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Colegio de Postgraduados



# Root density and accumulation of Myrobalan plum tree grafted with Methley Japanese plum

González-Pérez J. S.<sup>1</sup>; Becerril-Román A. E.<sup>1\*</sup>; Quevedo-Nolasco A.<sup>1</sup>; Jaén-Contreras, David<sup>1</sup>; Velasco-Cruz C.<sup>1</sup>

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## ABSTRACT

The following variables were analyzed at different soil depths, during one phenological cycle: root density, root dry matter percentage, root accumulation, both growing and absorbing roots, and intermediate and conducting roots. The aim of the study was to determine the root phenology of Myrobalan plum tree grafted with Methley Japanese plum. A quota sampling was used to select five trees from the experimental orchard. Within the volume of soil adjacent to tree's roots, 330 cm<sup>3</sup> of soil were sampled and collected each month, in order to identify root type, quantify their fresh and dry weight, and carry out statistical analyses. The highest densities of growing and absorbing roots were observed at 0-25 cm soil depth during the phenological cycle. A higher density of intermediate and conducting roots was recorded at 25-50 cm soil depth, just at the beginning of the ecodormancy. The highest root accumulation was recorded when moisture and soil temperature were not optimal but the cultivar did not record a significant vegetative and reproductive growth.

**Keywords:** *Prunus cerasifera*, *Prunus salicina*, root density, root accumulation.

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## INTRODUCTION

In the face of climate change and the limitation of natural resources, an interest has arisen to increase agricultural production without increasing crop areas (Lynch, 2007; Gregory *et al.*, 2013). Fruit trees have growing, absorbent, intermediate, and conducting roots (Kolesnikov, 1971), and some of the root functions include: water and nutrient uptake, reserve storage, and substance synthesis (Becerril *et al.*, 2009). Moreover, the roots of fruit species can be found at a soil depth of 10-90 cm, and 30 cm away from the trunk, depending on the vigor of the rootstock, biomass distribution (Ovando *et al.*, 1993), rootstock-cultivar combination, soil characteristics, orchard management (Gutiérrez *et al.*, 2006), and mulching and irrigation system (Wei *et al.*, 2002). Furthermore, deciduous fruit trees have three fluxes of root growth in each phenological cycle: 1) before bud swelling; 2) during flowering; 3) and after leaf abscission (Ryugo, 1988; Shaw, 1998). Specifically, the roots of fruit trees from the *Prunus* genus requires between 40 and 50 cm of soil depth (Silva and Alonso, 1976), and they tend to grow right under soil surface (Agusti, 2004). As a result of its successful growth in a wide range of soils (including sandy soils), the Myrobalan

plum tree is frequently used as rootstock for plum cultivars (Popescu and Caudullo, 2016). Nevertheless, unlike the canopy management, the management of the root system of fruit trees is still limited, because of the roots biology has not been entirely understood, and sustainable soil management has not been adapted to improve media conditions related with plant development (Becerril *et al.*, 2009), to achieve an efficient use of soil resources (Thorup and Kirkegaard, 2016) and optimize fertilization (Salazar *et al.*, 2015). Therefore, the phenology of the root as well as the rootstock-cultivar interactions must be studied, in order to determine if root development is a limiting factor and, if that is the case, how to overcome it (Thorup and Kirkegaard, 2016). Consequently, the objective of this study was to determine the root phenology of the Myrobalan plum tree during a phenological cycle.

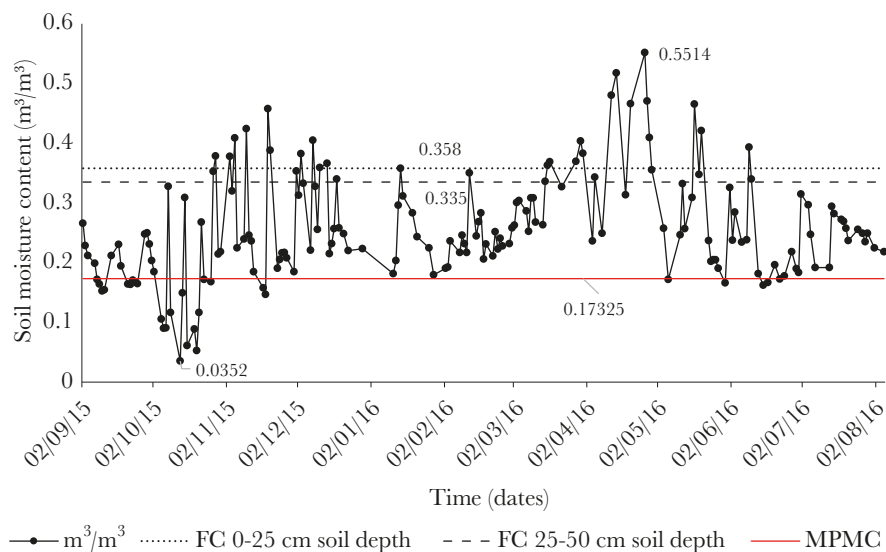
## MATERIALS AND METHODS

**Experimental site and plant material.** The research was carried out from September 2015 to August 2016, in the Colegio de Postgraduados (19° 29' N, 98° 54' W, and 2,252 m.a.s.l.). The climate is semi-dry temperate, with warm summers, and a 15.2°C mean annual temperature. The region has summer rains, with a 590 mm mean annual precipitation and a low percentage of winter rains (5%). It has a low thermal oscillation and its annual temperature progress is similar to the Ganges (García, 1988). The orchard is planted with Methley Japanese plum grafted on Myrobalan plum tree. The trees were 4 years-old (first year of a consistent fruit production, with a mean of 14.5 kg tree<sup>-1</sup>). They were planted in a 4×4 m square planting pattern, with a Tatura trellis. The soil is sandy loam, with 1.95% organic matter, 6.68 pH, and a 46.05% total porosity. Its color is brownish-grey when it is dry and darker grey when it is wet.

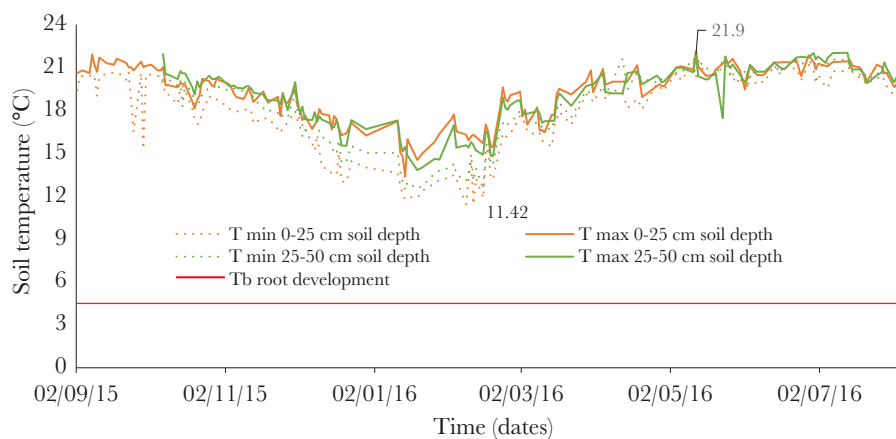
Suckers were removed continuously, and weedings were made with a manual weed remover or a chain brush cutter. Empirical fertilizations were carried out evenly spreading granular N, P, and K fertilizer around the trunk of the trees and under organic soil covers. The last ones, were permanently maintained. Soil moisture was monitored using a HH2 Delta-T Devices<sup>®</sup> (UK); in addition, water was sprayed, using two mini sprinklers per tree (water use: 16 L h<sup>-1</sup> 40 cm<sup>-1</sup> h<sup>-1</sup>), in order to maintain the moisture levels of the soil permanently close to field capacity (FC) (Figure 1).

Minimum temperature (MinT) and maximum temperature (MaxT) of the soil (Figure 2) were recorded daily, at 7:00 am and 3:30 pm, respectively. Both temperatures were measured at a depth of 25 and 50 cm, using a 1,450 mm digital thermometer with a fixed long probe (model 91000-021, Alla France<sup>®</sup>, France).

**Treatments and experimental design.** In the first study the treatments were the soil depths; a set of roots extracted at a depth of 0-25 and 26-50 cm, in the same sampling date, were analyzed. In the second study the treatments were certain phenological stages; a set of roots extracted at the same depth, during different phenological stages, were analyzed. A completely randomized experimental design was used. Through a quota sampling, five trees (EU) were selected for this study. The trees were 1.8 m tall and had a 1.7 m canopy, they were healthy and did not suffer nutritional disorders. Monthly, 330 cm<sup>3</sup> root samples were extracted with a California sampler (Figure 3A), at a depth of 0-25 and 26-50 cm, at 40 cm away from the trunk following the cardinal points (Figure 3B). The soil particles



**Figure 1.** Soil moisture content of an orchard with drip irrigation system, planted with Myrobalan plum tree grafted with Methley Japanese plum.



**Figure 2.** Temperature at a 25 and 50 cm depth in the soil of an orchard planted with Myrobalan plum tree grafted with Methley Japanese plum.



**Figure 3.** A) Sampling of the roots was made with a California Sampler; B) 330 cm<sup>3</sup> root sample extracted from a specific depth, at 40 cm away from the trunk following the cardinal points.

in the roots were separated using a #2 sieve, following a modified version of the method described by Cossio *et al.* (2008). Subsequently, the roots were washed with running water in the lab. Two groups of roots were created: a group of white growing roots and light brown absorbing roots ( $\leq 5$  mm thick) (GA) and a group of intermediate and conducting roots ( $\geq 5$  mm thick) (IC) (Becerril *et al.*, 2009; Kolesnikov, 1971).

### Response of the variables

**Thermal regime of the soil.** Considering 4.5 °C as the root base temperature (Kolesnikov, 1971), the MinT and MaxT of the soil were replaced in the Residual Method proposed by Snyder (1985).

**Root density.** The GA and IC root groups were measured with an EY-2200a digital scale (Asleep<sup>®</sup>, USA); afterwards, they were dried in a 620 forced air stove (Napco<sup>®</sup>, USA) for 72 h, at 70 °C, in order to establish dry weight (g). Root density was equal to the g of root dry matter found in 330 cm<sup>-3</sup>.

**Root accumulation.** Root accumulation was determined transforming the root densities into relative percentages (Cossio *et al.*, 2008).

**Phenology of the cultivar.** The phenology stages of the cultivar Methley used in this study were based on González-Pérez *et al.* (2018).

### Statistical analysis

In order to determine potential significant differences, an ANOVA and a Student's t-test were used to analyze the depth of the extraction of the roots for the treatment. The data about the phenological stages treatment were analyzed with an ANOVA and a Tukey Multiple Comparison Test (P=0.05). The SAS 9.4 statistical package was used.

## RESULTS AND DISCUSSION

The soil moisture content was influenced by the soil texture and the organic matter content of the soil, as well as the irrigation. It fluctuated between FC and MPMC (Minimum Permissible Moisture Content). This condition was appropriate for the growth of roots of fruit trees (Ley, 1994). On the other hand, the color of the soil affected its temperature: darker (wet) soils absorbed a higher solar radiation, and even more warmer as the dates in which sun rays hits the place in a perpendicular angle got closer (Forsythe, 2002). From May to September, soil temperature was closer to the optimum T shape of the root of the plum tree (23-25 °C) (Ley, 1994) and it was permanently higher than the Tb of the root (4.5 °C) (Kolesnikov, 1971). Consequently, the thermal regime of the soil during the study recorded 4,631.8 heat units. This physiological period allowed the formation of root mass and the monitoring of the development of the root during the phenological cycle. This thermal period can replaces the use of the calendar (Snyder, 1985), which does not accurately predict root development (Mendoza *et al.*, 2004; Slafer and Savin, 1991).

**Root density.** The density of the GA roots during the phenological cycle was higher at a depth of 0-25 cm than at a depth of 26-50 cm (Table 1), as a consequence of the tendency of the plum roots to grow right under soil surface (Agustí, 2004). In addition, mulching and irrigation promote the growth of the root in the upper layer of the soil (Wei

*et al.*, 2002), through the increase of temperature and the reduction of water percolation (Zhang *et al.*, 2017). In fact, roots can adapt to the environment through the formation and elongation of lateral roots, which help them to survive changing nutritional conditions (Malamy, 2005). This difference in root density at both depths can be the consequence of the high variability of the vigor of the Myrobalan plum tree (Popescu and Caudullo, 2016) and the distribution and availability of nutrients in the soil (Hodge, 2006).

Meanwhile, the IC roots recorded the highest density at a depth of 0-25 cm, at the beginning of the ecodormancy (21/09/15) and leaf abscission (27/11/15) (Table 1). This biological response was consequence of the caulinar origin of the roots of the rootstocks (given their clonal propagation) and their tendency to develop horizontal roots (Barlow, 1986). In addition, the organic soil cover caused roots to grow closer to the surface (Gutiérrez *et al.*, 2006). During the rest of the sampling dates, similar root densities were recorded in IC roots, at both depths. Concerning these observations, a significant proportion of the total root system is composed of suberized roots (Azcón-Bieto and Talón 2000), which allow to maintain the fruit set (Becerril *et al.*, 2009; Gutiérrez *et al.*, 2006); this explains why the roots of fruit trees are found at a depth of 25-50 cm.

Although the Myrobalan plum tree can successfully grow in sandy soils (Popescu and Caudullo, 2016), no GA or IC roots were found at a depth of >50 cm. These data confirm that an appropriate development of the plum trees requires a depth of 40-50 cm (Silva and Alonso, 1976). Nevertheless, Gutiérrez *et al.* (2006) have reported a different scenario, in which the highest root densities of fruit trees are found at a depth of 10-90 cm.

Regarding root density affected by the phenological stage of the cultivar, GA recorded the greatest root densities during bud swelling (18/02/2016), at both depths (Table 2). These results are similar to root formation before budding (Ryugo, 1988; Shaw, 1998). Consequently, the maximum root growth rates recorded during flowering (Ryugo, 1988; Shaw, 1998) seem to be the result of the roots that appeared during bud swelling and budding. Although the Myrobalan plum tree can successfully grow in sandy soils

**Table 1.** Root density based on soil depth.

Date	Root density (g 330 cm <sup>-3</sup> )			
	GA 0-25	GA 25-50	IC 0-25	IC 25-50
21/09/15	0.199 a	0.031 b	1.735 a	0.415 b
26/10/15	0.143 a	0.264 a	1.642 a	0.939 a
27/11/15	1.286 a	0.173 b	2.706 a	2.845 a
28/12/15	0.747 a	0.09 b	2.421 a	2.938 a
18/02/16	1.479 a	0.658 a	1.592 a	1.655 a
21/03/16	0.56 a	0.046 a	1.399 a	1.569 a
6/06/16	0.877 a	0.169 b	0.517 a	0.714 a
11/07/16	1.422 a	0.48 b	1.496 a	0.59 b
4/08/16	1.161 a	0.454 b	0.357 a	0.988 a

GA 0-25 = growing and absorbing roots at a depth of 0-25 cm. GA 25-50 = growing and absorbing roots at a depth of 25-50 cm. IC 0-25 = intermediate and conducting roots at a depth of 0-25 cm. IC 25-50 = intermediate and conducting roots at a depth of 25-50 cm. According to the Student's t-test, the values with the same letter are equal for each group of roots (GA, IC) at both depths.



**Table 2.** Root density based on the phenological stage of the cultivar.

Phenological stage	Root density (g 330cm <sup>-3</sup> )			
	GA 0-25	GA 25-50	IC 0-25	IC 25-50
Beginning ecoletargy	0.199 ab	0.031 c	1.735 a	0.415 a
Beginning endoletargy	0.143 b	0.264 abc	1.642 a	0.939 a
Beginning endoletargy	1.286 ab	0.173 bc	2.706 a	2.845 a
Middle of endoletargy	0.747 ab	0.09 bc	2.421 a	2.938 a
Bud swelling	1.479 a	0.658 a	1.592 a	1.655 a
Full bloom	0.56 ab	0.046 bc	1.399 a	1.569 a
3rd stage of fruit development	0.877 ab	0.169 bc	0.517 a	0.714 a
Foliar abscission	1.422 ab	0.48 ab	0.59 a	1.496 a
Foliar abscission	1.161 ab	0.454 abc	0.357 a	0.988 a

GA 0-25 = growing and absorbing roots at a depth of 0-25 cm. GA 25-50 = growing and absorbing roots at a depth of 25-50 cm. IC 0-25 = intermediate and conducting roots at a depth of 0-25 cm. IC 25-50 = intermediate and conducting roots at a depth of 25-50 cm. Based on the Tukey's Test (P=0.05), values with the same letter are equal for both GA and IC groups, in each phenological stage.

(Popescu and Caudullo, 2016), the lowest root density rates were recorded during the ecodormancy (21/09/2015) and endodormancy (26/10/2015), at a depth of 0-25 and 25-50, respectively.

Meanwhile, no significant differences in IC root density were recorded at both depths (Table 2), during the phenological stages. Most of the root system of woody species is composed of suberized roots (Azcón-Bieto and Talón 2000), which are a permanent conducting structure of the plant (Esau, 1976; Robbins *et al.*, 1976). In addition, they store reserves and absorb water (Becerril *et al.*, 2009), which could have standardized their densities.

**Root growth accumulation.** From the beginning of both the ecodormancy (21/09/2015) and during the endodormancy (27/11/2015), a constant root accumulation took place in all the different types of roots (Figures 4-7). During full endodormancy (28/12/2015), the accumulation of GA roots decreased, while the IC roots recorded a considerable accumulation. During bud swelling (18/02/2016), the GA roots increased their root accumulation, while the IC roots decreased theirs. These tendencies were the result of the highly variable vigor of the Myrobalan plum tree (Popescu and Caudullo, 2016) and the capacity of the simultaneous primary and secondary growth of the different parts of the tree (Eshel and Beeckman, 2013).

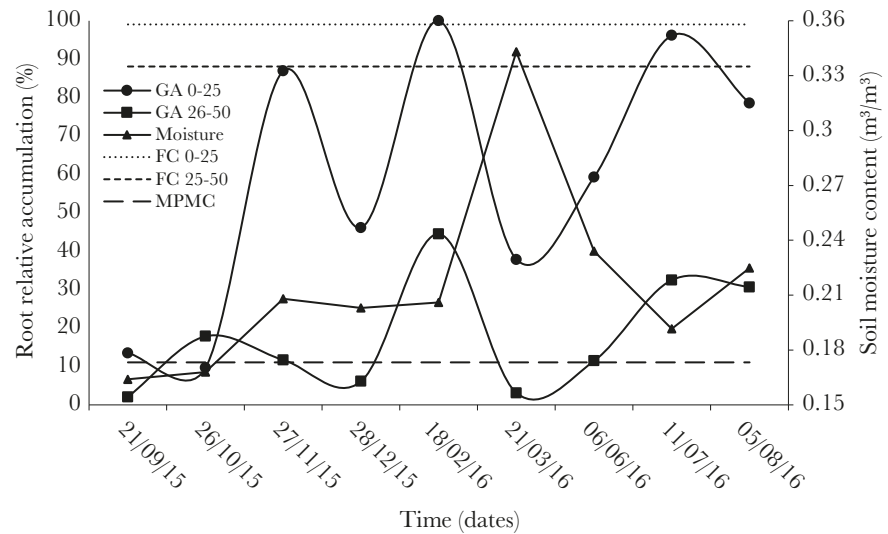
Consequently, the overall maximum root accumulation was recorded from the beginning of the ecodormancy to the bud swelling, when the aerial growth and development decreased. During that period, soil moisture content fluctuated between MPMC and CF, while the temperature was closer to the Tb of the root (4.5 °C, Kolesnikov, 1971) and very distant from the optimal root temperature (23-25 °C, Ley, 1994). These results show that the dormancy of the cultivar is more influential on the root accumulation than soil temperature. These findings are different from those obtained by Ryugo (1988) and Shaw (1998), who pointed out that the root grows more during flowering, regardless of



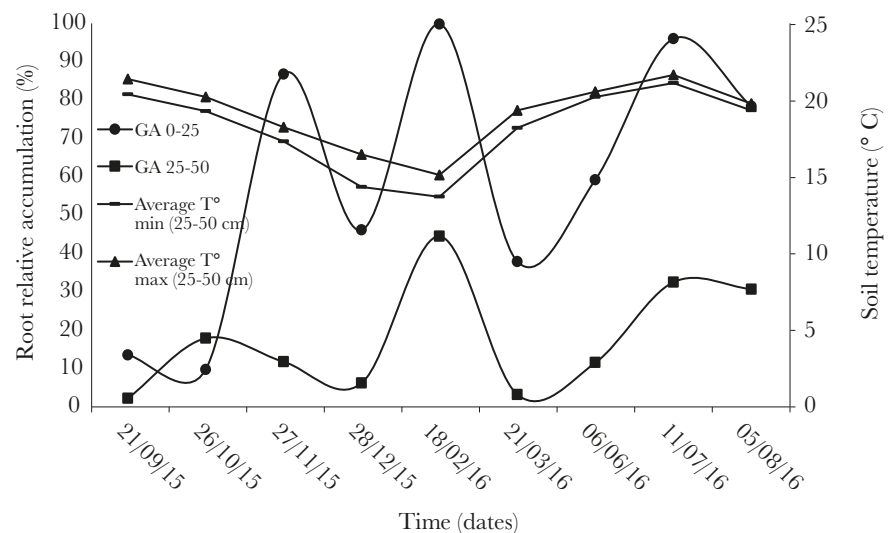
the type of fruit tree. In addition, both authors reported significant root growth fluxes before leaf abscission.

During flowering (21/02/2016), GA recorded a high and a medium root accumulation, at a depth of 0-25 cm and 26-50 cm, respectively (Figures 4 and 5). Meanwhile, the IC roots recorded a medium root accumulation at both depths. This difference in root accumulation is the result of the biomass distribution relationship between organs (Ovando *et al.*, 1993).

During fruit development (06/06/2016) until the harvest ripeness, a medium increase of root accumulation was recorded in GA roots, determined by the presence of fruits (Palmer *et al.*, 1991). These results differ from the findings of León (1994), who reported that the root development of the Japanese plum tree is highly limited by plant activity

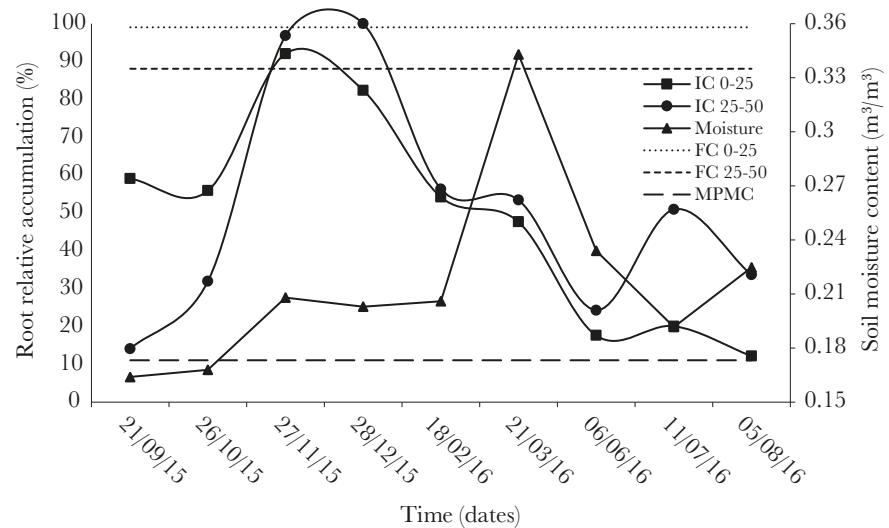


**Figure 4.** Accumulation of growing and absorbing roots and soil moisture content, from September 2015 to August 2016.

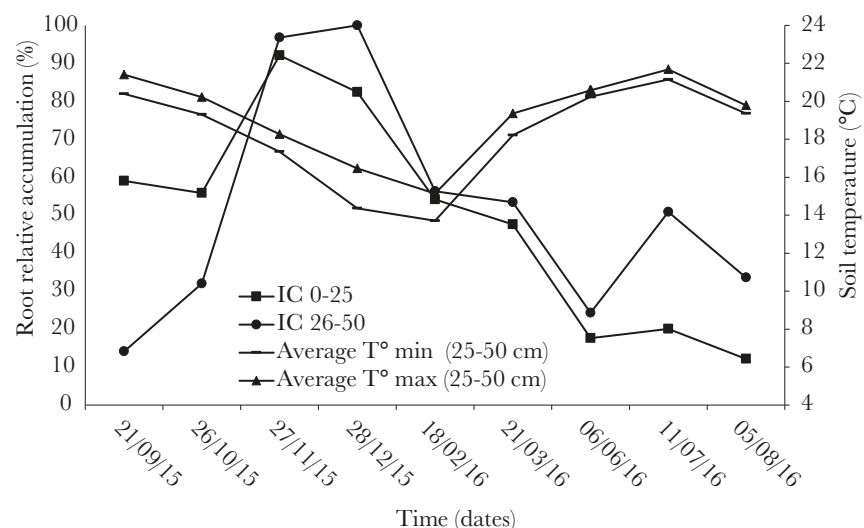


**Figure 5.** Accumulation of intermediate and conducting roots and soil temperature, from September 2015 to August 2016.

and reproductive processes, as well as root accumulation only occurs in the absence of canopy growth or reproductive stages, or when both have decreased. In this regard, the proportion of photo assimilates aimed to the roots diminishes as the tree bears a greater fruit load (Palmer *et al.*, 1991). This phenomenon was also recorded in this study, despite the appropriate water condition of the soil, which favored root accumulation (Shock *et al.*, 1998). The presence of intermediate roots during the whole phenological cycle (Figures 6 and 7), shows that the growing roots formed in previous phenological phases experienced a secondary growth (Esau, 1976; Robbins *et al.*, 1976). In addition, during the endodormancy, the trees generated the greatest portion of conducting roots, which is normal considering the high vigor of Myrobalan plum (Popescu and Caudullo,



**Figure 6.** Accumulation of intermediate and conducting roots and soil moisture content, from September 2015 to August 2016.



**Figure 7.** Accumulation of intermediate and conducting roots and soil temperature, from September 2015 to August 2016.

2016). These roots are the anchorage structure of the root system (Esau, 1976; Robbins *et al.*, 1976).

Finally, the uninterrupted generation of root material during the study period—even during the simultaneous phenological stages that took place in the canopy—is important, because roots synthesize cytokines, which carry information about the nutrient status of the root system (particularly nitrogen) to the aerial part of the tree (Sakakibara *et al.*, 1998; Yong *et al.*, 2000).

## CONCLUSIONS

The results reported in this study are useful to plan agronomic activities such as fertilization and irrigation. This is due to the punctual identification of development stages where roots carry out primary and secondary growth (with their respective proportions) during the phenological cycle in certain soil depth. In conjunction with development stages of the canopy, for example, flowering or fruit development, allow to have a complete notion about the most appropriated moments to supply nutrients and water.

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