelements and sodium) of tomato roots

Item		Nutrient (g kg^{-1} d.m.)						
		N	P	K	Ca	Mg	Na	S
Phosphorus (mg dm^{-3})	15	22.5	1.7	33.4	24.3	4.2	2.6	4.1
	50	23.3	3.1	34.6	22.7	4.1	2.6	4.5
Growing medium	rockwool	23.9	2.9	36.5	27.5	5.4	2.3	4.4
	coconut fiber	22.0	1.9	31.6	19.6	3.0	2.9	4.3
Growth stage	I*	21.5	2.8	33.2	26.5	4.6	2.6	4.4
	II	24.5	2.1	34.9	20.6	3.8	2.6	4.4
F for:								
phosphorus		ns	sd	ns	ns	ns	ns	ns
growing medium		sd	sd	sd	sd	sd	sd	ns
growth stage		sd	sd	sd	sd	sd	ns	ns

Note: * I – fruit setting on the 6rd cluster – 71 DAT; II – the end of cultivation – 113 DAT, DAT – days after transplanting, ns – non-significant differences, sd – significant differences

Roots of tomato plants grown in rockwool, regardless of the level of P in the medium and plant growth stage, contained significantly more of all macroelements, except S and Na (Table 1), as well as microelements, except Zn (Table 3), as compared to plants grown in coconut fiber. Noteworthy was the fact that the content of Fe and Mn was several times higher in the roots of plants grown in rockwool than in coconut fiber. The substrate type did not affect the P content in leaves (Table 2).

Table 2

Effects of the phosphorus content in a nutrient solution, growth stage and medium on the P content of tomato leaves

Item		P content in leaves (g kg^{-1} d.m.)
Phosphorus (mg dm^{-3})	15	3.6
	50	7.8
Growing medium	rockwool	5.7
	coconut fiber	5.7
Growth stage	I*	5.0
	II	6.4
F for:		
phosphorus		sd
growing medium		ns
growth stage		sd

Note: *I – fruit setting on the 6rd cluster – 71 DAT; II – at the harvest of fruits from the 3rd and 4th cluster – 92 DAT; ns – non-significant differences, sd – significant differences

Table 3

Effects of the phosphorus content in a nutrient solution, growth stage and medium on the chemical composition (microelements) of tomato roots

Item		Nutrient (mg kg^{-1} d.m.)					
		Fe	B	Cu	Mn	Mo	Zn
Phosphorus (mg dm^{-3})	15	1022.25	20.64	9.42	92.87	1.65	73.33
	50	1430.99	21.63	10.94	104.73	1.49	75.63
Growing medium	rockwool	2408.46	22.30	14.41	174.14	2.34	76.76
	coconut fiber	44.78	19.97	5.94	23.46	0.79	72.20
Growth stage	I	1718.08	23.13	12.44	131.85	1.55	78.2
	II	735.16	19.13	7.92	65.75	1.57	70.76
F for:							
phosphorus		sd	sd	sd	ns	sd	ns
growing medium		sd	sd	sd	sd	sd	ns
growth stage		sd	sd	sd	sd	ns	ns

Note: See Table 1

Regardless of the level of P in the medium and type of substrate, older roots (113 DAT) contained significantly more N and K than younger ones (71 DAT), but less P, Ca and Mg as well as most of the analyzed microelements with the exception of Mo and Zn (Tables 1 and 3). Conversely, the P content in leaves of older plants (92 DAT) was significantly higher than in leaves of younger plants (71 DAT) – Table 2.

Our analysis of the interaction between the experimental factors was limited to the assessment of the content of P in roots and leaves. A decrease of the P content in roots and leaves due to the reduction of the P content in the medium did not depend on the type of substrate. However, a significant decrease in the P content in roots of older plants (DAT 113) occurred only in plants treated with the medium containing 50 mg P dm^{-3} (interaction: P level in nutrient solution x stage of growth; $P < 0.05$, Figure 1).

The reduction of the P content in roots of older plants (113 DAT) was especially pronounced in plants grown in rockwool (interaction: substrate type x stage of growth; $P < 0.05$, Figure 2).

A higher content of P in leaves of older plants (92 DAT) was found only in plants grown in the medium with the higher content of P (interaction: P level in the nutrient solution x stage of growth; $P < 0.05$, Figure 3).

No significant interaction between the level of P in a nutrient solution and the type of substrate or the stage of plant growth on the P content was observed either in roots or in leaves ($P > 0.05$).

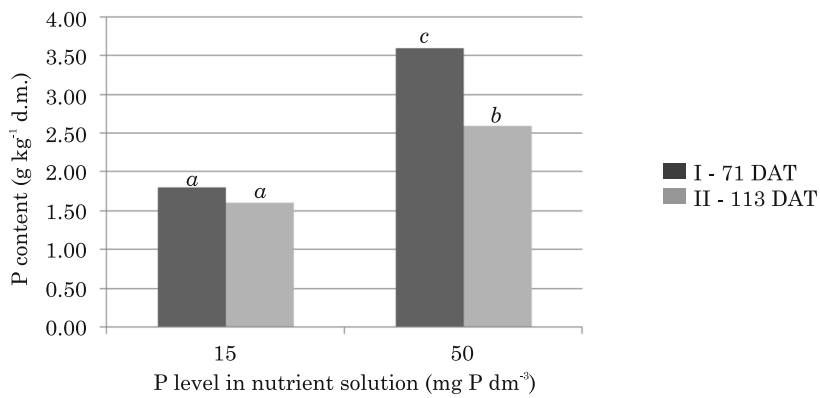

Fig. 1. The content of P in roots of tomato plants; interaction: P level in nutrient solution x phase of growth; $P < 0.05$

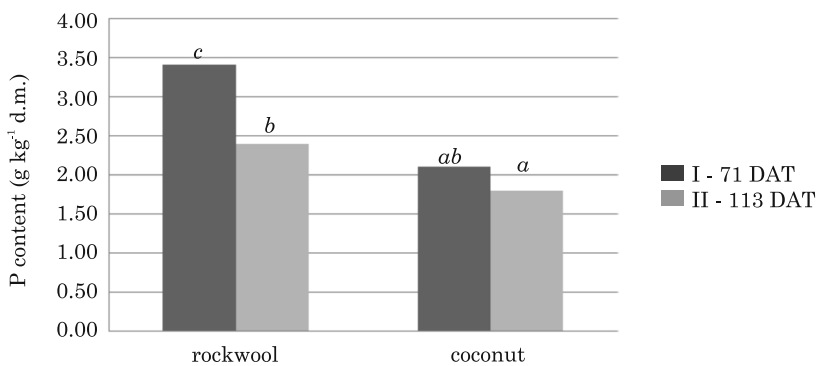

Fig. 2. The content of P in roots of tomato plants; interaction: substrate type x stage of growth; $P < 0.05$

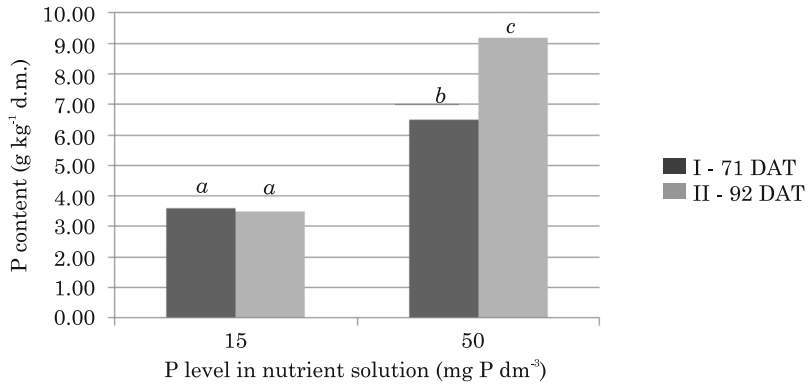

Fig. 3. The content of P in leaves of tomato plants; interaction: P level in nutrient solution x stage of growth; $P < 0.05$

The RT-PCR amplification yielded the transcripts of lengths of approximately 180 pb – 300 bp (*LePT1* – 287 bp, *LePT2* – 262 bp, *LePT3* – 206 bp and *LePT4* – 179 bp) – Figure 4A-4D. Generally, the transcript levels depended on the amount of P in the nutrient solution, plant growth stage as well as the type of substrate.

The expression of genes studied was generally higher (except *LePT4*) in plants treated by the nutrient solution with 15 mg P dm^{-3} in comparison to 50 mg P dm^{-3}. The expression of genes *LePT1-LePT4* in roots of older plants

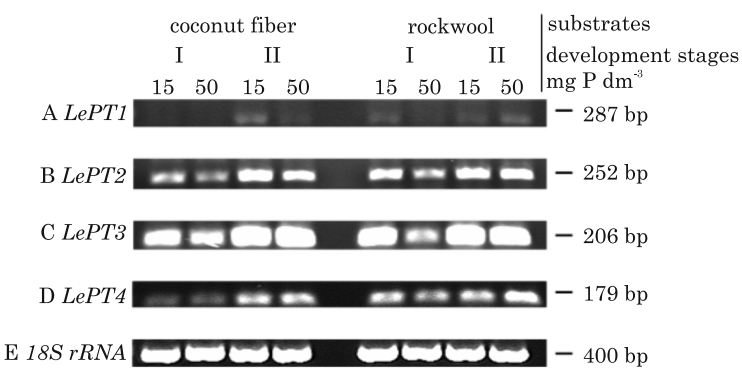

Fig. 4 A-E. Expression of the genes *LePT1-PT4* and *18S rRNA* at two fruit developmental stages, types of medium and phosphorus concentrations in a nutrient solution determined by RT-PCR

(113 DAT, Figure 4A-D) was generally higher than in young plants (71 DAT, Figure 4A-D). The effect of substrates was noted on the expression of *LePT1* and *LePT4*. The expression of these genes was stronger when the plants were grown in rockwool. Furthermore, regardless of the substrate type, the expression of *LePT2* and *LePT3* was also stronger in roots of young plants grown in the medium supplied with 15 mg P dm^{-3} than in roots of plants grown on 50 mg P dm^{-3}. However, there was no evident effect of the substrate tested

on the expression of genes *LePT2* and *LePT3* in older plants, regardless of the P level in a nutrient solution. However, in roots of older plants (113 DAT) grown in coconut fiber, the level of *LePT1* and *LePT2* transcripts was lower in plants fed by the nutrient solution with 50 mg P dm^{-3}. On the other hand, the expression of *LePT3* and *LePT4* genes in roots of plants (113 DAT) grown in coconut fiber was similar and independent of the P concentration in the medium. Finally, roots of plants (113 DAT) grown in rockwool and supplied the nutrient solution with 50 mg P dm^{-3} showed a generally higher expression of the genes (except *LePT3*) than plants fertilized with 15 mg P dm^{-3}.

As shown in Figure 5, genes encoding phosphorus transporters in the Neighbour-Joining tree were well clustered into four distinct groups. BLASTn analyses confirmed the identity of *LePT* genes and, therefore, the specificity of the primers. The sequence showed that *LePT1* was closely related to AK324945.1, AF022873.1 and Y14214.1, while *LePT2* was akin to XM004233991.1 and NM001247114.1. The nucleotide sequence of *LePT3*

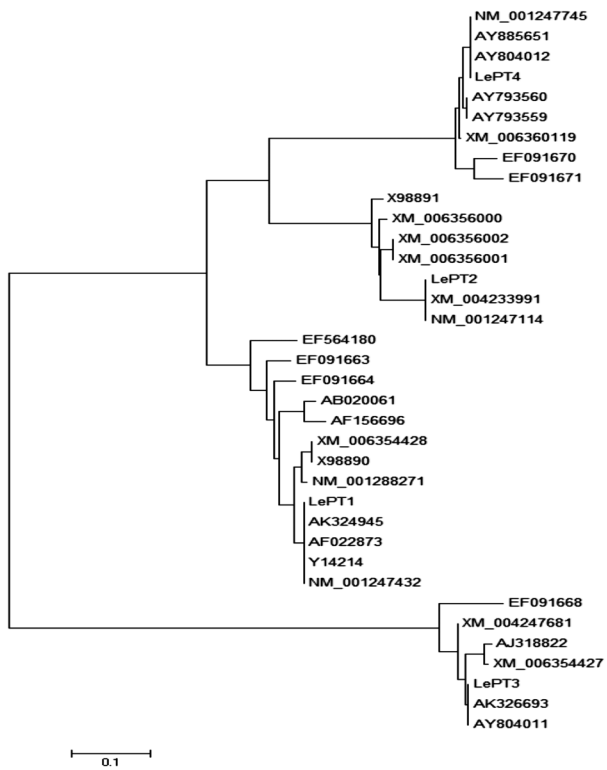

Fig. 5. Phylogenetic analysis of tomato *LePT1-LePT4* genes and other plant *PT* homologs. An unrooted phylogenetic tree of the plant genes encoding phosphorus transporters was constructed using the neighbour-joining method with the MEGA 6.0 programme. An optimal tree with the sum of branch lengths = 1.97070575 is shown. The tree is drawn to scale, with branch lengths in the same units as those of the evolutionary distances used to infer the phylogenetic tree. The evolutionary distances were computed using the Maximum Composite Likelihood method (Tamura et al. 2004) and are in the units of the number of base substitutions per site

revealed 100% identity with PT3 (accession no. AY804011.1) and probable inorganic phosphate transporter 1-7-like (accession no. XM004247681.1), whereas *LePT4* revealed 100% identity with PT4 (accession no. AY804012.2, AY885651.1 and NM001247745.1).

DISCUSSION

The aim of this study was to determine the effect of the P content in a nutrient solution on the expression of genes encoding proteins involved in the transport of P in plants, as well as on the P content in roots and leaves of tomatoes grown in different substrates. The observations were performed on young and older plants.

Reduction of the P content in the medium from the standard value (50 mg dm^{-3}) to a much lower content (15 mg dm^{-3}) decreased the P concentration in roots and leaves, which is in line with the results of ROSERWARNE et al. (1999). The mean content of P in leaves of plants supplemented with 15 mg P dm^{-3} was slightly below the range (0.40-0.65% d.m. – g kg^{-1} d.m.) considered by ATHERTON and RUDISCH (1986) and SADY et al. (1998) as optimal for tomato plants. On the other hand, the P content in leaves of plants grown on the nutrient solution with 50 mg P dm^{-3} (7.8 g kg^{-1} d.m.) was above the upper limit value in tomatoes grown in rockwool. According to REUTER and ROBINSON (1997), the mean P concentration in leaves equal 0.65% d.m. (6.5 g kg^{-1} d.m.) was adequate for the optimal growth of tomatoes. These differences might be explained by the difference between sampled leaves in our study and in other studies.

In our study, the reduction of P content in roots and leaves as a result of the lower P content in a nutrient solution occurred regardless of the type of substrate and stage of growth. On the other hand, older plants contained less P in roots than younger ones, when the nutrient solution with 50 mg P dm^{-3} was applied, whereas leaves of older plants contained more P than younger ones. These changes might have resulted from the transport of P from roots to leaves and accumulation of organic compounds containing P in leaves. It should be also noted that the P content in leaves was 2-3 times greater than in roots, which also confirms the accumulation of P in vegetative parts of plants. Similarly, a higher P content in tomato leaves than in roots was also observed by ROSEWARNE et al. (1999). Greater accumulation of P in leaves of older plants is opposite to the results of BAR-YOSEF and IMAS (1995), who found a decrease in the P content in leaves of tomato from 0.50 to 0.25% d.m. (5.0 to 2.5 g kg^{-1} d.m.) during the growing season. With the low P supply in our study (15 mg dm^{-3}), there were no differences in the P content in roots or leaves between younger and older plants. It is quite possible that in response to such a poor P supply older plants decreased the accumulation of organic compounds in leaves.

Phosphorus stimulates healthy root growth, which helps to improve the absorption of water and nutrients (Abd-Alla et al. 1996, Sainju et al. 2003). Reduction of the content of the most of micronutrients in roots as a consequence of a lower P level in the tested nutrient solution might have been caused by the deficit of P as an essential component of high energy compounds (e.g. ATP), necessary for the uptake of microelements (Marschner 1995). Moreover, phosphate ions are involved in the activation of many genes, including transporter protein participating in the transport of ions through the membrane (Liu et al. 1998*a*, Raghothama 2000).

The content of almost all macro- and microelements was higher in roots of plants grown in rockwool than in coconut fiber. Regardless of the level of P in the medium, plants grown in rockwool also contained significantly more P in roots, but not in leaves. Both substrates were fertigated by the same dose of a nutrient solution. Coconut as a substrate has a lower water holding capacity than rockwool, which may decrease the volume of solution kept in the root zone and consequently the amount of nutrients available, which is especially important for anions. The differences between the substrates in the P content of roots were observed only in younger plants (71 DAT), which is not easy to explain. Chohura and Komosa (2003) observed a higher P content in leaves of plants grown in expanded clay than in rockwool and polyurethane foam. The results of the above experiment show that the substrate type should be considered as an experimental factor in studies on the P supply to tomato plants. The effect of a substrate type on the expression of genes encoding P transporters is discussed below.

Expression of *LePT1-LePT4* genes and the plant's P status

The RT-PCR amplifications resulted in amplicons of the length varying from ~180 bp to ~300 bp, similar to those described by Balestrini et al. (2007).

Phosphorus acts as both a substrate and regulatory factor in photosynthesis and oxidative metabolism; it also participates in signal transduction by the way of protein phosphorylation/dephosphorylation. Its low solubility and high chemical sorption capacity in soils make it relatively unavailable to plants (Marschner 1995). As a result, P_i supply is one of the most important constraints to crop production worldwide (Marschner 1995, Raghothama 1999). It has long been known that P deficiency triggers a large number of physiological changes in plants, expected to enhance P_i acquisition from soil and to deal with suboptimal levels of P within the plant (Wang et al. 2002). These changes are related to the plant morphology (root architecture and root:shoot ratio) and physiology (stimulation of root P_i absorption, increases in the release of phosphatases and RNAses from roots, high energy phosphate bonds e.g. ATP (Raghothama 1999).

Molecular research on plant P nutrition has recently indicated that P deficiency also induces changes in the gene and protein expression in a way that indicates a number of genes and proteins involved in P nutrition which

are regulated by the plant's P status (Liu et al. 1998*a*, Muchhal, Raghothama 1999, Raghothama 1999, Mukatira et al. 2001). Also in our experiment, as we hypothesized, the expression of genes encoding P transporters depended on the P content in a nutrient solution and therefore on the plant's P status. At low P in the nutrient solution (15 mg P dm^{-3}), the expression of *LePT2* and *LePT3* genes was stronger than at 50 mg P dm^{-3}. According to Żebrowska and Ciereszko (2007), under conditions of low P availability (less than 20 mM) in the root zone, Pht1 (including *LePT1-4*) transporters are activated and these proteins with high affinity to phosphate (P_i) are responsible for the transport of P_i ions into the root cells. Liu et al. (1998*a*) reported that *LePT1* and *LePT2* were induced in a temporal and P_i - concentration-dependent manner. The induction of these genes was a rapid response to P_i starvation and is reversible upon replenishment of phosphorus. These reactions indicate some possible coordination between gene expression and an increased uptake in P_i. *LePT1-4* genes are homologous to those of plants from the family *Solanaceae* which belong to Pht1 and which encode transporters with high affinity to phosphorus.

A lower content of phosphorus in roots sampled from older plants (113 DAT) was accompanied by a stronger expression of *LePT2* and *LePT3* genes than during fruit setting. This may indicate an increased demand of older plants for phosphorus and its more intensive transportation from roots to leaves. At this stage of growth, tomato plants demonstrate higher intensity of photosynthesis than younger ones.

The higher content of phosphorus in roots of plants grown in rockwool than in coconut fiber was in line with the increased expression of *LePT2* and *LePT3* in roots of plants grown in rockwool (Figure 4B-C). This phenomenon may be explained by a more intense involvement of the products of these transcripts in the uptake of phosphorus from the medium.

The results obtained in our study provide a prospect for further studies on the molecular mechanism of adaptations of tomato plants to P-deficiency. Further characterization and functional analysis of the gene expression will help to improve its phosphorus utilization and overall productivity.

CONCLUSION

1. The level of P in the medium, regardless of the substrate type and stage of growth, had an effect on the content of phosphorus, iron, boron and copper in roots and phosphorus in leaves of tomato plants.

2. The content of macro- and microelements in roots of plants grown in rockwool was higher than in roots of plants grown in coconut fiber.

3. The stage of plant growth significantly affected the mineral composition of roots as well as the P content in tomato leaves.

4. At a low P content in a nutrient solution (15 mg P dm^{-3}), the expression of *LePT2* and *LePT3* genes in plant roots was stronger than at 50 mg P dm^{-3}.

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