

## Characterization of tomato genotypes under deficiency of phosphorus<sup>(1)</sup>

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**ABSTRACT:** Phosphorus essential is an macronutrient for the development of plants. Although it is not always required in larger amounts, its presence is often limited, since the Brazilian soils and other soils of the world are generally poor in this element. The objective of the research was to evaluate the development of the root system, gas exchange and efficiency in the absorption of phosphorus in tomato genotypes. The experimental design was a randomized block in the factorial scheme 3 x 4 (three tomato genotypes by four doses of phosphorus) with four replications. The tomato genotypes: "Globonnie" PI 121665 (Efficient in phosphorus absorption, crt/crt); TOM-584 considered sensitive in phosphorus uptake (normal, crt+/crt+) and an F1RC1 PI 121665 X TOM -584 genotype, supposedly effective in phosphorus absorption. The phosphorus doses: 0.0, 0.2, 0.5, 30, 60 and 100 mg L<sup>-1</sup>. It was concluded that the genotypes PI 121665 and F1RC1 PI 121665 x TOM-584 showed distinct characteristics which are associated with the efficiency of phosphorus uptake.

**Keywords:** *Lycopersicon esculentum*; WinRhizo; phosphorus.

#### INTRODUCTION

The tomato is produced in almost all geographical regions of Brazil and in different periods under different cropping systems and different levels of cultural management. It stands out as the second most widely cultivated vegetable in the world, only surpassed by the potato. The largest producer is China with 41.8 million tons at 8,700 million m<sup>2</sup> and a productivity of 48.0 tons per 10,000 m<sup>2</sup>. Brazil produced 3.69 million tons, for almost 6.1 e + 8 m<sup>2</sup>, placing 9<sup>th</sup> and 13<sup>th</sup> place respectively worldwide. A segment of the tomato contributed 63.4 percent of production (2.34 million tons) and the remaining 36.6 percent was allocated for industrial processing (1.35 million tons) (FAOSTAT, 2012).

Maximum productivity of the tomato has been associated with doses of fertilizer exceeding 300 kg of  $P_2O_5$  per 10,000 m<sup>2</sup> (Barbosa, 1993), reaching up to 1200 kg of  $P_2O_5$  per 10,000 m<sup>2</sup> (Filgueira, 2008).

The increased efficiency in the absorption of phosphorus by the tomato could provide an appreciable reduction in the doses of phosphate fertilizers provided, as well as, allow it to harness the most immediate fixed phosphorus in the soil. Consequently, it would bring favorable reflections towards agricultural sustainability in the net income of the rural producer, in the harnessing of marginal areas in terms of soil fertility, and in the cost of fertilizer at the national level. A reduction of only 100 kg of  $P_2O_5$  per 10,000 m<sup>2</sup> in the utilization of nutrients by tomato plants would represent a savings of approximately 85.36 dollars per 10,000 m<sup>2</sup>, totaling more than 4.69 million dollars on the national level of tomato production (Silva & Maluf, 2011).

Among the many nutrients necessary for the development and production of plants, phosphorus (P) occupies a prominent place due to the larger amount required by plants (Stauffer & Sulewski, 2004) and its deficiency is apparent in most soils (Lopes et al. 2004). There is a significant genetic variation inter- and intraspecific in the ability of plants to tolerate stress of phosphorus deficiency, called phosphorus use efficiency (Schröder et al., 2011). This efficiency can be based on superior ability to acquire P from soil through changes in morphology or architecture of roots, in the exudation of mobilizing components of P, or of alterations in the inorganic (Pi) transporters of phosphorus the plastic membrane (Kochian et al., 2004). Additionally, it can also involve smaller amounts required of P at the cellular level, or a more efficient remobilization of P within the plant (Yan et al., 2001).

#### MATERIAL AND METHODS

#### Growth conditions and plant material

The experiment was conducted in a greenhouse in 2013 in the Department of Soil Science at the Federal University of Lavras, Brazil. The climate is Cwa, according to Köppen, and characterized by a mean annual air temperature of 66.9 Fahrenheit, average relative humidity of 76.2% and rainfall 1529.7 mm (Dantas et al., 2007).

The seedlings were produced in phenolic foam that remained in the nursery for 30 days, after which

they were transplanted into plastic containers for the place definitive. In these structures, supporting stands were mounted for the first 15 days after transplantation. The tomato seedlings were maintained in five different kinds of nutrient solutions varying only in the phosphorus concentration which placed in plastic containers (virgin was polypropylene) with a capacity of 10 liters. The concentration was diluted to 1/4 of that recommended by the previous researchers. After this adaptation period, the plants were submitted to fully concentrated solutions until 150 days after transplant. The solutions were renewed every 15 days. It is noteworthy that, for the management of nutrient solutions along the study period, the pH was monitored daily, adjusting to 5.5 ± 0.5 using NaOH or HCI 0.1 M  $L^{-1}$  solution.

XXXV Congresso Brasileiro de

Ciência do Solo

The electrical conductivity of the nutrient solutions was in the range of 2.5 dS m<sup>-1</sup> and did not vary among the exchange period (15 days), indicating that the exchange of nutrient solution every 15 days was sufficient to maintain the electrical conductivity of 2.5 dS m<sup>-1</sup>. Distilled water was used for the replacement of the solution, with an oxygenated nutrient solution, and constantly renewed every 15 davs. Three tomato genotypes differing in phosphorus uptake efficiency were used: one efficient in absorbing phosphorus called "Globonnie" PI 121665 (efficient, crt/crt), another genotype considered sensitive in phosphorus uptake called TOM-584 (normal,  $crt^+/crt^+$ ) and an F<sub>1</sub>RC1 genotype, supposedly effective for absorbing P. All genotypes were tested under normal and hydroponic mediums deficient in P. The F1RC1 genotype arose from a cross between PI 121665 (efficient, crt/crt), and the  $(crt^{+}/crt^{+}),$ commercial strain TOM-584 and subsequent backcrosses to the commercial strain TOM-584 ( $crt^+/crt^+$ ). Each of these three genotypes was in a hydroponic system (Marques et al. 2007).

#### Imposition of stress and experimental design

The experimental design was a factorial design consisting of 3 (genotypes) x 4 dose of phosphorus (0.2; 30; 60 and 100 mg L<sup>-1</sup>) with four replications. The nutrient solution containing 60 ppm of P was calculated as recommended by Moraes and Furlani (1999). However, the solution deficient in P with 0.5 mg was based on Hochmuth et al. (1985), which was used by researchers for triage of tomatos for efficiency in P uptake and P deficient solution with 0.2 mg P (Marques et al. 2007). In this experimental stage, 20 plants were cultivated hydroponically for each type of nutrient solution as the concentration of P.

#### Analysis of root system morphology

The experiment was harvested at 150 days after transplanting. For the analysis of the root system, entire plants were collected (root system and aerial parts). The samples were submerged in ponds containing distilled water for 30 minutes. This aided the process of washing the roots. After the washing process, the plants were separated into root and shoot. The washed roots were stored in vials containing 70% ethanol solution to prevent dehydration and accommodate cold storage.

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For analysis of root system morphology: the system 2007 Pro "WinRhizo" was used (Regent Instruments, Sainte-Foy, QC, Canada), coupled to a professional Epson, Expression 10.000 XL scanner, (Epson America, Inc., USA) equipped with an additional light unit (TPU). A definition of 400 (dpi) was used for measurements of root morphology, as described by Bouma et al. (2000) and Costa et al. (2002). The roots were placed in an acrylic tub 7.87 inches wide by 11.81 inches long containing water.

The use of this accessory allowed obtaining three-dimensional images, also avoiding the overlapping of roots; readings were made on four plants per genotype. Then, the characteristics were determined as follows: length, surface area, volume and average diameter for tomato genotypes. To determine the dry weight of tomato plants, root, stem and leaflets were collected.

The roots were separated from aerial parts, through a cut in the plant lap, washed with running water to remove impurities. All components were dried in an oven at 60 Celsius, with forced ventilation, until constant mass. Other attributes involving morphological and dry mass data were as follows: specific root length (relationship between length and cm g) and root fineness (relationship between length and root volume cm cm<sup>-3</sup>).

#### **Statistical analysis**

Data were subjected to variance analysis and when significant differences occurred; the Scott-Knott test at 5% level of error probability was applied (Steel et al., 2006). Standard errors were calculated for all means. All statistical procedures were carried out with the SAS software (SAS, 1996).

#### **RESULTS & DISCUSSION**

#### **Root characteristics**

About the root morphology (Figures 1 A, B, C and D), was observed that Globonnie genotype had a significantly larger length root until the dose of 0.2 mg L<sup>-1</sup> of  $P_2O_5$  when compared to other genotypes of tomato (Tom-598 and Globonnie). However with the increasing of P doses, was observed that the length of roots was greater for the genotypes "Globonnie and F1" (Figure 1 A). This superiority with the



increasing of P doses of F1 may be explained by the gene interaction cross between the genotypes (Globonnie x TOM-598). To the root surface area (Figure 1 B) at dose of 0.2 mg  $L^{-1}$  of P<sub>2</sub>O<sub>5</sub> the Globonnie was higher in relation the others (TOM-598 and F1) in P limiting condition. However increasing the P doses the genotypes Globonnie and  $F_1$  were higher when compared with the genotype control, Tom 598, sensitive to lack of P. For the root volume (Figure 1 C) observed the same tendency to the Globonnie for greater production of root volume at the lowest dose 0.2 mg  $L^{-1}$  of  $P_2O_5$ , this concentration of P is considered limiting for tomato cultivation. These results confirm that the gene of Globonnie expressed the gene to resistance limitation P as compared with the F1 and TOM-598, for the same treatment. Already with increasing doses of P, the F1 and Globonnie were higher in the root volume increased when compared with TOM-598. The average diameter of root (Figure 1 D) at dose of 0.2 mg  $L^{-1}$  of P<sub>2</sub>O<sub>5</sub>, the genotype Tom-598 was higher than Globonnie and F<sub>1</sub>. However with the increasing of doses 30 and 60 mg  $L^{-1}$  of P<sub>2</sub>O<sub>5</sub>, the genotypes  $F_1$  and TOM 598 were higher than genotype Globonnie, when the tolerant lack of P was considered.

However, for the highest dose, 100 mg L<sup>-1</sup> of the  $P_2O_5$ , the Globonnie was higher than F1 and TOM-598. This results confirms that the "root Cotony" gene expressed efficiently in Globonnie to the lowest dose 0.2 mg L<sup>-1</sup> of  $P_2O_5$ , and the same is not happening with the F<sub>1</sub> hybrid.

By another aspect, in a higher concentration, but still deficient (30 mg L<sup>-1</sup> P<sub>2</sub>O<sub>5</sub>), the F1 and Globonie concentration didn't differ significantly and were significantly higher to the lineage Tom 589 indicating the presence of gene "Cottony root" for F1. In cultivation under concentration recommended for tomato (60 mg L<sup>-1</sup> P<sub>2</sub>O<sub>5</sub>), all genotypes registered increases in these same features but with the Tom-598 significantly lower.

All three evaluated characteristics (length, surface area and total volume) are important for phosphorus uptake. The root morphology ends up having a lot of importance on the efficient acquisition of phosphorus by plants because there is a relative immobility of P that makes their acquisition dependent on the further exploitation of the soil by the roots (greater length, volume and root surface area) (Ramaekers et al., 2010).

Previous studies of the tomato have identified a greater efficiency in the absorption of phosphorus. Hochmuth et al. (1985) identified in triage involving more than 200 accessions of tomato (*Solanum lycopersicon*), at least two highly efficient introductions in P extraction from poor nutrient solution in this nutrient. In one of these introductions

(PI 121665=Globonnie cultivate), the extraction efficiency of phosphorus was associated with a morphological characteristic associated to the roots when it was grown in nutrient solution with low levels of P. This characteristic, called "cottony root", proved simple inheritance (a recessive gene, referred to as CRT), and is associated with a number of roots, which can be observed under a microscope after staining with acetic carmine when plants are grown in solutions with low level (2 ppm) of P (Hochmuth et al., 1985). This result was not observed when higher concentrations of P (8 ppm) were used (Hochmuth et al., 1985). Imada et al. (2008) state that the surface area of the root, it is more related to the absorption of nutrients because a larger surface area can help the plant to obtain sources of nutrients that are deficient. Also an increase in the volume of roots when the nutrient concentration is the same throughout the root surface can lead to greater efficiency of nutrient absorption (Costa et al., 2002). In an unfavorable environment, is important for root growth (that the soil be explored) without loss in fertilization (Ryser, 2006). Theoretically, a higher specific root length is reflected in further exploration and acquisition of water and nutrients in the soil per unit of carbon invested (Ramaekers et al., 2010).

#### Chemical evaluation of plant tissue

For the concentration of phosphorus in the leaflet, stem and root on tomato genotypes (Figure 2 A, B and C) relative doses of P. Note that the highest concentration of P in the leaflet (Figure 8) was to Globonnie in the concentration of  $0.2 \text{ mg L}^{-1}$  of  $P_2O_5$ . Already with increasing concentration genotypes have different behaviors. At a dose of 30 mg L-<sup>1</sup> to phosphorus concentration was increased to F1 proportionally at a dose of 60 and 100 mg L<sup>1</sup> was the highest concentration Globonnie. However for the concentration of phosphorus in the stem (Figure 2 B) was superior Globonnie concentrations of 30, 60 and 100 mg  $L^{-1}$  of  $P_2O_5$ . However for the phosphorus concentration in the root (Figure 2 C) at a concentration of 0.2 mg  $L^{-1}$  Globonnie the P<sub>2</sub>O<sub>5</sub> had higher concentration than the other genotypes. With increasing doses of 60 and 100 mg  $L^{-1}$  of  $P_2O_5$ and Globonnie the F<sub>1</sub> genotypes were higher than those tomato genotypes.

One of the major changes in plans for the acquisition of phosphorus-deficient soils is to increase the exploitation of the soil through increased root growth and proliferation, mainly of those roots metabolically responsible for this function (roots of smaller diameter) (Zhang et al., 2010). Root growth, mainly fine and very fine roots observed in genotype Globonnie, can be connected to this root exploration in search of greater phosphorus acquisition. In contrast, significant differences



between genotypes (tolerant and non-tolerant) on the accumulation of dry matter in response to P deficiency have been reported for the maize crop (Li et al., 2007). Furthermore, the increase of the dry mass of roots by dry mass of the aerial part has been demonstrated as a major strategy in stress tolerance by phosphorus (Nielsen et al., 2001).

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Figure. 1 Root characteristics: (A) length, (B) surface area, (C) volume and (D) average diameter for genotypes de tomato (Globonnie, TOM 598, F<sub>1</sub>) with regard to different concentrations of phosphorus.

### Figures





Figure. 2 Phosphorus concentration root (A), stem (B), sheet (C) to tomato genotypes (Globonnie, TOM 598, F1) in relation to different concentrations of phosphorus.