EFFECTS OF NANOSCALE ZERO-VALENT IRON (nZVI) AND Fe_2O_3 NANOPARTICLES ON ROOT HYDRAULIC CONDUCTIVITY OF *Solanum licopersicum*

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Introduction

Recently, the synthesis and utilization of iron based nanomaterials with novel properties and functions have been widely studied, both for their nano size and for their magnetic characteristics (Komárek et al., 2015). For that reason, the application of iron nanoparticles (NPs) is being rapidly extended and environmental engineering has used them for remediation purposes, especially for the removal of metal/metalloids from contaminated waters and soils, or their stabilization therein (Gómez-Pastora et al., 2014; Martínez-Fernández et al., 2016b). However, their safety still represents an unknown barrier to their full application. Even when most of the available studies on phytotoxicity of iron NPs have focused mainly on their advantages, relatively few have examined the mechanisms of phytotoxicity, uptake, translocation, and bioaccumulation (Zhang et al., 2015). Considering the roots as the first organ which can suffer from NPs interference in the soil, there is a need to evaluate the impacts on plant physiology of NPs together with their potential ecotoxicity and interactions with the key processes in the rhizosphere (Elsaesser and Howard, 2012). The aim of this experiment was to study the effects of two iron nanomaterials (nZVI and nFe_2O_3) on the plant response of tomato, focusing on the root water conductivity and the effects derived directly from physical phytotoxicity.

Methods

To evaluate the root functionality of *Solanum lycopersicum* L. under nanoparticles exposure, a hydroponic experiment was conducted to obtain intact root systems, and then evaluate the consequences of the individual effects of the two selected NPs. It was performed in 5-l containers, with 4 plants in each, randomly distributed in a glasshouse (20-25°C, 13 h daylight/11 h darkness, PAR 225 µE m^{-2} s^{-1}, 60-80% H^a). The plants were growing 23 days in a modified nutrient solution (Martínez-Fernández et al., 2016a), until the root system had developed sufficiently to allow the determination of the root hydraulic conductivity (L_o). Then, five treatments were imposed: Control without NPs; 50 and 100 mg l^{-1} of nZVI or nFe_2O_3. After the 5 days of treatment with nZVI or nFe_2O_3, L_o was measured by pressurising the roots into a Scholander chamber (model 600; PMS instruments Co., Corvallis, OR, USA) (Martínez-Fernández et al., 2016a). Sap was collected every 60 seconds using a syringe, placed in Eppendorf tubes and weighed on a precision balance. Finally, the L_o (mg g^{-1} FW h^{-1} MPa^{-1}) was calculated as the slope of the line relating mg of gathered sap per g of fresh root and per hour to the external pressure applied (from 0.1 to 1.5 MPa). The plants (shoot and roots separately) were weighed just after harvesting (FW) and were dried to constant weight for the dry weight determination (DW). The Fe concentration was extracted from the plant material by acid digestion (HNO_3/H_2O_2) at 210°C, and determined by inductively-coupled plasma-optical emission spectrometry (ICP-OES) (Varian, VistaPro, Australia). The chlorophyll a (*Chl a*) and chlorophyll b (*Chl b*) contents were determined with the method of Lichtenthaler and Wellburn (1983).

Results

Table 1. Root hydraulic conductivity (L_o), Fe in the sap and shoot, and chlorophyll contents in FW of *S. lycopersicum* cultivated hydroponically, after 5 days of exposure to nZVI or nFe_2O_3. Mean values denoted by the same letter do not differ significantly according to Tukey's test ($p > 0.05$); *ns* not significant.

Nanomaterial	Treatment	*Lo* (mg g^{-1} h^{-1} MPa^{-1})	*Fe in the sap* (mg l^{-1})	*Fe in the Shoot*	*Chl a* (mg kg^{-1})	*Chl b* (mg kg^{-1})
nZVI	Control	2131	0.502	0.6314	1542 a	487 a
	+50 mg l^{-1}	2961	0.316	0.4941	922 b	289 ab
	+100 mg l^{-1}	2316	0.419	0.4336	621 b	276 b
	ANOVA	*ns*	*ns*	*ns*	**	*
nFe_2O_3	Control	2087 a	0.469	0.4824	1458 a	460
	+50 mg l^{-1}	1613 ab	0.451	0.5686	1116 ab	350
	+100 mg l^{-1}	1266 b	0.395	0.4040	669 b	299
	ANOVA	**	*ns*	*ns*	**	*ns*

The high reactive capacity of NPs could stimulate their adhesion to the epithelial root cell wall, and NPs particles appeared changing the color of the roots surfaces (gray for nZVI and red for nFe_2O_3), affecting their interactions with the external medium. Presumably, the accumulation of NPs in the epithelial cells of the root surface interfered with the water transport because of physical and chemical interactions, with the consequent L_o reduction. However, changes in FW or DW were not detected here (data not shown). The recorded effects of nano-oxides on plants indicate lack of uptake, insignificant translocation to the shoots (Martínez-Fernández et al., 2016 a, b). The impacts on the roots may have affected the uptake of elements such as Mg from the solution, a nutrient associated with the synthesis of chlorophyll.

Conclusions

Short-term treatments with nFe_2O_3 at concentrations of 100 mg l^{-1} had significant effects on the root water uptake of *S. licopersicum* plants, but no effects were detected for nZVI, which could be crucial during the maintenance of plant water relations during more privative water conditions.

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