RESEARCH NOTE

Growth, yield and iron deficiency tolerance level of six peach rootstocks grown on calcareous soil

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Abstract

C. Sotomayor, R. Ruiz, and J. Castro. 2014. Growth, yield and iron deficiency tolerance level of six peach rootstocks grown on calcareous soil. Cien. Inv. Agr. 41(3):403-409. Iron deficiency is a major problem that affects many fruit crops grown on calcareous soils. A four-year study was conducted to evaluate the effect of calcareous soils on the vegetative growth and productivity of six peach rootstocks (Nemaguard, Atlas, MRS 2/5, Cadaman, GF 677 and GxN15) grafted with 'Ruby Diamond' nectarines. Trees were planted in 100 L containers filled with a sandy loam textured, calcareous soil (6.3% active calcium carbonate) with a pH of 8.0 and an intermediate level of available iron (5 mg kg^{-1} of Fe^{+2}). The trunk diameter during the 4th growing season was larger in GF 677, GN 15, Cadaman and Atlas, whereas Nemaguard and MRS 2/5 showed smaller average values. Cadaman and Atlas presented the highest pruning weight (a measure of vegetative growth), GxN 15 and GF 677 were intermediate, and Nemaguard and MRS 2/5 exhibited the lowest weight. In the fourth year, yield per tree was higher in Atlas and Cadaman, while Nemaguard had the lowest yield. MRS 2/5 and GxN 15 had the highest individual fruit weights, particularly when compared with Cadaman. Regarding leaf chlorosis, the most severe symptoms were exhibited by Nemaguard (12.3 mg kg^{-1} of active iron), with mild symptoms in Atlas and slight to no symptoms in Cadaman, MRS 2/5 and GxN 15. Leaf chlorosis was not observed in GF 677 (26.4 mg kg^{-1} of active iron). All rootstocks, especially GF 677, showed better performance than Nemaguard with respect to chlorophyll measurements of the leaves obtained through the SPAD technique.

Key words: Calcareous soil, Fe^{+2}, iron chlorosis, peach rootstock, SPAD.

Introduction

Iron (Fe) deficiency is a major problem for fruit crops grown on calcareous or lime (high pH) soils (Tsipouridis *et al.,* 2005). Leaf chlorosis is the most characteristic symptom, in which the leaves are pale or yellow and the veins remain green. If the deficiency is mild to moderate, it only affects young leaves; however, if it is severe, it will affect the entire plant (Tagliavini and Rombolà, 2001). Another characteristic symptom is a reduction in growth and productivity; in grapevines, growth may be affected before leaf chlorosis is observed (Romheld, 2000). It is believed that calcareous

soils lack available iron, apart from any problems with absorption and translocation within the plants (Mengel, 1995). Koseoglua (1995) noted that HCl-extractable iron content is negatively correlated with the calcium carbonate and calcium bicarbonate contents in soils.

Peaches are an important fruit crop in many countries, and mineral nutrition is one of the factors that most significantly affects tree productivity and fruit quality. Many orchards are located in areas with calcareous soils, increasing the possibility that they may develop iron chlorosis (Ruiz *et al.*, 1984; Toselli *et al.*, 2000). Despite the high Fe concentrations in the roots and leaves of vines grown in calcareous soils, the plants can suffer from Fe deficiency (Gruber and Kosegarten, 2002). According to Koseoglua (1995), the availability of Fe and its absorption by peach roots is significantly affected by high pH and the presence of carbonates and bicarbonates in the soil, and the resulting iron deficiency often manifests as leaf chlorosis. However, there is no relationship between the total Fe concentration in leaf tissue and the development of iron chlorosis (Ruiz *et al.,* 1984; Alvarez-Fernández *et al.,* 2003; Razeto and Valdés, 2006). Peach trees are Strategy I plants, and Fe uptake by these plants is preceded by a reduction step from Fe^{+3} to Fe^{+2} (Tagliavini and Rombola, 2001). The soluble Fe extracted with water from dried soil samples has been shown to be ineffective as a diagnostic tool to determine the amount of Fe available to leaves (Sadzawka *et al.*, 2008; Razeto and Valdés, 2006). It has been suggested that the analysis of active iron (Fe^{+2}) extracted with acid from fresh samples would be a better indicator of the iron that is available to plants (Zohlen, 2000). Using this analysis, Sadzawka *et al.* (2008) found that there was a greater correlation between the degree of leaf symptoms and the level of active iron (Fe^{+2}) found in the sample.

Determinations of active iron have also been found to be useful in studies of iron deficiency in avocado leaves (Neaman and Aguirre, 2007; Ruiz *et al.*, 2008) and in peach leaves (Koseoglu

and Acikgoz, 1995). Other tissues of fruit trees, such as flowers, have been used as diagnostic tools for iron (Abadia *et al.*, 2000).

Iron deficiency decreases vigor, yield and fruit quality in many species, and it affects bud development in the subsequent season (Tagliavini and Rombolá, 2001). Additionally, iron deficiency decreases carbohydrate reserves in the roots (Ruiz *et al.*, 2008). The development of iron chlorosis symptoms in peach orchards is known to affect tree growth and to reduce fruit yield and quality (Alvarez-Fernándes *et al.*, 2003). Of all the tree species, peaches tend to be more susceptible to developing iron deficiency chlorosis, which can be expensive and difficult to correct (Mengel and Kirkby, 2001; Tagliavini and Rombolá, 2001). The use of Fe deficiency-tolerant genotypes as rootstocks is a reliable solution to prevent iron chlorosis (Socías *et al.*, 1995). Indeed, one of the successful strategies for solving problems with calcareous soils and iron chlorosis is the use of tolerant or resistant rootstocks, which appear to improve iron assimilation by the plant. Giorgi *et al.* (2005) found that the type of peach rootstock grown in calcareous soils can affect vegetative qualities, productivity, fruit quality and nutritional attributes. The well-known Nemaguard rootstock, which is used worldwide, has been reported to be an iron chlorosis-sensitive rootstock (Ruiz *et al.*, 1984; Tagliavini and Rombolá, 2001).

There have been reports of new rootstocks that resist the development of chlorosis in calcareous soils. According to Romera *et al.* (1991), the rootstock Nemaguard is more susceptible to iron chlorosis induced by calcium bicarbonate than the hybrid GF 677. Additionally, Tsipouridis *et al.* (2005) found that the GF 677 rootstock was very efficient in dry, calcareous soils, and its grafted scion cultivars achieved higher concentrations of foliar iron. Loreti and Massai (2006) also reported that the rootstocks MRS 2/5 and Cadaman were fairly tolerant of iron deficiency chlorosis in calcareous soils. According to Iglesias *et al.* (2004), the Nemaguard and Nemared rootstocks

showed the greatest sensitivity to iron chlorosis in calcareous soils, and while GxN 15 and MRS 2/5 were less affected, GF 677 was the only rootstock that did not develop chlorosis.

The objective of this study was to evaluate the effect of calcareous soils on vegetative growth, productivity and intensity of leaf chlorosis symptoms in six peach rootstocks, including the traditional Nemaguard rootstock and the new rootstocks Atlas, MRS 2/5, Cadaman, GF 677 and GxN 15, grafted with 'Ruby Diamond' nectarines. The study was conducted over a period of four years.

Materials and methods

'Ruby Diamond' nectarine scions were grafted (by the Univiveros and Requinoa Nurseries) onto Nemaguard, Atlas, MRS 2/5, Cadaman, GF 677 and GxN 15 peach rootstocks and were grown for four years in 100 L containers at the Estación Experimental de Pirque UC located in Pirque (near Santiago), Chile (33º43' S and 71º11' W). This location has a warm temperate climate. The 100 L containers were filled with a sandy loam calcareous soil (previously sterilized with methyl bromide) containing 6.3% active $CaCO_3$ (pH 8.0) with an intermediate level of available iron (5 mg kg^{-1} DTPA-extracted). All containers were drip-irrigated to achieve tensiometer readings of 20 cb, and the soil fertility level (originally N: 18 mg kg^{-1}, P: 9 mg kg^{-1}, K: 324 mg kg^{-1}, Zn: 1 mg kg^{-1} and Mn: 5 mg kg^{-1}) was maintained with annual applications of 100 g potassium nitrate (through the irrigation water). The experiment followed a completely randomized design with 12 replications. The experimental unit was each grafted rootstock in an individual container (spaced 5 m apart), and the treatments were the rootstocks Nemaguard, Atlas, MRS 2/5, Cadaman, GF 677 and GxN 15, all of which were grafted with Ruby Diamond nectarine.

Pruning was performed by removing half of all the shoots produced in a year during winter rest. Measurements of the fruit (yield and fruit number), trunk diameter and weight of pruning material were performed at the end of the experiment (4th season). The iron concentration was evaluated in 30 randomly collected leaves per plant in January using a chemical analysis method based on the active iron fraction (Fe^{+2}) (Katyal and Sharma, 1980, 1984). The procedure was carried out using 30 fresh leaves per plant at 24 hours after collection under acidic conditions with ortho-phenanthroline. All these determinations were complemented by leaf chlorophyll measurements using a Minolta SPAD-502 chlorophyll meter; 30 leaves per plant were measured at random. According to these measurements, a leaf chlorosis scale was established, with 1.0 for no chlorosis (more than 40 Spad units), 2.0 for mild chlorosis (30 to 40 Spad units), and 3.0 for severe chlorosis (less than 30 Spad units). The soil in the containers was regularly analyzed to determine the pH, electrical conductivity and basic fertility.

All collected data were analyzed with ANOVA, and the means were separated by the Tukey-Kramer test at P = 0.05.

Results and discussion

The trunk diameter of the different rootstocks increased significantly during this study, with GF 677, GxN 15, Cadaman and Atlas showing larger values when compared with Nemaguard and MRS 2/5. Cadaman and Atlas had the highest pruning weight (a measure of vegetative growth), while GF 677, GxN 15 and MRS 2/5 had intermediate weights. Nemaguard showed the lowest weight and vigor (Table 1).

When measured at the end of the experiment, the trunk diameters of GF 677, GxN 15, Cadaman and Atlas were significantly larger than those of Nemaguard and MRS 2/5. Cadaman and Atlas had significantly higher pruning weights, GF 677 and GxN 15 had intermediate weights, and MRS 2/5 together with Nemaguard had the lowest weights (Table 1).

Table 1. Vegetative growth of Ruby Diamond nectarine trees grafted onto different peach rootstocks during the 4th growing season.

Rootstock	Trunk diameter (mm)	Pruning weight (g tree[-1])
Cadaman	43.8 ab	705 a
Atlas	40.2 ab	691 a
GxN 15	46.6 ab	450 b
GF 677	52.8 a	442 b
MRS 2/5	20.8 c	254 c
Nemaguard	30.2 bc	250.0 c

Different letters in a column indicate significant differences according to the Tukey-Kramer test at P= 0.05.

The Cadaman and Atlas peach rootstocks produced the greatest overall vegetative growth in the calcareous soil utilized in this study. In contrast, Nemaguard and MRS 2/5 showed the least vegetative growth, which could also be due to the mild dwarf phenotype of MRS 2/5 (Loreti and Massai, 2006). Because Nemaguard is not a dwarfing rootstock, the low growth could be due to a greater sensitivity to iron deficiency.

By the fourth growing season, the yield per tree was highest in Atlas and Cadaman, while GF 677, GxN 15 and MRS 2/5 showed intermediate yields. Nemaguard exhibited the lowest yield (Table 2). The individual fruit weight of MRS 2/5 was significantly higher than that of all other rootstocks except GxN 15, while Cadaman followed by Atlas had the lowest fruit weights (Table 2).

Table 2. Yield and fruit weight in Ruby Diamond nectarine trees grafted onto different peach rootstocks during the 4th growing season.

Rootstock	Individual fruit weight (g)	Yield per tree (kg tree[-1])
Atlas	128 b	7.4 a
Cadaman	110 c	7.2 a
GxN 15	148 ab	4.8 b
GF 677	120 b	4.4 b
MRS 2/5	192 a	4.2 b
Nemaguard	132 b	3.6 c

Different letters in a column indicate significant differences according to the Tukey-Kramer test at P= 0.05.

The highest total iron concentrations in leaves were observed in GF 677, GxN 15 and Cadaman when compared with the other rootstocks (Table 3). These leaf iron values are high according to the standards indicated by Reuther and Robinson (1997). These results confirm the insensitivity of the total Fe content for the diagnosis of leaf deficiency, as has been reported by many other authors. Conversely, as shown in Table 3, leaf ferrous iron (Fe^{+2}) content in most rootstocks was higher than that in Nemaguard. In Atlas, this difference was not statistically significant. The inaccuracy of using total Fe content to evaluate the iron status in peaches must again be emphasized here. According to the visual evaluation scale used for determining the degree of iron chlorosis, the symptoms were severe in Nemaguard; mild in Atlas; mild to non-existent in GxN 15, Cadaman and MRS 2/5; and non-existent in GF 677. These observations agree with those of Cinelli *et al*. (2004) in the sense that the GF 677 rootstock has a high tolerance to iron chlorosis. Similar results have been reported by Iglesias *et al*. (2004). With regard SPAD measurements, Nemaguard exhibited the lowest value (21.6 units) when compared with the other rootstocks, whereas GF 677 showed the highest value (42.6 units). These data may indicate that, for reasons not yet well understood, these rootstocks are able to extract Fe^{+2} from soil more efficiently and enhance chlorophyll synthesis in leaves when compared with Nemaguard. Normally, under aerobic conditions, plants actively absorb Fe^{+2} after the reduction of soil Fe^{+3} by a reductase in the root plasma membrane (Romheld, 1987). It is possible that the enhanced iron nutrition obtained when using GF-677, GxN 15 and Cadaman rootstocks is due to the plants' ability to efficiently absorb Fe. According to Mengel and Kirby (2001), these plants are capable of excreting organic substances with iron-chelating abilities, such as citric acid, caffeic acid and others, into the rooting medium, thus improving iron absorption by the roots and preventing further oxidation or inactivation.

Table 3. Leaf iron content (total Fe and Fe^{+2}), SPAD values and degree of leaf chlorosis during the 4th growing season.

Rootstocks	Total Fe (mg kg^{-1})	Fe^{+2} (mg kg^{-1})	Leaf SPAD values	Degree of chlorosis
GF 677	357 a	26.4 a	42.6 a	1.0 (none)
GxN 15	356 a	25.1 a	42.1 a	1.5 (mild to none)
Cadaman	359 a	23.0 a	41.9 ab	1.5 (mild to none)
MRS 2/5	319 b	23.5 a	42.0	1.5 (mild to none)
Atlas	326 b	19.2 ab	36.8 b	2.0 (mild)
Nemaguard	330 ab	12.3 b	21.6 c	3.0 (severe)

Different letters in a column indicate significant differences according to the Tukey-Kramer test at P= 0.05.

It can be concluded that under the experimental conditions described above, the rootstocks GF 677, GxN 15, Atlas, MRS 2/5 and Cadaman were more better able to resist iron deficiency and iron chlorosis compared with Nemaguard. Cadaman and Atlas showed higher vegetative growth, higher yields, and an increased trunk diameter when compared with Nemaguard and MRS2/5. These experiments confirm the inefficiency of total iron analysis in leaves as a tool for diagnosing iron deficiency. Leaf ferrous iron (Fe^{+2}) was found to be a very sensitive tool for the determination of iron status, with higher Fe^{+2} levels in GF 677, GxN, Cadaman and MRS 2/5 and lower Fe^{+2} levels in Nemaguard. These results are closely linked to leaf foliar symptoms and SPAD values and are generally in agreement with the reports by Romera *et al.* (1991) and Tsipouridis *et al.* (2005) in peach trees.

Resumen

C. Sotomayor, R. Ruiz y J. Castro. 2014. Crecimiento vegetativo, productividad y tolerancia a la deficiencia de hierro en seis portainjertos de duraznero creciendo en suelo calcáreo. Cien Inv. Agr. 41(3):403-409. La deficiencia de hierro es un problema mayor que afecta a muchos cultivos frutales en suelos calcáreos. Se realizó un estudio durante cuatro años para evaluar el efecto de suelos calcáreos en el comportamiento vegetativo y productivo de seis portainjertos de duraznero (Nemaguard, Atlas, MRS 2/5, Cadaman, GF 677 y G×N 15) injertados con el nectarino Ruby Diamond. Los árboles fueron plantados en contenedores de 100 L conteniendo un suelo franco-arenoso y calcáreo (6,3% de carbonato de calcio activo), con pH 8,0 y un nivel medio de hierro aprovechable (5 mg kg^{-1} de Fe^{+2}). El diámetro de tronco a la cuarta temporada y final de este estudio fue mayor en GF 677, G×N 15, Cadaman y Atlas mientras que Nemaguard y MRS 2/5 mostraron diámetros promedio menores. Con respecto al peso de la poda (una medida del crecimiento vegetativo) Cadaman y Atlas presentaron los pesos mayores, G×N 15 y GF 677 fueron intermedios, mientras que MRS 2/5 y Nemaguard lograron los menores. Al cuarto año la producción por planta fue mayor en Atlas y Cadaman, mientras que en Nemaguard fue la más baja. En cuanto a peso individual de fruto, MRS 2/5 y G×N 15 alcanzaron la cifra mayor, particularmente comparado con Cadaman. Respecto a la deficiencia de hierro (clorosis férrica), los síntomas más severos los mostró Nemaguard (12 mg kg^{-1} de Fe^{+2}). No se desarrolló clorosis en GF 677, hubo ligera clorosis en Atlas y leve a ninguna en los otros portainjertos. Todos los portainjertos, especialmente GF 677, mostraron una mejor respuesta que Nemaguard en las mediciones de clorofila a través de la técnica SPAD.

Palabras clave: Clorosis férrica, Fe^{+2}, portainjerto de duraznero, SPAD, suelo calcáreo.

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