

Differential responses of root system and gas exchange in contrasting tomato genotypes under phosphorus starvation

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Abstract

Phosphorus (P) is an essential macronutrient for the development of plants. Although it is not always required in larger amounts, its presence is often limited, since the Brazilian 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 contrasting tomato genotypes. The experimental design was a randomized block in the factorial scheme with three tomato genotypes (Globonnie, Tom-598 and F₁) and four phosphorus levels (0.2, 30, 60 and 100 mg L⁻¹) with four replications. Assessing the root morphology, it was observed that the Globonnie genotype has higher root length value in 0.2 mg L⁻¹ of P; however, with the increasing of P levels it was greater in F₁ genotype. This study concluded that Globonnie and F₁ presented better performances under phosphorus starvation, due to better root characteristics and gas exchange that promote higher P efficiency. Besides, they may further uptake and improve the use of the phosphorus. Here, we revealed a possible reduction of phosphorus and consequently production costs in tomato farming.

Keywords: *Solanum lycopersicon*, phosphorus, photosynthesis, root system.

Abbreviations: ATP_adenosine triphosphate, CO₂_carbon dioxide, E_transpiration rate, EAP_efficient absorption of phosphorus, gs_stomatal conductance, NADPH_nicotinamide adenine dinucleotide phosphate, P_phosphorus, Pi_inorganic phosphorus, Pn_photosynthesis, P₂O₅_Phosphorus pentoxide, RUBISCO_ribulose biphosphate carboxylase oxygenase, RuBP_Ribulose-1,5-bisphosphate.

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. This is the second vegetable most widely cultivated in the world, which only surpassed by the potato. The largest producer is China with 41.8 million tons at 8,700 million m² and a productivity of 48.0 tons per 10,000 m². Brazil produced 3.69 million tons. 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; IBGE, 2012).

Maximum productivity of the tomato has been associated with levels of fertilizer exceeding 300 kg of P₂O₅ per 10,000 m² (Barbosa, 1993), reaching up to 1200 kg of P₂O₅ per 10,000 m² (Filgueira, 2008). The increased efficiency in the absorption of phosphorus by the tomato could provide an appreciable reduction in the levels of phosphate fertilizers, as well as, allow harnessing 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 P₂O₅ per 10,000 m² in the utilization of nutrients by tomato plants would represent a savings of approximately 85.36 dollars per 10,000 m², totaling more than 4.69 million dollars on the national level of tomato production (Silva and 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 and 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 linked to phosphorus deficiency, called phosphorus use efficiency (Lynch, 1998; 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 exudation of mobilizing P components, or by the modifications of inorganic phosphorus (Pi) transporters in plasmatic membrane (Kochian et al., 2004). Additionally, it can also involve smaller P amounts required in cellular level, or a more efficient remobilization of P within plant (Yan et al., 2001).

Introductions of tomato plants with greater efficiency in the absorption of phosphorus have been identified. Study conducted by Hochmuth et al. (1985) evaluated more than 200 tomato types (*Solanum lycopersicon*), with at least two of these introductions being highly efficient in extracting P from the poor-P solution. In one of these introductions (PI 121665 was described as Globonnie cultivar), the efficiency in the extraction of phosphorus was associated with a morphological characteristic in the roots, when this type was cultivated in nutrient solution containing a low content of P. This characteristic, called "cottony root", showed up in simple heritage (a recessive gene, termed crt), and is associated with a large number of roots that can be observed in a microscope after being stained with carmine acetic, when plants are grown in solutions with low content (2 ppm) P, while that this response is not observed when higher concentrations of P (8 ppm) are used (Hochmuth et al., 1985). The availability of tomato germoplasm with efficiency in the extraction of phosphorus is associated with a morphological characteristic (large number of root hairs when cultivated under low levels of P), and the simple heritability to this trait (controlled by a recessive gene), makes the *Solanum lycopersicon* specie appropriate for the genetic improvement aiming improves the efficiency in the absorption of P. Thus, this research was aimed to evaluate the development of the root system, gas exchange and efficiency for absorption of phosphorus in contrasting tomato genotypes under phosphorus starvation.

Results

Modifications on root characteristics

It was observed that the root morphology of Globonnie genotype (Fig. 1) had higher root length value in 0.2 mg L⁻¹ of P (Fig. 1A), when compared to other tomato genotypes (Tom-598 and F₁). However, with the increasing of P levels, it was observed that the length of roots was greater in genotypes F₁ (Fig. 1A). This superiority with the increasing of P levels of F₁ may be explained by the gene interaction in genotypes Globonnie and Tom-598. For the root surficial area (Fig. 1 B) under P limiting condition of 0.2 mg L⁻¹ P, Globonnie was higher compared to others (Tom-598 and F₁). However, with increasing the P levels, the genotypes Globonnie and F₁ showed higher values, when compared to Tom-598, which is sensitive to lack of P. For the root volume (Fig. 1 C) we observed the same tendency to the Globonnie for greater production of root volume at the lowest level 0.2 mg L⁻¹ of P, considering this P concentration as limiting for tomato cultivation.

These results confirm that the gene of Globonnie is expressed further under P limitation to tolerate the limiting factor, compared with the F₁ and Tom-598, for the same treatment. Moreover, with increasing of P levels, the F₁ and Globonnie showed higher root volume, when compared with Tom-598. The root medium diameter (Fig. 1 D) at 0.2 mg L⁻¹ of P was higher in Tom-598 than Globonnie and F₁. However, at the highest level (100 mg L⁻¹ of the P) Globonnie presented superior than F₁ and Tom-598.

For the fineness of root (Fig. 2 A) at 0.2 mg L⁻¹ P the genotypes Globonnie and F₁ were higher than Tom-598. With the increasing of levels up to 60 mg L⁻¹ P, the F₁ and Globonnie were lower to control material and sensitive to the lack of P (Tom-598). Additionally, only on excess level (100 mg L⁻¹ of P), the F₁ hybrid was more efficient in producing fine root, when compared to the Tom-598 and Globonnie. For the specific root length (Fig. 2B) at 0.2 mg L⁻¹ of P, the

Globonnie was superior to F₁ and Tom-598. With the increasing of P level up to 30 mg L⁻¹, the F₁ hybrid was superior to the Tom-598 and Globonnie. For the level of 60 and 100 mg L⁻¹ P, the F₁ showed superiority to other genotypes. The highest value of specific length was observed in the genotype Globonnie in conditions of phosphorus deficiency at 0.2 mg L⁻¹ level. However, with increasing P levels, the F₁ expressed increase of this parameter.

For root diameter relative to specific length (Fig. 3) it was shown that at limiting level (0.2 mg L⁻¹ of P) the Globonnie showed superiority compared to F₁ and Tom-598 (Fig. 3 A). With proportional increasing of P levels (Fig. 3 A, B and C) the hybrid vigor of F₁ showed superiority, if compared with Tom-598 and Globonnie.

For surface area of root (Fig. 4 A and B), Globonnie (the tolerant genotype to P under 0.2 and 30 mg L⁻¹ levels of P) was better than F₁ and Tom-598. However, with increasing of P levels to 60 and 100 mg L⁻¹, the hybrid vigor of root was expressed higher in F₁ than Globonnie and Tom-598 (Fig. 4 C and D). These results are very important for selection of tolerant genotypes for P limitation in soils tropicalized countries. The Tom-598 is considered sensitive to the lack of P, which only presents superior with increase in concentration P solution.

For volume of root (Fig. 5 A, B, C), the Globonnie genotype was higher than F₁ and Tom-598 at levels of 0.2, 30 and 60 mg L⁻¹ of P. Only at highest concentration of P that hybrid vigor was expressed in F₁ to root volume relative to root diameter, proving that the Globonnie is expressing at root the larger area to result in greater efficiency in lack of P. Noting that at all concentrations the genotype Tom-598 showed the least surface area of fine roots. Relative to volume by diameter classes, the genotype Globonnie presented, under lack P, the greater volume of fine roots and reduction for thick roots.

Moreover, the genotype Tom-598 produce greater amount of thick roots, only responsible for supporting and fixing of plant. In short, Globonnie showed greater length, volume and surface area of fine roots and higher specific length and thickness roots. However, relative the root structure of fine roots, which is important in absorption of P values were below (Fig. 2).

The dry matter of leaf (Fig. 6) was greater in Globonnie at lowest level of P (0.2 mg L⁻¹) when compared to F₁ and Tom-598. The F₁ hybrid was superior in production of leaf with the increased P level up to 30 mg L⁻¹ when compared to Tom-598 and Globonnie. However, at the optimal level (60 mg L⁻¹ of P) and the excess level (100 mg L⁻¹ of P) no significant difference was observed between the F₁, Tom-598 and Globonnie genotypes.

The production of stem was superior in Globonnie at 0.2 mg L⁻¹ of P, comparing to other genotypes. Applying the maximum level of P (100 mg L⁻¹), the Tom-598 was superior to F₁ and Globonnie. However, the dry matter of root was superior in Globonnie at 60 mg L⁻¹ of P. With the excess P level (100 mg L⁻¹ of P) Tom-598 was superior to the other genotypes (F₁ and Globonnie). It was observed that P deficiency caused reduction in dry matter, perceiving that the genotype Globonnie produced more roots than leaf area under P deficiency (Fig. 6 A, B and C).

Genotype responses linked to leaf gas exchange

The photosynthesis level was higher in Globonnie at the limiting P (0.2 mg L⁻¹) (Fig. 7) compared to F₁ and Tom-598. At the 30 mg L⁻¹ of P the F₁ exhibited superior photosynthetic

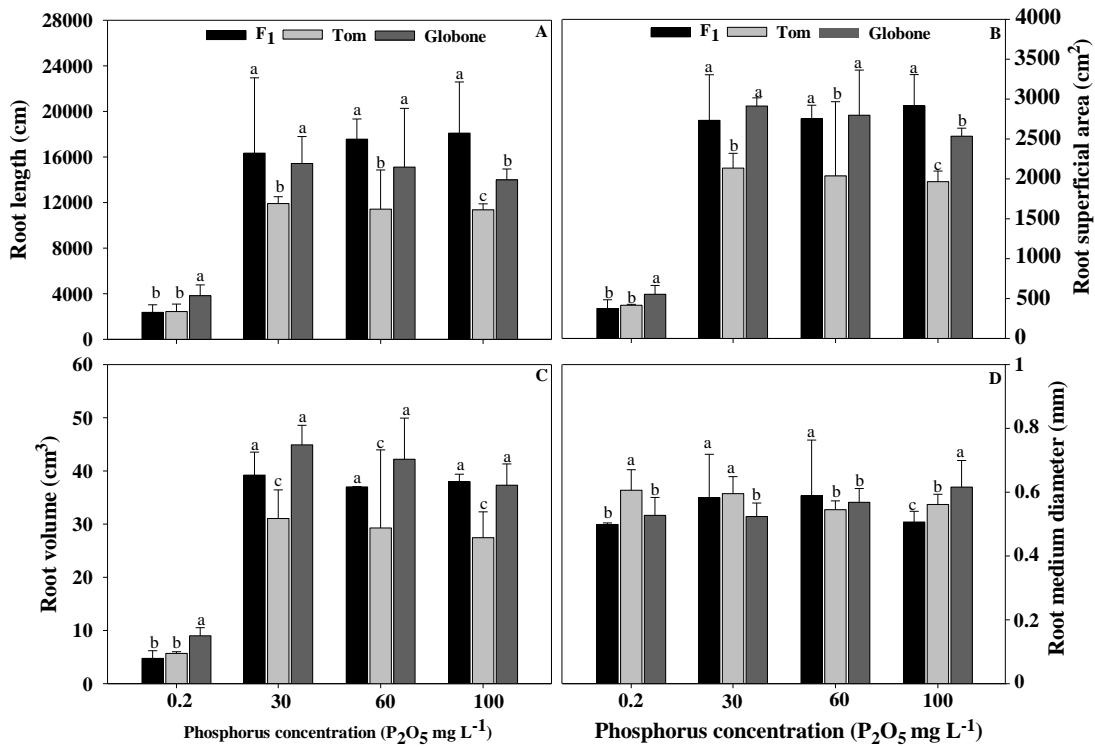


Fig 1. Length (A), surface area (B), volume (C) and average diameter (D) in tomato genotypes (Globonnie, Tom-598 and F₁) subjected to different phosphorus concentrations (0.2; 30; 60 and 100 mg L⁻¹).

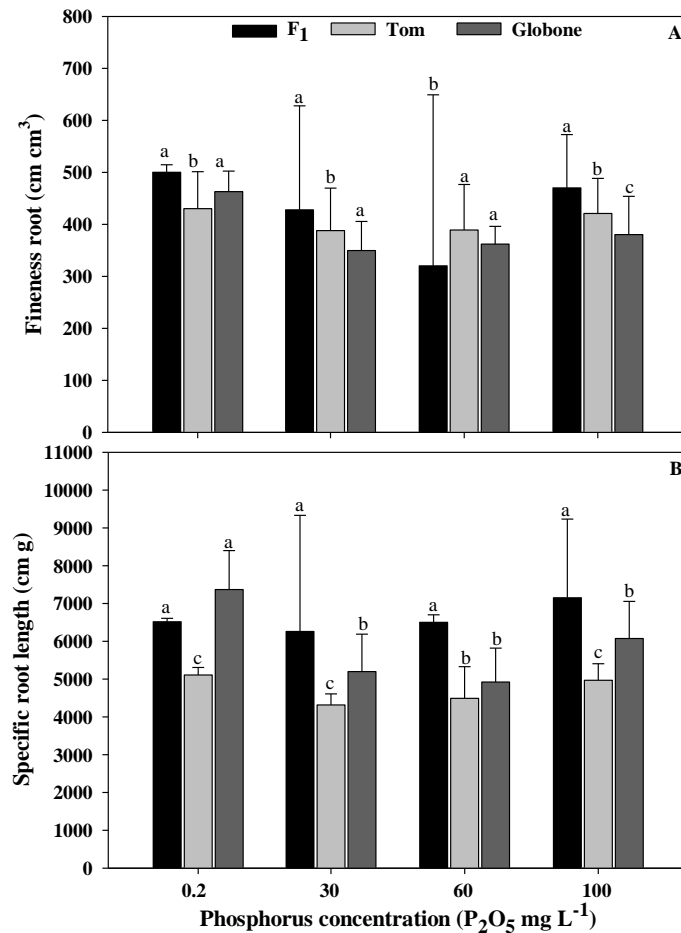


Fig 2. Fineness root (A), specific root length (B) in tomato genotypes (Globonnie, Tom-598 and F₁) subjected to different phosphorus concentrations (0.2; 30; 60 and 100 mg L⁻¹).

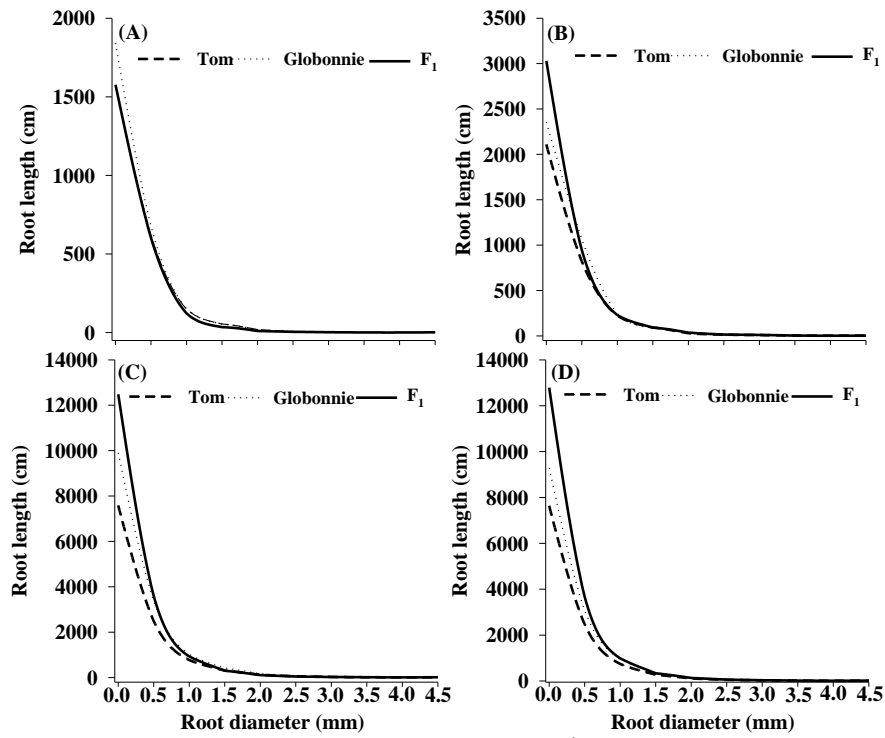


Fig 3. Root relative length under 0.20 (A); 30 (B); 60 (C) and 100 (D) (mg L^{-1}) into root diameter of tomato genotypes (Globonnie, Tom-598 and F₁).

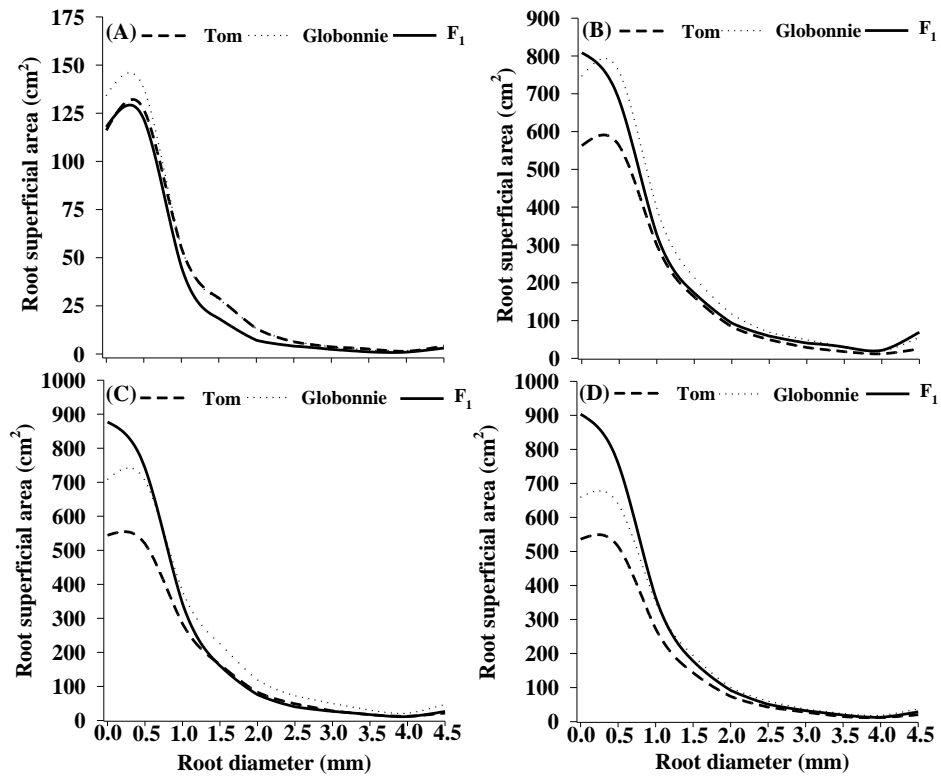


Fig 4. Root relative superficial area under 0.20 (A); 30 (B); 60 (C) and 100 (D) (mg L^{-1}) into root diameter of tomato genotypes (Globonnie, Tom-598 and F₁).

rate. This is still considered as limiting P level recommended for tomato culture. However, with increasing the level to 60 and 100 mg L⁻¹ the superiority of P Globonnie was expressed in most photosynthetic rate compared to F₁ hybrid and Tom-598. The rate of leaf transpiration (Fig. 7 B) was higher in Globonnie with increasing the P level, compared to F₁ and Tom-598. However, the stomatal conductance (Fig. 7 C) in leaf was noted independent of P. The stomatal conductance of Globonnie increased with increasing levels of P, showed superiority to F₁ and Tom-598. It is observed that the higher photosynthetic rates, stomatal conductance and transpiration occur at higher concentrations of applied phosphorus. We noted that Globonnie showed better results at different concentrations of phosphorus, when compared with the other genotypes of tomato (Tom-598 and F₁). Interestingly, the Globonnie expressed tolerance trait to the deficit of phosphorus having the highest rates of photosynthesis, transpiration and stomatal conductance at lower concentration of phosphorus, showing its superiority in metabolic activity being a possible adaptation of the plant to limited amount of phosphorus.

Concentration and absorption of phosphorus in plant tissue

The P concentrations in the leaf, stem and root of tomato genotypes are presented in Fig 8A, B and C, being described the higher P concentration in leaves of Globonnie under P concentration of 0.2 mg L⁻¹ of (Fig. 8 A). However, the increased P concentrations produced different behaviors in the genotypes. After 30 and 60 mg L⁻¹ P application, the phosphorus concentrations were higher in F₁; however, at 100 mg L⁻¹ P it was highest in Globonnie. The P concentration in stem (Fig. 8 B) was higher in Globonnie at the 30 and 60 mg L⁻¹ of P. The P concentration was higher in root of Globonnie (Fig. 8 C) under 0.2 mg L⁻¹, compared to other genotypes. With increase of P level to 100 mg L⁻¹, the Globonnie and F₁ genotypes showed higher values than other tomato genotypes.

The efficient absorption of phosphorus (EAP) of genotypes and phosphorus levels are shown in Fig. 9. The EAP of leaves (Fig. 9 A) was higher (efficient) in Globonnie under 0.2 mg L⁻¹ P, compared to others. At concentrations of 60 and 100 mg L⁻¹ P, the Globonnie was more efficient in use of this mineral. For the EAP in stem (Fig. 9 B), Globonnie also had higher efficiency than other genotypes at the lowest P level. With increasing concentrations to 30, 60 and 100 mg L⁻¹ of P, genotypes showed differential EAP especially Tom-598 with the highest efficiency. For the EAP of roots (Fig. 9 C), the genotype Tom-598 presented more efficiency under concentrations of 60 and 100 mg L⁻¹ P. This greater efficiency of Tom-598 in root may be associated with the genotype that present a thicker root, when compared to the root Globonnie, which is characterized by the production of fine roots (Fig 1).

Discussion

All three evaluated root 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 different groups of genotypes involving more than 200 accessions of tomato (*Solanum lycopersicon*), at least two highly efficient genotypes in P extraction from poor nutrient solution in this nutrient. In one of these introductions (Globonnie genotype), 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) stated that the surface area of the root is more related to the absorption of nutrients because a larger surface area can help 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). Therefore, the roots of tomato plants have lower expense of linear construction (grams carbon/length), because they presented greater specific length. On the other hand, increasing the thickness of the root reflects a greater exploration of the soil by functional roots, because the efficiency of the roots with high values of specific length can vary between species, possibly because the very fine roots have reduced longevity, which may hinder root functioning. Thus, separating specific length in thick root component (length/volume) and density of tissue roots (root mass /volume) can provide information on the relationship between root characteristics and development strategies of plants (Ryser and Lambers, 1995).

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, with mainly very fine roots observed in genotype Globonnie, can be connected to this root exploration in search of greater phosphorus acquisition.

One of the possible reasons that tomato genotypes have shown greater length, surface area and volume of fine roots (Fig. 3) is favoring the exploration and acquisition of water and nutrients, because the fine roots have an important role in these functions (Eissenstat et al., 2000).

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).

Root growth is less affected in phosphorus deficient plants than the aerial part, often resulting in an increase in root dry mass/aerial part. This increase in the ratio of root/aerial part in plants deficient in phosphorus is correlated with an increase in carbohydrate partitioning in root, indicated

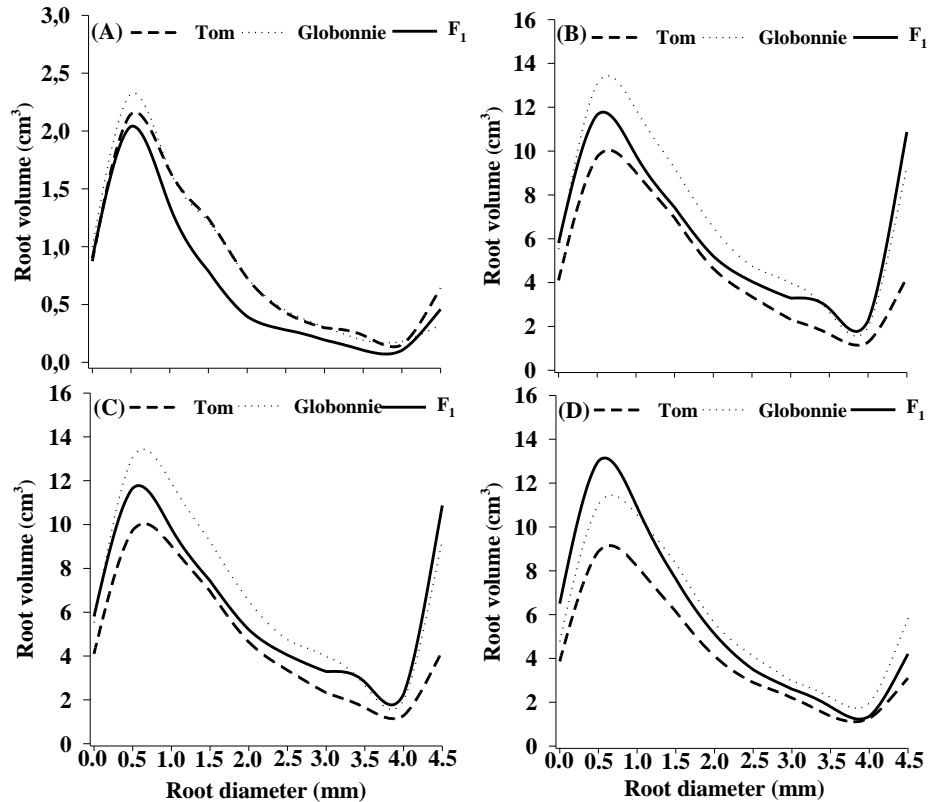


Fig 5. Root relative volume under 0.20 (A); 30 (B); 60 (C) and 100 (D) (mg L^{-1}) of phosphorus in tomato genotypes (Globonnie, Tom-598 and F_1).

by a sharp increase in sucrose content in roots (Marschner, 1995).

The plants well supplied with phosphorus stock store the excess inorganic phosphorus (Pi) in cells (85 to 95%) mainly in the vacuole or non-metabolic manner of storage. This vacuolar Pi is used to maintain the levels of cytoplasmic Pi, which may change due to different metabolic activities and / or the variation in the external supply means (Jain et al., 2007). The Pi absorbed by plants can remain in its inorganic form or can be incorporated into organic molecules, such as the formation of simple phosphate ester or by binding to other Pi by connecting with energy, forming pyrophosphate, such as the example of the ATP molecule.

Briefly, the phosphorus (P) is involved in several cellular roles. It acts as a structural element of molecules such as nucleic acids, proteins, coenzymes; participates in cellular processes such as energy storage and transfer, cell division and growth; has regulatory function of metabolic pathways in the cytoplasm and in chloroplasts, acting directly in the processes of photosynthesis and respiration (Marschner, 1995).

For gas exchange, it was noted that Globonnie presented higher value. Phosphorus is connected, or regulates the biosynthesis of starch/sucrose, regulates the activation of RUBISCO and the provision of energy by ATP in the Calvin cycle. In this manner a smaller amount of phosphorus leads to an inhibition of these processes. Tolerant genotypes as Globonnie have higher photosynthesis at low concentrations of P. Silva et al. (2010) observed a decrease in stomatal conductance of coffee plants under conditions of phosphorus deficiency. This occurred when comparing the highest concentrations of P with the smallest, yet Globonnie kept higher conductance compared to the other genotypes at low P concentrations, facilitating gas exchange. In the

photosynthetic apparatus, phosphate plays a regulatory role in starch/sucrose biosynthesis and RUBISCO activation, and a role in metabolites as it is used to phosphorylate intermediates of the Calvin Cycle, and in energy availability (ATP and NADPH) (Sawada et al., 1992). P limitation may affect photosynthesis through changes in the activity of Calvin cycle enzymes, RuBP regeneration, and/or RUBISCO activity (Jacob and Lawlor, 1991). As with nitrogen, a feedback limitation of photosynthesis has been suggested as a cause of decreased CO_2 at low P supply (Pieters et al., 2001). However, for tomato plants, a decrease in starch accumulation with decreasing P supply suggested that the production, rather than the utilization of photosynthates, was limiting (De Groot et al., 2001).

Phosphorus deficiency in tomatoes leads to decreased gas exchange parameters (photosynthesis). In a tomato cultivar (*Solanum lycopersicon*. cv. Capita), De Groot et al. (2003) observed that phosphorus deficiency affects the capacity of carboxylation of CO_2 and thus decreases the capacity to produce photo-assimilates.

Materials and Methods

Experiment localization and climatic conditions

The experiment was conducted in a greenhouse in 2013 in the Department of Soil Science at the Federal University of Lavras-MG, 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.

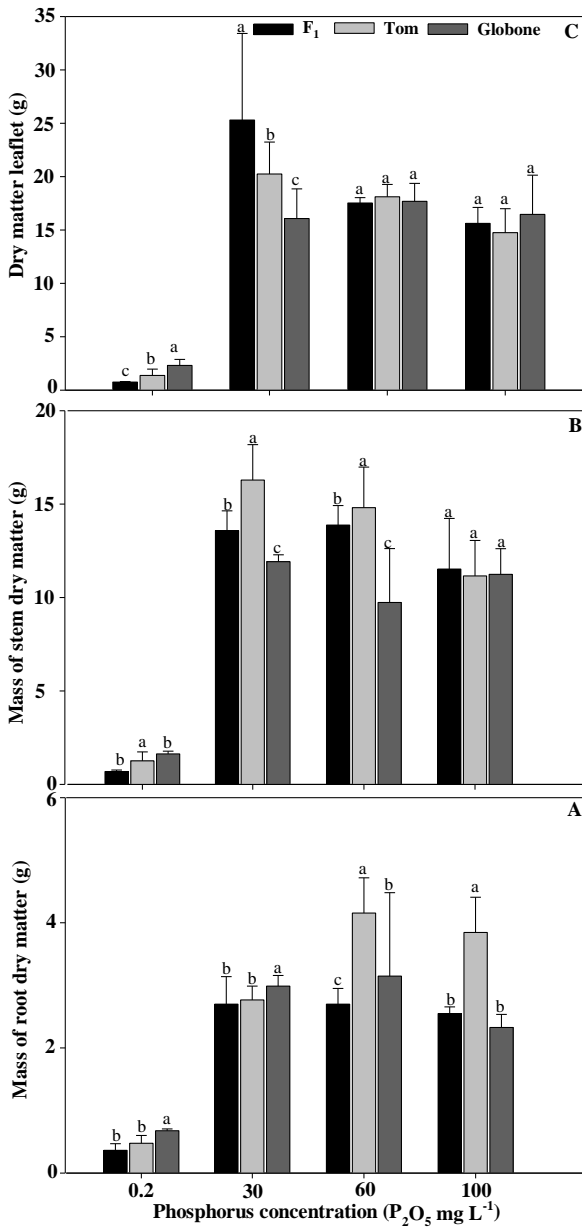


Fig 6. Root dry matter (A), stem dry matter (B) and leaf dry matter (C) in tomato genotypes (Globonnie, Tom-598 and F₁) submitted to different phosphorus concentrations (0.2; 30; 60 and 100 mg L⁻¹).

Plant material

Three tomato genotypes differing in phosphorus uptake efficiency were used, being one efficient in absorbing phosphorus called Globonnie or PI 121665 (efficient, crt/crt), another genotype considered sensitive in phosphorus uptake called Tom-598 (normal, crt⁺/crt⁺) and F₁. This genotype was created from a cross between Globonnie (efficient, crt/crt), and the commercial strain Tom-598 (crt⁺/crt⁺). All three genotypes were evaluated in a hydroponic system (Marques et al., 2007) under different phosphorus levels.

Growth conditions of genotypes

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

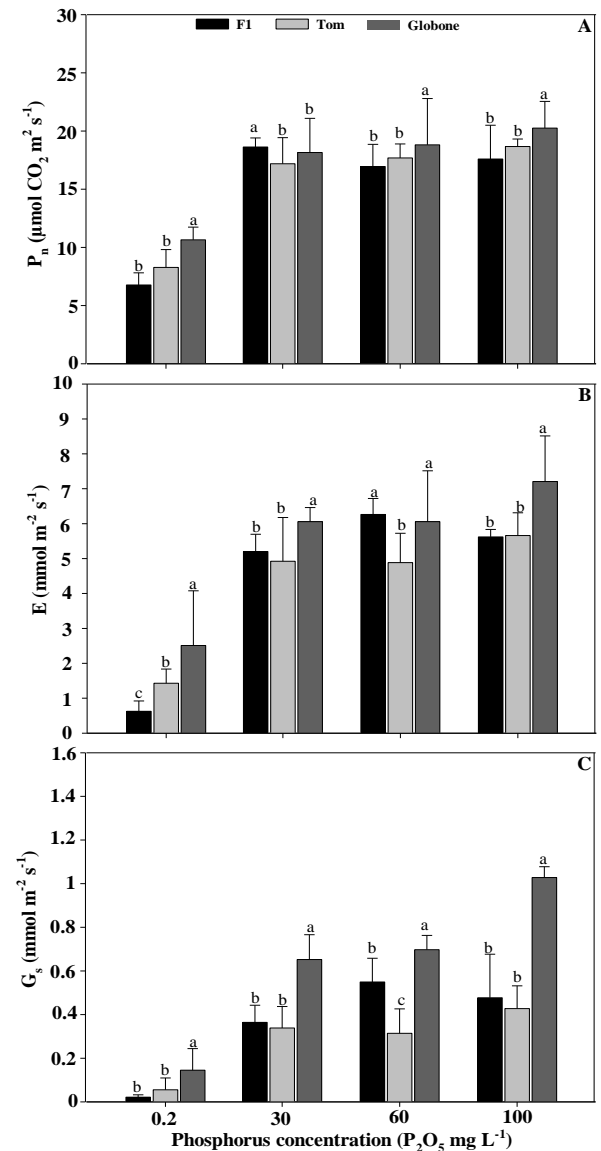


Fig 7. Photosynthesis (A), transpiration (B) and stomatal conductance (C) in tomato genotypes (Globonnie, Tom-598 and F₁) submitted to different phosphorus concentrations (0.2; 30; 60 and 100 mg L⁻¹).

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 was placed in plastic containers (virgin polypropylene) with a capacity of 10 L. The concentration was diluted to 0.25 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 HCl 0.1 M L⁻¹ solution.

The electrical conductivity of the nutrient solutions was in the range of 2.5 dS m⁻¹ 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

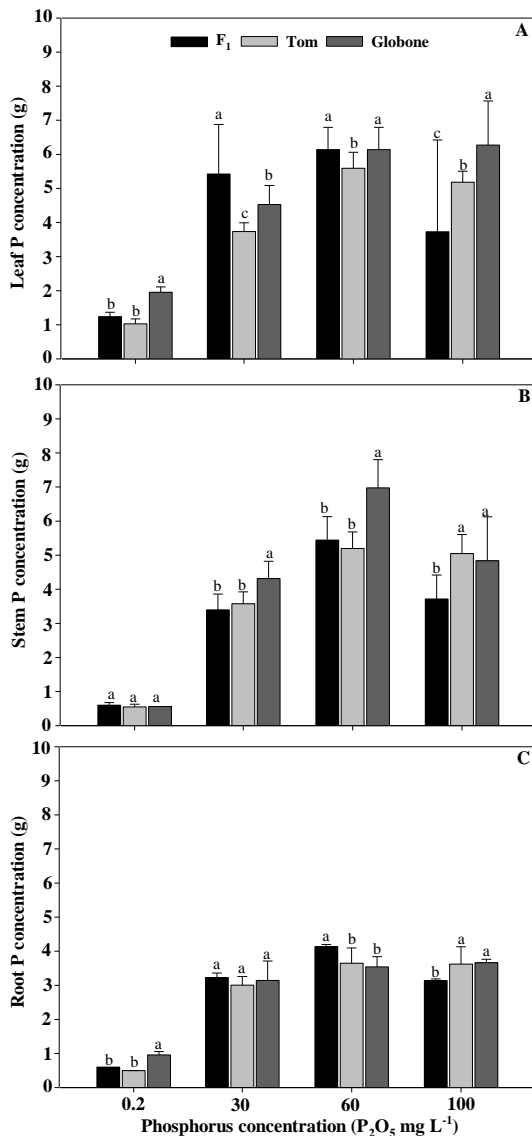


Fig 8. Phosphorus concentration leaf (A), stem (B), root (C) in tomato genotypes (Globonnie, Tom-598 and F₁) submitted to different phosphorus concentrations (0.2; 30; 60 and 100 mg L⁻¹).

electrical conductivity of 2.5 dS m⁻¹. Distilled water was used for the replacement of the solution, with an oxygenated nutrient solution, and constantly renewed every 15 days.

Application of stress and experimental design

The experimental design was a factorial design consisting of three genotypes (Globonnie, Tom-598 and F₁) combined with four phosphorus levels (0.2; 30; 60 and 100 mg L⁻¹ P₂O₅), composed by 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 made according to Hochmuth et al. (1985), which was used by researchers for triage of tomato 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.

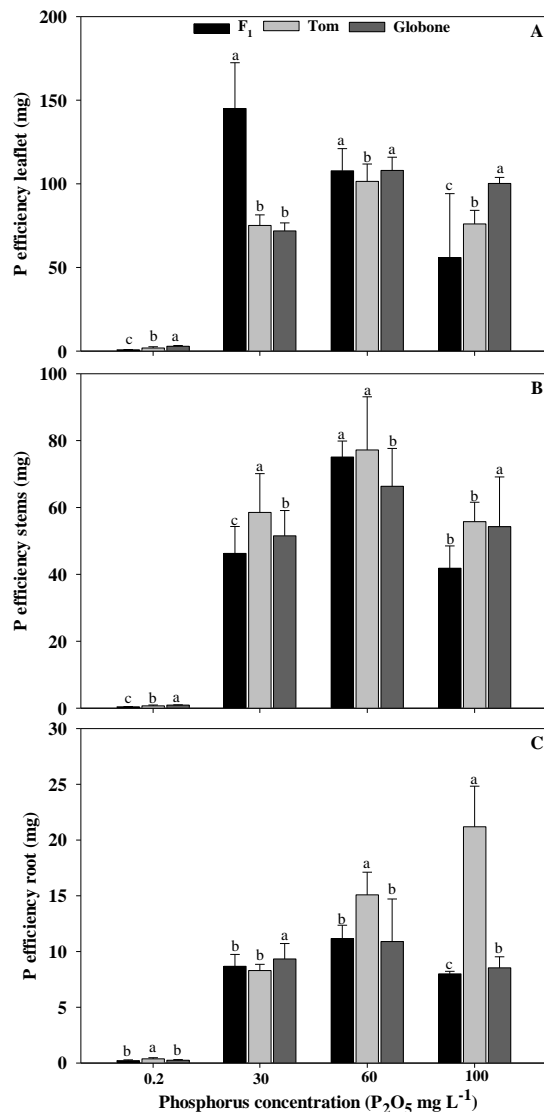


Fig 9. Phosphorus absorption efficiency in leaf (A), stem (B), root (C) in tomato genotypes (Globonnie, Tom-598 and F₁) submitted to different phosphorus concentrations (0.2; 30; 60 and 100 mg L⁻¹).

Analysis of root system morphology and dry matter characteristics

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 stored in cold storage.

For analysis of root system morphology, the system 2007 Pro WinRhizo was used (Regent Instruments, Sainte-Foy, QC, Canada), coupled with 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 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 leaf 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 °C, 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⁻³).

Leaf gas exchange

For the determinations of gas exchange, a measurement at 70 days was performed after transplant using a portable photosynthesis system (IRGA, Model LI-6400XT, Li-Cor, Lincoln, Nebraska, USA). All measurements were performed in the morning between 9:00 am and 11:00 am on fully expanded terminal leaf. The parameters measured were leaf photosynthesis (P_n), stomatal conductance (g_s), and transpiration rate (E). Measurements were made on a leaf area of 6 cm² with a flow of air in the chamber with a CO₂ concentration of 380 $\mu\text{mol mol}^{-1}$. The air was collected outside the greenhouse and transported into a protective gallon and then pumped into the chamber. One photon flux density (PPFD) of 500 $\mu\text{mol m}^{-2} \text{s}^{-1}$ of a light source red-blue LED was used. Other environmental factors such as air humidity and temperature were not controlled allowing for natural variation.

Nutritional determination in tissues

Samples of root, stem and the leaf were analyzed for mineral composition to 105 days after planting. The analyses were conducted in the laboratory of mineral nutrition of plants, Department of Soil Science-UFLA methodologies as described by Malavolta et al. (1997).

Absorption efficiency of phosphorus

The absorption efficiency of P is defined as the product of efficiencies of acquisition and internal use of P, corresponding to the dry mass of the leaf, stem and roots produced by 2.2 lb of phosphorus (Moll et al., 1981).

Statistical analysis

Data were subjected to variance analysis and when significant differences occurred; 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).

Conclusion

Based on this research, it can be concluded that the Globonnie genotype showed superiority in root characteristics, gas exchange and efficiency in the phosphorus absorption considering that the gene “cottony root” was expressed in the genotype. Globonnie and F₁

presented better performances under phosphorus starvation, based in root characteristics and gas exchange that promote higher P efficiency, besides improving the uptake and use of the phosphorus. Therefore, this research reveals a possibility of reduction of phosphorus application and consequent reduction of costs linked to tomato production.

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References

- Alvarez JM, Rocha JF, Machado SR (2008) Bulliform cells in *Loudetiopsis chrysothrix* (Nees) Conert and *Tristachya leiostachya* Nees (Poaceae): structure in relation to function. *Braz Arch Biol Tech.* 51:113-119.
- Jain A, Poling DM, Karthikeyan SA, Blakeslee JJ, Wendy PA, Titapiwatanakun B, Murphy SA, Raghothama GK (2007) Differential effects of sucrose and auxin on localized phosphate deficiency-induced modulation of different traits of root system architecture in *Arabidopsis*. *Plant Physiol.* 144:232-247.
- Bouma TJ, Nielson LK, Koutstaal SAB (2000) Sample preparation and scanning protocol for computerized analysis of root length and diameter. *Plant Soil.* 218:185-196.
- Costa CLM, Dwyer X, Zhou P, Dutilleul C, Hamel ML, Reid LD (2002) Root morphology of contrasting maize genotypes. *Agron J.* 94:96-101.
- De Groot CC, Marcelis MFL, Van Den Boogaard R, Kaiser MW, Lambers H (2003) Interaction of nitrogen and phosphorus nutrition in determining growth. *Plant Soil.* 248:257-268.
- De Groot CC, Marcelis MFL, Van Den Boogaard R, Lambers H (2001) Growth and dry mass partitioning in tomato as affected by phosphorus nutrition and light. *Plant Cell Environ.* 24:1309-1317.
- Eissenstat DM, Wells EC, Yanai DR (2000) Building roots in a changing environment: implications for root longevity. *New Phytol.* 147:33-42.
- FAO (2012) Food and agriculture organization of the United Nation. In: FAO statistical database, 2012. <http://www.fao.org>.
- Filgueira FAR, Obeid CP, Marais JH, Santos VW, Fontes RR (1999) Staked tomato. In: Ribeiro CA, Guimarães GTP, Avarez HV (Eds). Recommendations for the use of lime and fertilizers in Minas Gerais, 5th Approach. CFSEMG, Viçosa, 207-208.
- Hochmuth GJ, Gabelman HW, Gerloff CG (1985) The gene affecting tomato root morphology. *Hortscience.* 20:1099-1101.
- Imada S, Yamanaka N, Tamai S (2008) Water table depth effects *Populus alba* fine root growth and whole plant biomass. *Funct Ecol* 22:1018-1026.
- IBGE (2013) Brazilian Institute of Geography and Statistical. In: Systematic survey of agricultural production. Available in: <http://www.ibge.gov.br>.
- Jacob J, Lawlor WD (1999) Dependence of photosynthesis of sunower and maize leaves on phosphate supply, ribulose-1,5-biphosphate carboxylase/oxygenase activity and ribulose-1,5-bisphosphate pool size. *J Plant Physiol.* 98:801-807.

- López-Bucio J, Cruz-Ramirez A, Herrera-Estrela L (2003) The role of nutrient availability in regulating root architecture. *Curr Opin Plant Biol.* 6:280-287.
- Lynch J (1998) The role of nutrient efficient crops in modern agriculture. *J Crop Prod.* 1:241-264.
- Kochian LV, Hoenkenga AO, Piñeros AM (2004) How do crop plants tolerate acid soils? Mechanisms of aluminum tolerance and phosphorus efficiency. *Ann Rev Plant Biol.* 55:459-469.
- Marschner H (1995) Mineral nutrition in plants 2nd edn. Academic Press, San Diego.
- Marques DJ, Silva EC, Moraes LFR, Caetano E, Silva AR, Araújo HT, Abreu MV, Maciel AMG (2007) Introgression of resistance and characterization of the gene phosphorus deficiency in tomato. In: IV Brazilian Congress of Plant Breeding. Summaries. São Lourenço, Brazil: ABMP (CD-ROM).
- Malavolta E, Vitti CG, Oliveira AS (1997) Evaluation of nutritional status of plants: principles and applications. 2nd edn. Potafos, Piracicaba.
- Moraes CAG, Furlani RP (1999) Cultivation of fruit vegetables in hydroponics in greenhouses. *Inf Agropec.* 20:105-113.
- Moll RH, Kamprath JE, Jackson AW (1981) Analysis and interpretation of factors which contribute to efficiency of nitrogen utilization. *Agron J.* 74:562-564.
- Nielsen KI, Eshel A, Lynch PJ (2001) The effect of phosphorus availability on the carbon economy of contrasting common bean genotypes. *J Exp Bot.* 52:329-339.
- Ramaekers L, Remans R, Rao MI, Blair WM, Vanderleyden J (2010) Strategies for improving phosphorus acquisition efficiency of crop plants. *Field Crops Res.* 117:169-176.
- Ryser P (2006) The mysterious root length. *Plant Soil.* 286:1-6.
- Ryser P, Lambers H (1995) Root and leaf attributes accounting for the performance of fast- and slow-growing grasses at different nutrient supply. *Plant Soil.* 170:251-265.
- SAS Institute 1996. SAS/STAT User's Guid, Version 6. 12 SAS Institute, Cary.
- Sawada S, Usuda H, Tsukui T (1992) Participation of inorganic orthophosphate in regulation of the ribulose-1,5-biphosphate carboxylase activity in response to changes in the photosynthetic source-sink balance. *Plant Cell Physiol.* 33:943-949.
- Silva L, Marchiori REP, Maciel PC, Machado CE, Ribeiro VR (2010) Photosynthesis, water relations and growth of young coffee plants in relation to phosphorus availability. *Pesq Agropec Bras.* 45:965-972.
- Silva EC, Maluf RW (2012) Hydroponic technique for screening tomato genotypes on the efficiency of phosphorus uptake. *Hortic Bras.* 30:317-321.
- Stauffer MD, Sulewski G (2004) Phosphorus: Essential for Life. In: Yamada, T, Abdalla SRS (Eds.). Phosphorus in Brazilian Agriculture. Potáfos, Piracicaba, 1-12.
- Schröder JJ, Smit LA, Cordell D, Rosemarin A (2011) Improved phosphorus use efficiency in agriculture: A key requirement for its sustainable use. *Chemosphere.* 84:822-831.
- Steel RGD, Torrie JH, Dickey DA (2006) Principles and procedures of statistics: a biometrical approach, 3rd edn. Academic Internet Publishers, Moorpark.
- Pieters AJ, Paul JM, Lawlor WD (2001) Low sink demands limits photosynthesis under Pi deficiency. *J Exp Bot.* 52:1083-1091.
- Yan XL, Liao H, Trull CM, Beebe ES, Lynch PJ (2001) Induction of a major leaf acid phosphatase does not confer adaptation to low phosphorus availability in common bean. *J Plant Physiol.* 125:1901-1911.
- Zhang F, Shen J, Zhang J, Zuo Y, Li L, Chen X (2010) Rhizosphere processes and management for improving nutrient use efficiency and crop productivity: implications for China. *Adv Agron.* 107:1-32.