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ORIGINAL PAPER

Impact of arbuscular mycorrhizal fungi on the allergenic potential of tomato

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Abstract Arbuscular mycorrhizal (AM) fungi influence the expression of defence-related genes in roots and can cause systemic resistance in plants probably due to the induced expression of specific defence proteins. Among the different groups of defence proteins, plant food allergens were identified. We hypothesized that tomato-allergic patients differently react to tomatoes derived from plants inoculated or not by mycorrhizal fungi. To test this, two tomato genotypes, wild-type 76R and a nearly isogenic mycorrhizal mutant RMC, were inoculated with the AM fungus *Glomus mosseae* or not under conditions similar to horticultural practice. Under such conditions, the AM fungus showed only a very low colonisation rate, but still was able to increase shoot growth of the wild-type 76R. Nearly no colonisation was observed in the mutant RMC, and shoot development was also not affected. Root fresh weights were diminished in AM-inoculated plants of both genotypes compared to the corresponding controls. No mycorrhizal effects were observed on the biomass and the concentration of phosphate and nitrogen in fruits. Real-time quantitative polymerase chain reaction analysis revealed that six among eight genes encoding for putative allergens showed a significant induced RNA accumulation in fruits of AM-colonised plants. However, human skin reactivity tests using mixed samples of tomato fruits from the AM-inoculated and control plants showed no differences. Our data indicate that AM colonisation under conditions close to horticultural practice can induce the expression of allergen-encoding genes in fruits, but this does not lead necessarily to a higher allergenic potential.

Keywords Allergy · Defence proteins · *Glomus mosseae* · RNA accumulation · Skin prick test

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Introduction

Arbuscular mycorrhizal (AM) fungi are ubiquitous soil microorganisms which form a mutualistic symbiosis with the roots of 80% of all land plants (Smith and Read 2008). Their main function is the improvement of plant nutrition by supplying mineral nutrients (Marschner and Dell 1994). In addition to this function, AM fungi can induce a systemic response in plants altering the interaction of upper plant parts with pathogens, probably due to the differential expression of pathogenesis-related and defence proteins (e.g. Gernns et al. 2001; Lingua et al. 2002; Garmendia et al. 2004; Fritz et al. 2006; Liu et al. 2007).

Tomato (*Solanum lycopersicon* Mill.) belongs to the relevant allergenic food causing skin symptoms, such as urticaria, angioedema and dermatitis, but also oral allergy

symptom, rhinitis and abdominal pain (Zacharisen et al. 2002). The prevalence of tomato allergy ranges from 1.5% (Northern Europe; Petersen et al. 1996) up to more than 16% (Italy; Ortolani et al. 1989) among food-allergic populations. At the moment, 18 potential tomato allergens, including the different isoforms, have been reported and were published in different structural databases of proteins related to allergens (http://www.allergome.org; http://fermi.utmb.edu/SDAP/). Among these allergens are proteins of pathogenesis or defence, such as chitinase, glucanase, peroxidase or lipid transfer proteins (Breiteneder 2009).

In addition, research data revealed that several proteins related to defence mechanisms of the plants are responsible for latex allergy, for the accompanying cross-reactivity or directly for food related allergies (Hoffmann-Sommergruber 2002; Wagner and Breiteneder 2002; Yagami 2002; Foetisch et al. 2001). Hence, one can hypothesize that food derived from AM fungal inoculated plants result in a different reaction pattern of food-allergic patients compared with food from plants free of mycorrhizal colonisation. Until now, variations in the allergic reactions were reported after testing eight horticultural cultivars of bell pepper (Jensen-Jarolim et al. 1998), four cultivars of hazelnut (Wigotzki et al. 2000) and also 21 apple cultivars (Vieths et al. 1998; Bolhaar et al. 2005). In contrast, nothing is known on how production conditions of food crops affect hypersensitivity, although it is clear that such conditions influence protein expression patterns and in consequence could also induce the occurrence of specific allergens.

We tested this hypothesis by inoculating a tomato cultivar (76R) and a corresponding isogenic mycorrhizal mutant (RMC) with an AM fungus under conditions similar to commercial cultivation in horticulture. Plants were harvested and first analyzed for growth characteristics and their fruits for nitrogen and phosphorus content in order to assess the influence of the mycorrhiza under regular phosphate conditions. Secondly, RNA extracts from tomatoes were analysed for the expression of allergene-encoding genes and tomato extracts for their allergenic potential.

Materials and methods

General treatment conditions

A greenhouse experiment was carried out at the Leibniz Institute of Vegetable and Ornamental Crops in Großbeeren, Germany. Seven weeks after germination in coarse sand, 16 tomato plants at eight-leaf stage were transferred each into one 10-l bucket filled with a sand/vermiculite mixture (1/1 *v*/*v*). Mean temperature, daily radiation and humidity were 23.1°C, 20.2 molm^{-2} and 61.3%. Four treatments were imposed in a 2×2 factorial design with four replications. Factor one (mycorrhiza treatments) was chosen to test a reaction of the tomato against mycorrhiza inoculation consisting in a mock-inoculated and a mycorrhizal treatment. For the latter, mycorrhizal inoculum, *Glomus mosseae* (Nicol.&Gerd.) BEG 12 (Biorhize: Dijon, France) was uniformly mixed (5% *v*/*v*) with the sand/vermiculite substrate in the pots before planting the seedlings. Filtered drain of BEG 12 inoculum (589/3 blue ribbon paper filter, Schleicher & Schuell Bioscience GmbH, Dassel, Germany) was added to the mock-inoculated treatment. In addition, mock-inoculated treatments were supplied with autoclaved BEG 12 inoculum (121°C for 20 min). Factor two consisted in two genotypes. 76R is a commercial tomato cultivar (Petoseed Company, Santa Maria, CA, USA), and RMC is an isogenic mutant showing less mycorrhizal colonization of the roots (Barker et al. 1998). Tomatoes were irrigated manually until drain started. Once a day, pots were supplied with tap water and twice a week with nutrient solution: NH_4NO_3 (1), $Ca(NO_3)_2{\cdot}4H_2O$ (2.75), KNO_3 (6.5), KH_2PO_4 (1.25), K_2SO_4 (1.5) and $Mg(NO_3)_2$ (1), in millimolar; FeEDTA (25), $MnSO_4$ (10), H_3BO_4 (20), MoO_3 (0.5) $CuSO_4$ (0.75) and $ZnSO_4$ (4) in micromolar.

Plant analysis

One sub-sample of the root system of each treatment and each replication was taken with an auger (200 mm length, 225 ml) to measure root length and mean root diameter. Roots were washed out from the substrate and fresh weighed. Diameter was measured on 20 roots randomly sampled from each treatment, and each replication and mean root diameter was calculated. Considering a root as a cylinder, length of the sub-sample was calculated based on diameter and fresh weight, i.e. volume. Total root length was calculated by the ratio sub-sample to total substrate volume in the buckets (factor 44.25). Fruits were harvested once a week when red-ripe, stage 8–9 of the colour screening scale for tomato (Anonymous 1992). At the end of the experiment (9 weeks after inoculation), shoots were separated from roots, and fresh and dry weighed. Total nitrogen in fruits was measured using an Auto Analyzer (Dumas combustion; Heraeus, Hanau, Germany). Phosphorus was analyzed spectrophotometrically using the vanadate method (Merck 14842).

Arbuscular mycorrhizal colonisation

In order to determine mycorrhizal colonisation characteristics, roots were cleared with 10% KOH, acidified with 2 N HCl and stained with 0.05% Trypan blue in lactic acid (Phillips and Hayman 1970). In 30 root segments each with 10-mm length inoculation frequency, relative colonisation intensity and

arbuscular frequency were determined under a microscope and calculated according to Trouvelot et al. (1986).

RNA accumulation analyses

Frozen fruit material from each treatment and replication was ground in liquid nitrogen, and RNA was extracted using the RNeasy Plant Mini Kit (Qiagen, Hilden, Germany) following manufacturer's instructions. Total RNA was treated with the RNase free DNase Set (Qiagen) and reverse transcribed with an M-MLV Reverse Transcriptase System using oligo-dT primer (Promega, Mannheim, Germany). The primers for eight target genes selected were designed using the DNAStar Primer Select software (GATC Biotech, Konstanz, Germany) based on tomato mRNA sequences deposited in NCBI Database (Tables 1 and 2). One representative isoform was taken from each tomato allergen nucleotide sequence known. Quantitative real-time polymerase chain reaction (PCR) was carried out using the 7500 Fast real-time PCR System (Applied Biosystems, Darmstadt, Germany) with the following temperature program: 50°C for 2 min, 95°C for 10 min, 40 cycles of 15 s at 95°C and 1 min at 60°C. The real-time PCR reactions were performed in triplicate. Relative gene expression values were calculated as $2^{\Delta Ct}$. ΔCt describes the difference between Ct values obtained from the geometric mean of two reference genes encoding the clathrin adaptor complexes medium subunit and the SAND family protein recommended for RNA accumulation analyses in tomato fruits (Expósito-Rodríguez et al. 2008) and Ct values obtained from the target gene.

Patients screening, skin prick test and specific IgE

All patients were recruited from the Allergy-Centre-Charité, Berlin, Germany. The study was approved by the Local Ethics Committee (Charité—Universitätsmedizin Berlin, EC-No. 1832/Si.258), and patients gave written informed consent. Inclusion criteria for the selection of patients were an age ≥18 years, a history of adverse reactions to and a positive skin prick test response with tomato. Among

Table 1 Mycorrhization characteristics of tomato cultivars, wild-type 76R and mutant RMC, after 9 weeks growing in the greenhouse

Mycorrhization	76R	RMC	Effect
Colonization frequency in %	29.20	4.17	–*
Relative colonization intensity in %	4.20	0.75	–*
Relative arbuscular frequency in %	1.89	0.00	–*

Roots of mock-inoculated plants harboured no mycorrhizal structures
*Significant differences at P=0.05 (one-way ANOVA; n=4)

Table 2 Pulative allergens in tomato, their function, accession (AC) number, references for their identification and primer pairs used in the current analysis

Allergen	Function	AC number	Reference	Forward primer	Reverse primer
Lyc e 1	Profilin	AJ417553	Westphal et al. (2004)	TGGGCTCAATCTGCTAAT	AGTCATCGGCTCGTCATA
Lyc e 2	β-Fructofuranosidase	S70040	Westphal et al. (2003)	CGTACCCCGCCAACTTATCTG	CAATGCCGGGTGGAGGAAC
Lyc e 3	Lipid transfer protein	AM051295	Asero et al. (2000)	TGGCTCCTTGTCTCCCTTATCT	TTGCCCAAATTGAGTCCTGTA
Lyc e Chi	Chitinase	Z15140	Diaz-Perales et al. (1999)	ATGGGGTTACTGTTTCCTTAGAGA	TGGCCCATAGTTGTAGTTGTGT
Lyc e Glc	Glucanase	M80608	Fuentes-Silva et al. (2006)	TCAAACATCATGGCTACCTCACAA	TCCCCATCATTCCATAACAAACAC
Lyc e PG	Polygalacturonase	X05656	Kondo et al. (2001)	GGCTTTGGATTGCTTTTGATAGTG	CAGAAGGTTAAGGCCGTTGGTG
Lyc e PE	Pectinesterase	X74638	Foetisch et al. (2001)	GGGCTGAGTGGGACGGAGATT	CCAGGCCACTTGACACGCTTACT
Lyc e NP24	Osmotin/PRP23	AF093743	Kondo et al. (2001)	TATATGGGGTCGTACTGGTTGC	GCTAGGGTGTTTGGGGGTTTG

20 persons with a clinical history of tomato allergy recruited for this study, ten patients had a positive reaction in the skin prick test. Skin prick tests were performed according to the recommendations of GA^{2}LEN (Heinzerling et al. 2005) by using the prick-to-prick method with native material. Prick-to-prick tests were performed in duplicate on the surface of the interior forearm with standardized prick needles. Histamine dihydrochloride (10 mg/ml, ALK Scherax, Wedel, Germany) and saline solution (pH 7.4, ALK-Scherax) served as positive and negative controls, respectively. Allergenicity of the different tomatoes was analyzed by measuring the wheal diameters. The skin prick test response was considered positive when the wheal diameter was ≥3 mm after 15 min in the absence of a reaction in the negative control (Heinzerling et al. 2005). The specific IgE (sIgE) was measured with the Phadia CAP System FEIA® (Uppsala, Sweden), according to the manufacturer's instruction. Measurement of the specific IgE to tomato makes sure that the patient is sensitised towards tomato and can reflect patient's reaction presupposed it matches his clinical history.

Statistical analyses

Tomato data and real-time PCR results were subjected to two-way analysis of variance (ANOVA) procedures using Statistica software (StatSoft Inc. 2004, Tulsa, OK, USA). Means were separated by Tukey's test procedure at $P=0.05$. Prick test data were subjected to non-parametric test. Medians were separated by Mann–Whitney U test procedure at $P=0.05$.

Results

Impact of mycorrhization on tomato vegetative growth

Tomato roots of the wild-type 76R were successfully inoculated with the AM fungus *G. mosseae* under normal phosphate fertiliser conditions, although the values of the different mycorrhization parameters were relatively low (Table 1). These values were even lower in the mutant RMC: Inoculation frequencies were reduced from 29.20% to 4.17%, and colonization intensity and arbuscular frequency amounted to 4.20% and 1.89% for wild-type 76R but almost 0 for the mutant RMC (Table 1). Fresh mass of shoots was significantly increased after inoculation of wild-type 76R plants, but not of the mutant RMC (Table 3). In contrast, root fresh masses were lower in inoculated plants of both genotypes compared to the corresponding controls. Other vegetative growth characteristics were not significantly affected by the mycorrhizal treatment of the wild-type 76R, but nearly all growth characteristics showed significant differences between the two genotypes (Table 3).

Impact of mycorrhization on tomato fruits

For revealing, if mycorrhization has any systemic influence on generative organs, fruits were analysed in detail. The overall fruit yield was similar between treatments, but the dry matter content of mature fruits of the mutant RMC was 5 $g kg^{-1}$ higher compared with the wild-type 76R irrespective of AM (Table 3). Nitrate, total nitrogen and phosphate

Table 3 Characteristics of tomato genotypes (gt), wild-type 76R and mutant RMC, 70 days after inoculation with arbuscular mycorrhizal (AM) fungus *Glomus mosseae* or mock-inoculated (C)

Parameter	Unit	76R		RMC		Effects		
		C	AM	C	AM	gt	AM	IA
Shoot fresh mass	$g plant^{-1}$	673	792[a]	804	825	–*	ns	ns
Dry mass	$g plant^{-1}$	97.8	110.7	120.4	122.4	–*	ns	ns
DMC	$g kg^{-1}$	145	139	150	149	–*	ns	ns
Root fresh mass	$g plant^{-1}$	199	163	221	151	ns	–*	ns
Length	$m plant^{-1}$	2270	2023	2551	1650	ns	ns	ns
Diameter	mm	0.340	0.324	0.334	0.342	–*	ns	ns
Fruit yield	$g plant^{-1}$	629	633	549	624	ns	ns	ns
DMC	$g kg^{-1}$	70.1	70.4	75.6	74.6	–*	ns	ns
NO_3 conc.	$g kg^{-1}$	0.325	0.300	0.300	0.375	ns	ns	ns
Total N conc.	$g kg^{-1}$	28.9	29.2	28.2	27.7	–*	ns	ns
Total P conc.	$g kg^{-1}$	6.40	6.57	5.73	5.60	–*	ns	ns

A two-way ANOVA ($P=0.05$; $n=4$) was carried out with gt and AM as factors (IA=interactions between factors)

DMC dry matter content, *ns* non-significant differences

*Significant differences

[a] Shoot fresh masses were significantly increased in wild-type 76R after inoculation with *Glomus mosseae*

concentration in fruits were not significantly affected by the mycorrhizal treatment, but fruits of the mutant RMC showed significantly lower values for total nitrogen and phosphate compared with the wild-type 76R (Table 3).

In order to assess whether the production of proteins, which represent allergens in fruits, is influenced by mycorrhization of tomato plants upon regular phosphate conditions, the mRNA profile of eight genes encoding for putative allergens was analysed in fruits (Table 2). All eight genes investigated showed an induction after inoculation with mycorrhiza of wild-type 76R and mutant RMC plants. This was independent of the genotype since the two-way ANOVA did not reveal any differences between them. Although variations between biological replicates were high, resulting in high standard errors, the difference between AM- and mock-inoculated plants was significant for the allergens Lyc e 1 (profilin), Lyc e 2 (β-fructofuranosidase), Lyc e Chi (chitinase), Lyc e Glc (glucanase), Lyc e PE (pectinesterase), Lyc e NP24 (osmotin/PR P23) but not for Lyc e 3 (lipid transfer protein) and Lyc e PG (polygalacturonase; Fig. 1).

Allergenic potential of tomato

To investigate if the differential expression of genes encoding for putative allergens influences the allergenic potential of

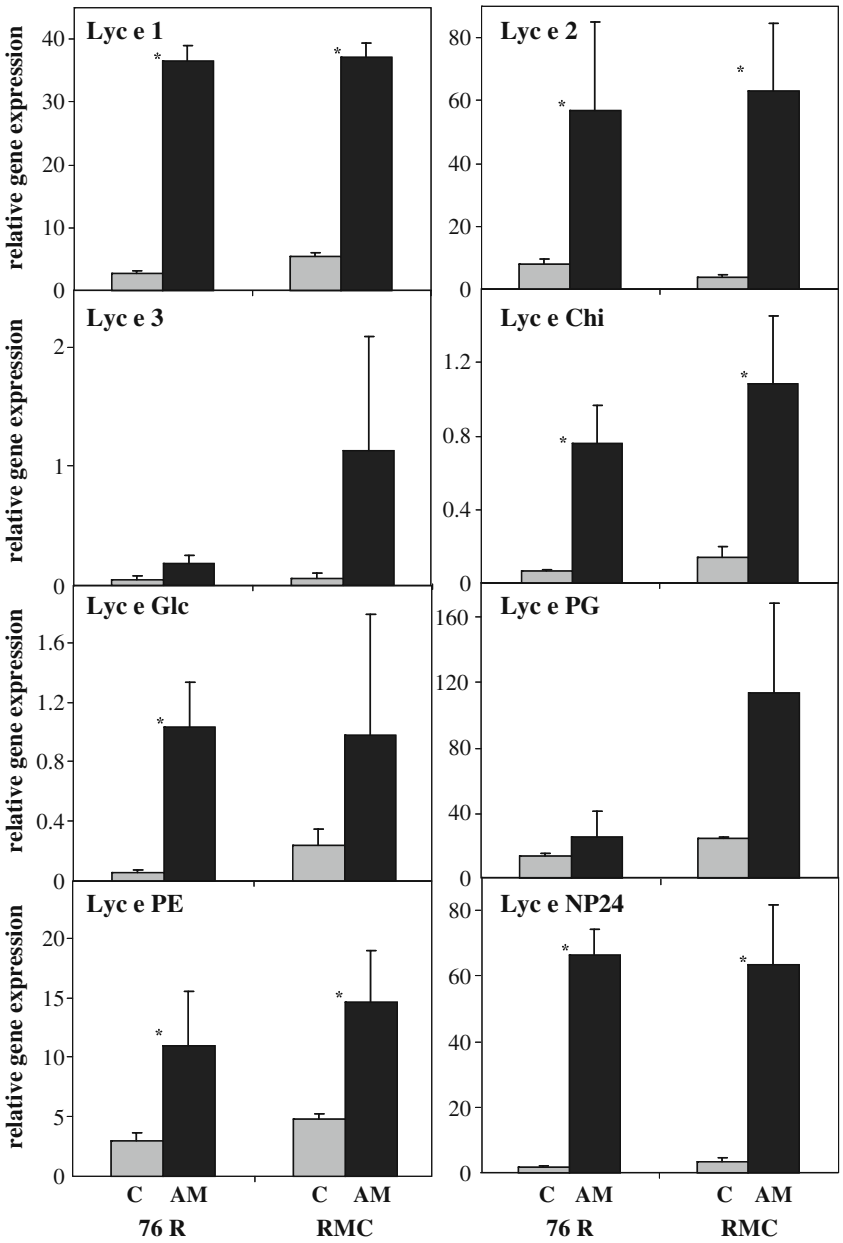

Fig. 1 Influence of mycorrhiza inoculation on gene expression of the putative allergens Lyc e 1 (profilin), Lyc e 2 (β-fructofuranosidase), Lyc e 3 (lipid transfer protein), Lyc e Chi (chitinase), Lyc e Glc (glucanase), Lyc e PG (polygalacturonase), Lyc e PE (pectinesterase) and Lyc e NP24 (osmotin/PR P23) in tomato fruits from wild-type 76R and mutant RMC plants. Quantitative real-time PCR was carried out using total RNA from fruits and relative RNA accumulation rates are shown. *Bars* represent standard errors. Significant differences between plants inoculated with the arbuscular mycorrhizal (AM) fungus *Glomus mosseae* (dark grey bars) and mock-inoculated controls (C; light grey bars) are indicated by asterisks (two-way ANOVA; $P=0.05$; $n=4$). Interactions between the factors genotype and inoculation or any significant influence of the genotype were not detected

Table 4 Characteristics of recruited patients

Patients	Age, years	Sex	Total IgE, kU/l	sIgE, kU/l	Symptoms to tomato, case history
2004-01	40	M	114	2.02	GIT
2004-02	41	M	184	<0.35	OAS
2004-03	36	M	197	0.86	OAS
2004-04	43	M	51	<0.35	OAS
2004-05	38	M	155	<0.35	R
2004-06	18	F	383	0.82	OAS
2004-07	18	F	234	nd	OAS
2004-08	36	F	405	0.92	GIT
2004-09	36	F	255	nd	OAS
2004-10	18	F	476	0.65	OAS

Age, sex, total IgE and specific IgE (sIgE) to tomato and subjective symptoms are listed

Age years: median, 36; min/max, 18/43; sex: F/M, 5/5; total IgE: median, 216; min/max, 51/476; sIgE: positive, 5; total, 10 *GIT* symptoms of the gastrointestinal tract, *OAS* oral allergy syndrome, *R* rhinitis

tomato fruits, a group of 13 patients with allergic symptoms after tomato consumption (Table 4) was recruited. Skin prick tests were carried out with these patients, and differences were identified between the genotypes tested (Fig. 2). A trend towards an overall increased skin reactivity of the mutant RMC compared to the wild-type 76R was observed, but the differences failed to be statistically significant. Inoculation of the plants with the AM fungus had no influence on wheal diameters at all. A reason for that observation might be the large variation of patient's individual skin reactivity.

Discussion

Tomato roots of the wild-type 76R were successfully inoculated with the AM fungus *G. mosseae* (Table 1), although the colonisation rate was low compared with other reports on the same cultivar (e.g. Gao et al. 2001). This is probably due to the level of phosphate fertilisation in the present experiment, as it is well known that a normal supply inhibits the development of the mycorrhizal symbiosis (Gerdemann 1968; Gianinazzi-Pearson and Diem 1982). Nevertheless, a significant impact of mycorrhization on vegetative growth parameters was observed: Shoot fresh weights increased, but roots developed less fresh masses (Table 3). Because plants were optimal fertilised and irrigated, the reason cannot be an improved nutrient or water supply. An alternative explanation might be that changes in the phytohormone balance of the plant occurred (Hause et al. 2007). Increased shoot growth might be due to increased cytokinin levels (Allen et al. 1980; Drüge and Schönbeck 1990). While indeed several vegetative growth characteristics were affected by mycorrhization, fruit characteristics did not differ between plants either inoculated with the AM fungus *G. mosseae* or mock inoculated (Table 3). The low colonisation of the roots in this study upon the conditions of regular phosphate fertilisation seems to have no influence on yield, dry matter content or concentrations of nitrogen and phosphorus in the fruits. This result was expected and helps to study mycorrhizal effects on allergenicity without confounding mycorrhizal effects on plant growth and nutrient uptake.

A clear difference in numerous parameters was observed between the wild-type cultivar 76R and its isogenic mutant RMC: Colonization frequencies were significantly reduced, and arbuscules were not observed (Table 1). This is in accordance with a previous report on interactions between the genotype RMC and *G. mosseae*: Though the AM fungus is able to penetrate the epidermis of the root, cortex colonisation is rare (Gao et al. 2001). In contrast to the wild type, shoot fresh masses were not higher in colonised roots of the mutant RMC (Table 3). If growth promotion depends on changes in the phytohormone concentrations as dis-

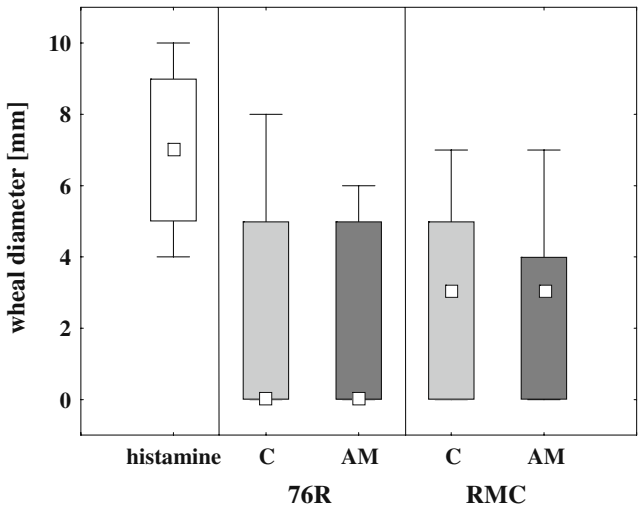

Fig. 2 Skin prick tests with tomato extracts of genotypes, wild-type 76R and mutant RMC, 9 weeks after inoculation with the (AM) fungus *Glomus mosseae* or from the corresponding mock-inoculated controls (C). Histamine dihydrochloride (10 mg/ml) was used as positive control. The median is marked as a *white square*. The *boxes* depict the quantiles and the *lines* the range of wheal sizes of ten tomato-sensitized patients. Mann–Whitney *U* test procedure at $P=0.05$ did not reveal any significant differences

cussed above, the absence of any differences in this parameter is probably based on the low colonisation. At least for jasmonate, it has been shown that the production is regulated in arbusculated cells (Isayenkov et al. 2005). Interestingly, root fresh weights were reduced as in the wild type (Table 3). This seems not to be dependent on cortex colonisation, but on the presence of the fungus in the soil, the physical contact with the root and/or the penetration of the epidermis. Such a phenomenon has up to now not been described for the AM symbiosis, but ectomycorrhizal fungi do affect plant root morphology before and during first contact (e.g. Herrmann et al. 1998).

AM fungal inoculation of tomato plants can lead to increases in fruit yield (Subramanian et al. 2006) and in the content of phosphorus in the fruits (Al-Karaki and Hammad 2001). This was not the case in the current experiment probably due to the low colonisation level upon the regular phosphate fertilisation. The nitrogen content was also not affected. However, more characteristics were affected by the genotype. In general, fruits from the mutant RMC had a higher dry matter content accompanied by similar fresh weights but lower nitrogen and phosphorus content. Because, the vegetative growth was enhanced in the mutant compared to the wild-type, it seems to be a matter of biomass distribution between different plant organs (Heuvelink 1996).

In contrast to the lack of difference in the basic characteristics between the fruits of mycorrhizal and control plants, six out of eight investigated genes showed a significant mycorrhiza-induced RNA accumulation. Systemic effects of mycorrhization on gene expression have been previously described in tomato (Taylor and Harrier 2003) and in *Medicago truncatula* (Liu et al. 2007), but it is the first time that fruits were analysed in this respect. This phenomenon could be based on changes in phytohormone levels, as it was already observed in leaves of mycorrhizal plants (Hause et al. 2007). Interestingly, significant induction of five genes was also evident in the mutant RMC, indicating that signalling processes between AM fungus and plant before, during contact or at epidermal penetration, seem to be sufficient for this type of response, as it was discussed above for the reduction in root fresh weight. This has been already observed in the roots of the RMC mutant, where pathogenesis- and defence-related genes were similarly induced by different AM fungi as in the corresponding wild-type plants (Gao et al. 2004). It is not likely that the induction of the genes is part of the systemically induced resistance or tolerance by mycorrhiza against abiotic or biotic stress. A fully developed mycorrhiza is usually necessary to develop this symbiotic function (Slezack et al. 2000). All of the encoded proteins are, however, described to interact with sera from tomato-allergic patients (see references in Table 2) and might be the background for a different reaction of such patients to fruits of mycorrhizal and control plants. Our study showed individual reactions of tomato-allergic patients to fruits regardless of the inoculation of tomato plants, but the analysis of the overall patterns revealed no influence of the AM fungus. A different reactivity pattern was only observed between the two genotypes independent of the treatment. Fruits harvested from the mutant plants seemed to harbour a slightly higher allergenic potential as those from the wild type. Different reactions between sera from food-allergic patients to fruits from different cultivars have been described many times (Jensen-Jarolim et al. 1998; Wigotzki et al. 2000; Vieths et al. 1998; Bolhaar et al. 2005). However, skin prick tests showing differences in the reaction of apple-allergic patients to various apple cultivars have been reported only once (Bolhaar et al. 2005). The two genotypes of the current issue are, however, described to be nearly isogenic (Barker et al. 1998), and it was therefore surprising to find these differences independent of mycorrhizal inoculation and of the mycorrhiza-increased RNA accumulation of putative allergen-encoding genes. This could be based on translational control or post-translational modifications of the corresponding proteins which are important for their allergenic potential (Altmann 2007; Ballmer-Weber et al. 2002). Future Western blot experiments using antisera of the tomato-allergenic patients with tomato extracts from mycorrhizal and non-mycorrhizal plants of the different genotypes could help to clarify this question.

The present analyses showed that upon horticultural practice conditions, AM fungi exert a significant effect on tomato plant growth. Most pronounced was the systemic induction of several genes in the fruits. Although these genes encode proteins with allergenic potential, this was not sufficient to change the skin reactivity of the fruits in a group of patients being allergic to tomato. Future research has to show if massive colonisation of the roots by an AM fungus under artificial low phosphorus conditions or the infection of a plant by pathogens is sufficient to increase the allergenic reactivity of tomato plants.

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