

RESEARCH ARTICLE

Cultivar-rootstock interactions on growth, yield and mineral nutrition of newly planted peach trees in a pot experiment

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ABSTRACT

Two peach cultivars, i.e. 'Andross' and 'Mercil' were budded onto four rootstocks of varying vigor (the vigorous rootstocks 'Garnem' and 'GF 677', the semi vigorous to vigorous rootstock 'Rootpac R' (RPR) and the dwarf to semi dwarf rootstock 'Rootpac 20' (RP20)) and planted in pots. The aim of the present trial was to study the effect of rootstock and cultivar on growth characteristics, yield and on plant nutrient status in order to plan the most efficient fertilization program fitted to each rootstock specific properties. The plants were grown for three years, and each year the growth of the trees (in terms of trunk cross sectional area, tree height, and shoot length), fruit production and leaf nutrient concentration were assessed. The lowest tree height in both cultivars was recorded when these were grafted on RP20 (the most dwarfing rootstock of all four used). The yield per tree was highest when the most vigorous rootstocks were used. 'Garnem' resulted in the highest upper plant dry weight, while 'RP20' in the lowest. Cultivar exhibited a significant effect regarding leaf nutrient concentration, as 'Andross' presented higher concentrations of N, K, Ca and Fe in most combinations and lower concentrations of P and Cu. The discriminant analysis, using all growth and nutrient data from the last two years, revealed that irrespective of the cultivar budded, 'RP20' and 'GF 677' were clearly distinguished from each other and from 'RPR' and 'Garnem'. On the other hand, the hierarchical agglomerative analysis pointed out the crucial role of 'RP20' and 'Garnem' on tree growth and leaf nutrient concentration, with the cultivar budded on them playing a minor role. In conclusion, the fertilization program of a young, newly established orchard, should take into account the singularity of each scion-rootstock combination, to achieve the optimum tree performance.

Keywords: 'Garnem'; 'GF 677'; nutrients; 'Rootpac R'; 'Rootpac 20'

INTRODUCTION

Peaches and nectarines are the most worldwide known stone fruits and are highly appreciated by consumers (Drogoudi and Tsiouridis, 2007; Reig et al., 2016; Seker et al., 2017; Santana et al., 2020). The expansion of peach cultivation and nursery production is mainly based on the introduction to the market of a high number of cultivars each year, satisfying every consumer's needs and expectations. Like many of the fruit tree crops, peach is characterized by the so-called replant disease, which hopefully is solved in this case, by the use of the proper clonal rootstock (Jiménez et al., 2011).

Therefore, nowadays, commercial peach trees consist of two different parts, i.e. the rootstock and the scion cultivar (Zarrouk et al., 2005; Zarrouk et al., 2006; Ben Yahmed

et al., 2020; Shahkoomahally et al., 2021). The right choice of the rootstock and scion cultivar combination is probably the most important factor of the peach cultivation success (Reig et al., 2016; Özdemir et al., 2019; Santana et al., 2020; Shahkoomahally et al., 2020). Rootstocks are selected based on some good properties, such as the aforementioned peach replant tolerance, resistance to soil pathogens and lime-induced chlorosis, to root-knot nematodes, to root asphyxia etc. (Caruso et al., 1996; Bouhadida et al., 2009; Font i Forcada et al., 2012; Ben Yahmed et al., 2016; Jimenes et al., 2018; Iglesias et al., 2019; Seker et al., 2017; Santana et al., 2020). The rootstock affects also the tree vigor, tree precocity, yield efficiency, fruit size and quality, as well (Boyhan et al., 1995; Reig et al., 2018; Reig et al., 2020; Seker et al., 2017; Shahkoomahally et al., 2020). The effects of rootstock on the grafted cultivar characteristics are mainly attributed to the

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change in the hormone balance induced by the rootstock, the uptake and the translocation of nutrients and water (Zarrouk et al., 2005; Mestre et al., 2015; Ben Yahmed et al., 2020; Reig et al., 2020; Shahkoomahally et al., 2020).

Genetic improvement practices worldwide have yielded a plethora of stone fruit rootstocks, which can be used according to the needs and requirements of the crop. Therefore, the necessity of screening these genotypes has become obvious (Font i Forcada et al., 2012; Ben Yahmed et al., 2020).

During the last years, dwarf rootstocks have been introduced to the peach and almond industry, aiming at increasing plant density, as has been the case in other species such as apple, cherry, etc (Weibel et al., 2003; Basile et al., 2003; Tombesi et al., 2009). Dwarf rootstocks have been used for many years in the apple and cherry industry, acknowledging their positive effects on early fruit production and quality as well as the early occupation of the allotted space (Weber, 2001; Lordan et al., 2017; Hrotkó and Rozpara, 2017). The superior fruit quality achieved with dwarf rootstocks has been partly attributed to the better nutrient distribution and balance among different organs within the tree (Mestre et al., 2015).

The most common rootstocks used in Mediterranean countries are the ‘almond x peach’ hybrids, mainly due to their tolerance to Fe chlorosis (Zarrouk et al., 2005; Aras et al., 2021). The most widespread peach rootstock in Greece and generally southern Europe is ‘GF 677’ (Assimakopoulou et al., 2011), a ‘peach x almond’ rootstock, characterized by high vigor, resistance to lime-induced chlorosis, and replant disease (Jiménez et al., 2011). Another popular ‘peach x almond’ rootstock is ‘Garnem’, with similar characteristics to ‘GF 677’, regarding tree vigor and resistance to lime-induced chlorosis, as well as drought stress and root-knot nematode resistance (Reig et al., 2020). During the last years, a ‘myrobalan x almond’ (*Prunus cerasifera* x *Prunus dulcis*) hybrid was introduced, called ‘Rootpac R’ (Rootpac® R or RPR) by ‘Agromillora Iberia’ nursery, characterized as a medium-vigor rootstock, with root-knot nematode, root asphyxia and replant disease resistance (Pinochet, 2010; Özdemir et al., 2019). The most dwarf though plum based hybrid rootstock (*Prunus besseyi* x *P. cerasifera*) Rootpac® 20 (Densipac or RP20) (also by ‘Agromillora Iberia’), is a relatively new rootstock, characterized by moderate resistance to iron chlorosis, salinity and root-knot nematodes and by sensitivity to water deficit (Opazo et al., 2020). ‘RP20’ is suitable for high-density systems with closely spaced trees (1200- 3000 trees ha⁻¹), achieving fast farm space coverage, as has been clearly shown in the case of almond.

The present trial aimed to assess the effects of different cultivar-rootstock combinations (using two peach scion cultivars, i.e. ‘Andross’ and ‘Mercil’ budded on four *Prunus* rootstocks, differing in vigor, i.e. ‘GF 677’, ‘Garnem’, ‘RPR’ and ‘RP20’) on tree growth, early years’ productivity, and plant nutrition.

MATERIALS AND METHODS

Plant material and growing conditions

The present experiment was carried out during three successive growing seasons (2016-2019) at the orchard of the Agricultural University of Athens (Latitude: 37°58'N, Longitude: 23°32'E, Altitude: 30 m from the sea level). Two mid-season peach cultivars, i.e. ‘Mercil’ and ‘Andross’ were used as plant material, budded (dormant bud) on four rootstocks, i.e. the vigorous peach-almond hybrids ‘GF 677’ (*Prunus dulcis* x *Prunus persica*) and ‘Garnem’ (*Prunus amygdalus* x *Prunus persica*), the medium vigor plum-almond hybrid ‘RPR’ (*Prunus cerasifera* x *Prunus dulcis*) and the dwarfing rootstock ‘RP20’ (*Prunus besseyi* x *Prunus cerasifera*).

The plants were about 30 cm high (at budding height) and 5 mm thick planted in a 250 ml Teku® pot (Agromillora Iberia nurseries, S.L., Barcelona, Spain). Sixteen plants of every scion-rootstock combination were transplanted in the winter of 2016, in 45-liters plastic pots (one plant per pot), filled with soil and digested cattle manure in a ratio of 2:1 (v/v). The trees were grown in an open field and were trained as central leader. At planting 250 g of the fertilizer Basacote Starter 6M 16-25-6(+2) (COMPO EXPERT) were applied per tree, whereas during the second and third year, a proper fertilization program was applied to ensure optimum plant growth and productivity. They were drip irrigated and all the necessary phytosanitary actions were taken to ensure plant health and unhindered growth. The area where the trial took place, is characterized by a common Mediterranean type climate with the mean summer daily temperature being 33 °C whereas the mean annual rainfall 400 mm.

Field assessments and measurements

During the first 10 days of July of every year, young, 20 fully expanded leaves from plants of every ‘scion x rootstock’ combination, from the middle part of non-bearing shoots, were collected for nutrient analysis. During the dormant season, the tree height from soil level to the top of the freestanding shoot and the trunk circumference, 5 cm above the graft union, were recorded, to determine tree height and trunk cross-sectional area (TCSA). Furthermore, during the dormant season of the second and third year, the total length of the lateral shoots was also recorded. In August of the second and third year, when the fruits had

obtained the commercial size and typical color for each cultivar, they were collected in two hands, in a 5-7 days interval, and the fruit production per tree was recorded. The yield efficiency (kg tree⁻¹/TSCA) and the cumulative yield efficiency (the sum of 2018 and 2019 yields / TSCA of 2019) were calculated as well. In the autumn of the third experimental year, before the leaves fall, every tree was divided into leaves, upper plant part and roots, the relevant fresh (FW) and dry (DW) weights were recorded and the total plant FW and DW were calculated as well.

Nutrient element concentration determinations

The leaf samples were analyzed in order to determine the concentrations of nitrogen (N), phosphorus (P), potassium (K), calcium (Ca), magnesium (Mg), iron (Fe), manganese (Mn), zinc (Zn), copper (Cu) and boron (B). Each leaf sample was appropriately washed, dried at 80 °C to a constant weight, grounded in a fine powder and dry-ashed in a furnace at 500°C for 5 h; then the ash was extracted with 5% v/v HCl. P concentration was determined by the vanado-molybdo-phosphate yellow color method, B by azomethin-H and K, Ca, Mg, Fe, Mn, Zn and Cu concentration was determined by atomic absorption spectrometry (Varian SpectrAA, 240 FS) in the dry digest. The concentration of N in plant leaves was determined by the indophenol-blue method in the wet digest (Allen, 1989; Karla, 1998).

Experimental design and statistical analysis

The experiment was established as a completely randomized design with four replications; every replication consisted of four trees per scion-rootstock combination. Raw data of the measured variables were analyzed by the Analysis of variance (ANOVA) and significant differences were determined by the LSD test at P<0.05. Discriminant analysis was performed based on common available data of the years 2018 and 2019 (apart from the final measurements of fresh and dry weight of the trees), to assess any possible discrimination among rootstocks budded with the same cultivar and irrespective of the cultivar used. Furthermore, using the above data of the two last years, separately per cultivar-rootstock combination, an hierarchical clustering analysis took place, in order to get a piece of descriptive information on the influences of cultivar or rootstock on the measured variables.

Results

Growth parameters

Year 2017

'Andross' at the end of the first cultivation period (the year 2017) presented lower values of TCSA when grafted on 'GF 677' compared to other rootstocks (Table 1). Tree height was higher on 'RPR' and 'Garnem' followed by that on 'GF 677' and lastly by that on 'RP20'.

Table 1: Growth parameters of two peach cultivars 'Andross' and 'Mercil' grafted on four *Prunus* rootstocks, in 2017

Cultivar	Rootstock	TCSA	Tree height
		(cm ²)	(cm)
'Andross'	'GF 677'	5.68 b*	75.25 b
	'GARNEM'	7.21 a	80.31 a
	'RPR'	6.73 a	83.06 a
	'RP20'	7.03 a	71.08 c
'Mercil'	'GF 677'	5.59 bc	62.43 c
	'GARNEM'	6.57 b	65.43 b
	'RPR'	8.59 a	73.93 a
	'RP20'	4.79 c	52.31 d

*Means within the same column of each variety followed by the same letter do not differ significantly based on LSD at P < 0.05

'Mercil' on the other hand exhibited the lowest TCSA when grafted on 'RP20' and the highest one when 'RPR' was used as a rootstock. Similarly, 'RP20' resulted in the lowest height compared to the other rootstocks, with the highest one being found when 'RPR' was used as a rootstock.

Year 2018

At the end of the second cultivation period, 'Andross' grafted on 'RPR' exhibited the highest values of tree height, fruit production per tree, and yield efficiency (Table 2). On the other hand, 'RP20' gave the highest values of TCSA and 'GF 677' the lowest one.

Regarding 'Mercil', trees grafted on 'RPR' presented the highest values of tree height, fruit production (without a significant difference though from those grafted on 'GF 677') and yield efficiency (again without significant difference from 'GF 677'). The lowest fruit production per tree was recorded on trees grafted on 'RP20', even though those trees presented high values of TCSA along with those grafted on 'Garnem'.

Similar to 'Andross', the total length of the lateral vegetation per tree did not differ among the rootstocks tested.

Year 2019

At the end of the third cultivation period, 'Andross' presented the greatest height, fruit production per tree, yield efficiency, and cumulative yield when grafted on 'GF 677' rootstock (Table 3). 'Garnem' resulted in high TCSA values, along with 'RP20' and similar high values of fruit production per tree with those recorded on 'GF 677'. The lowest production per tree was recorded on 'RPR' as also the lowest yield efficiency, while the lowest tree height was recorded on trees grafted on 'RP20', which also exhibited the lowest cumulative yield efficiency.

'Mercil' plants grafted on 'GF 677' presented the lowest TCSA but the highest values of fruit production per tree and yield efficiency as well as the highest cumulative yield

Table 2: Growth parameters of two peach cultivars 'Andross' and 'Mercil' grafted on four *Prunus* rootstocks, in 2018

Cultivar	Rootstock	TCSA	Tree height	Length of lateral shoots	Fruit production per tree	Yield efficiency
		(cm ²)	(cm)	(cm)	(g)	(g cm ⁻²)
'Andross'	'GF 677'	14.0 c*	113.1 b	284.6 a	389.7 b	27.9 b
	'GARNEM'	17.2 b	115.4 ab	258.1 a	397.1 b	23.1 c
	'RPR'	18.8 ab	119.6 a	236.5 a	616.5 a	32.7 a
	'RP20'	19.2 a	105.2 c	271.5 a	299.8 c	15.5 d
'Mercil'	'GF 677'	16.5 b	97.0 b	279.3 a	530.0 a	32.1 a
	'GARNEM'	18.9 a	102.5a	276.5 a	444.6 b	23.5 b
	'RPR'	17.0 b	103.1 a	299.5 a	517.8 a	30.4 a
	'RP20'	18.3 a	90.5 c	271.6 a	358.4 c	19.6 c

*Means within the same column of each variety followed by the same letter do not differ significantly based on LSD at $P < 0.05$

Table 3: Growth and yield parameters of two peach cultivars 'Andross' and 'Mercil' grafted on four *Prunus* rootstocks, in 2019.

Cultivar	Rootstock	TCSA	Tree height	Length of lateral shoots	Fruit production per tree	Yield efficiency	Cumulative yield efficiency
		(cm ²)	(cm)	(cm)	(g)	(g cm ⁻²)	(g cm ⁻²)
'Andross'	'GF 677'	21.1 b	146.8 a	349.2 a	509.9 a	23.6 a	42.1 a
	'GARNEM'	27.0 a	140.8 ab	433.3 a	510.0 a	19.2 b	34.0 b
	'RPR'	23.0 b	138.2 bc	319.6 a	224.0 c	9.7 d	36.7 b
	'RP20'	29.4 a	134.6 c	425.0 a	385.2 b	13.1 c	23.2 c
'Mercil'	'GF 677'	23.2 c	122.9 b	409.0 ab	525.8 a	23.2 a	46.1 a
	'GARNEM'	26.5 ab	133.2 a	401.4 ab	513.6 a	19.5 b	36.3 b
	'RPR'	23.7 bc	122.4 b	335.3 b	220.6 c	9.3 d	31.1 bc
	'RP20'	27.2 a	115.6 c	434.4 a	353.9 b	13.0 c	26.3 c

*Means within the same column of each variety followed by the same letter do not differ significantly based on LSD at $P < 0.05$

(Table 3). The highest tree height was recorded in plants grafted on 'Garnem', which also presented similar high values of fruit production per tree, as with those grafted on 'GF 677'. On the other hand, plants grafted on 'RP20' presented the highest TCSA as well as the highest lateral shoot length, but the lowest tree height and cumulative yield efficiency.

At the end of the experimentation period, in 2019, it was clear that rootstock had a significant effect on tree growth traits (Table 4). 'Andross' grafted on 'Garnem' presented the highest upper plant part fresh weight, followed those trees grafted on 'RPR', similarly to 'Mercil'. The root fresh weight though was highest when 'RPR' and 'RP20' were used as rootstocks in 'Andross'. Interestingly, 'GF 677' used as a rootstock in 'Andross' resulted in the lowest leaves' fresh weight and total plant fresh weight. The upper plant part dry weight was found to be higher when 'Andross' was grafted on either 'Garnem' or 'RPR', but the root dry.

Leaf nutrient element assessment

Year 2017

Leaf nutrient status of both cultivars was significantly affected by the rootstock used (Table 5). In 'Andross' the highest nitrogen concentration was determined when 'GF 677' was used as rootstock, which also induced significantly high values of potassium, manganese, zinc, copper, and boron concentrations. On the other hand, 'RP20' was the

rootstock which resulted in increased concentration of calcium, magnesium and copper as well, in 'Andross' leaves, while it resulted in low concentration of both nitrogen and boron.

Low concentration of both potassium and manganese was detected in 'Mercil' leaves grafted on 'RP20' rootstock, while 'GF 677' induced significant increases in the concentration of calcium, magnesium, manganese, zinc, and copper.

Year 2018

'GF 677' in the year 2018 resulted again in a high concentration of nitrogen, calcium, magnesium, manganese, zinc, copper, and boron in the leaves of 'Andross' (Table 6). The highest concentration of potassium and iron in the leaves of 'Andross' was determined when 'Garnem' was used as rootstock and the lowest under 'RPR'.

On the other hand, in 'Mercil', the 'RPR' induced a significant increase in the concentration of potassium in the leaves, but the lowest concentration of calcium. 'GF 677' was the rootstock which along with 'RP20' induced a significant increase in the levels of magnesium and zinc, compared to the other two rootstocks. 'RP20' as the rootstock of 'Mercil' resulted also in increased calcium, iron, and manganese concentration but also in the lowest boron one.

Table 4: Growth characteristics of the two peach cultivars 'Andross' and 'Mercil' grafted on four *Prunus* rootstocks after three years of cultivation (2019)

Cultivar	Rootstock	Upper plant part FW	Root FW	Plant leaves FW	Total plant FW	Upper plant part DW	Root DW	Total leaf DW	Total plant DW
(g)									
'Andross'	'GF 677'	331.6 b*	321.7 b	98.5 c	734.1 c	190.7 ab	138.0 b	43.9 b	364.6 a
	'GARNEM'	392.17 a	322.0 b	134.4 ab	833.7 b	218.8 a	141.0 b	57.8 a	412.9 a
	'RPR'	334.7 b	483.7 a	118.0 bc	948.6 a	209.0 a	176.9 ab	50.5 ab	415.6 a
	'RP20'	324.5 b	478.7 a	157.6 a	962.2 a	176.8 b	232.0 a	58.5 a	405.3 a
'Mercil'	'GF 677'	332.5 b	278.4 a	160.0 a	770.9 a	180.5 bc	137.44a	51.8 bc	345.2 b
	'GARNEM'	398.9 a	309.1 a	137.2 ab	845.3 a	224.9 a	141.29a	59.7 ab	425.9 a
	'RPR'	343.4 ab	357.8 a	165.6 a	866.9 a	196.0 b	157.19a	64.8 a	407.5 a
	'RP20'	294.6 b	152.9 b	121.2 b	606.9 b	157.4 c	66.50b	48.0 c	294.4 c

*Means within the same column of each variety followed by the same letter do not differ significantly based on LSD at $P < 0.05$

Table 5: Leaf nutrient element concentrations of two peach cultivars 'Andross' and 'Mercil' grafted on four *Prunus* rootstocks, in 2017

Cultivar	Rootstock	N	P	K	Ca	Mg	Fe	Mn	Zn	Cu	B
(g kg ⁻¹ dw)											
'Andross'	'GF 677'	45.8 a	1.54 a-B	18.5 a	11.1 b	4.33 b	103.4 b	71.4 a-B	25.8 a-B	8.9 a-B	55.8 a-A
	'GARNEM'	37.5 c	1.46 a	16.9 ab-B	7.3 d-B	3.06 c-B	86.5 b	46.2 b-B	20.1 bc-B	7.1 b-B	46.3 b
	'RPR'	41.4 b-B	1.32 a	18.1 a	9.7 c	4.20 b	169.5 a-A	47.5 b	18.1 c-B	7.4 b-B	49.8 ab
	'RP20'	38.2 c	1.27 a-B	16.2 b	12.5 a-B	5.00 a-A	87.5 b	43.9 b	23.5 ab-B	8.4 a-B	36.4 c-B
'Mercil'	'GF 677'	43.9 a	1.85 a-A	17.6 b	12.8 a	5.10 a	100.63 a	95.8 a-A	35.6 a-A	10.9 a-A	47.2 a-B
	'GARNEM'	40.4 a	1.94 a	21.0 a-A	10.4 b-A	4.23 b-A	101.3 a	52.0 b-A	29.4 bc-A	8.6 c-A	53.8 a
	'RPR'	43.8 a-A	1.59 a	19.9 ab	9.9 b	4.63 ab	85.13 a-B	51.5 b	26.3 c-A	9.4 bc-A	47.6 a
	'RP20'	40.3 a	2.06 a-A	15.3 c	13.2 a-A	4.43 b-B	101.43 a	41.6 c	31.6 ab-A	10.3 ab-A	47.7 a-A

Means within the same column of each variety followed by the same small letter do not differ significantly based on LSD at $P < 0.05$. Means within the same column and for the same rootstock followed by the different capital letter differ significantly based on Student's *T*-test at $P < 0.05$

Table 6: Leaf nutrient element concentrations of two peach cultivars 'Andross' and 'Mercil' grafted on four *Prunus* rootstocks, in 2018

Cultivar	Rootstock	N	P	K	Ca	Mg	Fe	Mn	Zn	Cu	B
(g kg ⁻¹ dw)											
'Andross'	'GF 677'	37.5 a-A	2.08 a	13.5 b-A	8.0 a-A	3.53 a-A	95.6 ab-A	13.3 a-B	34.7 a-A	10.7 a-B	59.1 a
	'GARNEM'	28.1 b-A	1.83 a	15.9 a-A	7.2 b-A	2.93 c	101.4 a-A	9.1 b-B	24.5 b	10.2 b-B	48.3 b
	'RPR'	29.3 b-A	1.87 a	10.3 d-A	5.7 c-A	2.50 d	87.3 b-A	9.5 b-B	24.2 b	9.3 c-B	61.8 a-A
	'RP20'	27.1 b	1.82 a-B	12.0 c-A	7.7 ab	3.16 b	96.2 ab-A	12.9 a-B	26.6 b	9.6 c-B	37.1 c
'Mercil'	'GF 677'	24.2 a-B	2.05 a	10.4 c-B	5.0 b-B	2.96 a-B	61.5 b-B	17.6 b-A	29.6 a-B	13.3 a-A	58.5 a
	'GARNEM'	23.8 a-B	2.14 a	11.9 b-B	5.4 b-B	2.73 b	56.3 c-B	10.2 d-A	22.7 b	11.8 a-A	56.0 a
	'RPR'	23.7 a-B	1.66 a	13.0 a-B	4.1 c-B	2.56 b	62.1 b-B	11.3 c-A	24.2 b	12.4 a	50.6 a-B
	'RP20'	25.9 a	2.03 a-A	11.1 c-B	7.6 a	3.03 a	68.7 a-B	19.8 a-A	28.0 a	12.8 a	31.7 b

Means within the same column of each variety followed by the same small letter do not differ significantly based on LSD at $P < 0.05$. Means within the same column and for the same rootstock followed by the different capital letter differ significantly based on Student's *T*-test at $P < 0.05$

Year 2019

At the end of the experimentation period (2019) 'Andross' exhibited the highest concentration of phosphorus and calcium when grafted on 'GF 677' (Table 7). Nitrogen, magnesium, iron, and zinc were found in high concentration in plants grafted on 'GF 677' and 'RP20', with the latter rootstock inducing a significant increase of potassium and copper in the leaves, compared to the other rootstocks.

'GF 677' was the rootstock on which 'Mercil' presented the highest values of nitrogen, manganese, zinc, and copper concentration in the leaves. 'RP20' on the other hand induced a significant increase in the levels of

both P and K but resulted in the lowest concentration of copper.

The discriminant analysis revealed very interesting results. Concerning the effects of rootstocks on 'Andross' cultivar, it was obvious that both 'GF 677' and 'RP20' exhibited significant and distinct effects on the annual growth and mineral nutrient concentration of the cultivar, as much from each other as from 'Garnem' and 'RPR' too (Fig. 1). The latter two rootstocks could not be distinguished, as they shared a common area.

Concerning the effects of the rootstocks on 'Mercil' cultivar, on the other hand, all rootstocks were clearly distinguished

Table 7: Leaf nutrient element concentrations of two peach cultivars 'Andross' and 'Mercil' grafted on four *Prunus* rootstocks, in 2019

Cultivar	Rootstock	(g kg ⁻¹ dw)					(ppm)				
		N	P	K	Ca	Mg	Fe	Mn	Zn	Cu	B
'Andross'	'GF 677'	28.8 a	3.96 a-A	12.7 ab-A	11.7 a-A	2.80 a	98.5 a	20.8 b-B	30.8 a-A	8.2 b-B	52.1 b
	'GARNEM'	25.0 b	2.98 b	11.2 c	8.7 b	2.86 a	67.1 c-B	16.0 c	19.0 b	7.6 c-B	59.5 ab
	'RPR'	24.1 b	2.06 c-B	11.3 bc	7.8 b	2.36 b	87.9 b	16.2 c	19.8 b	5.5 d	65.6 a
	'RP20'	28.3 a-A	3.30 b-B	12.9 a-B	8.4 b-A	2.73 a	106.1 a-A	23.5 a-A	29.0 a-A	9.9 a	61.1 ab
'Mercil'	'GF 677'	29.4 a	3.00 b-B	11.3 d-B	10.4 a-B	3.00 a	94.5 a	36.6 a-A	25.4 a-B	11.0 a-A	51.9 b
	'GARNEM'	24.4 b	3.44 b	12.7 c	10.3 a	3.13 a	85.7 b-A	15.6 c	19.7 c	10.1 b-A	63.0 a
	'RPR'	24.5 b	3.14 b-A	14.3 b	9.7 a	2.76 b	100.2 a	21.1 b	22.7 b	10.7 ab	70.2 a
	'RP20'	25.5 b-B	4.39 a-A	15.5 a-A	7.2 b-B	2.66 b	82.2 b-B	21.8 b-B	22.7 b-B	9.0 c	60.2 ab

Means within the same column of each variety followed by the same small letter do not differ significantly based on LSD at $P < 0.05$. Means within the same column and for the same rootstock followed by the different capital letter differ significantly based on Student's *T*-test at $P < 0.05$

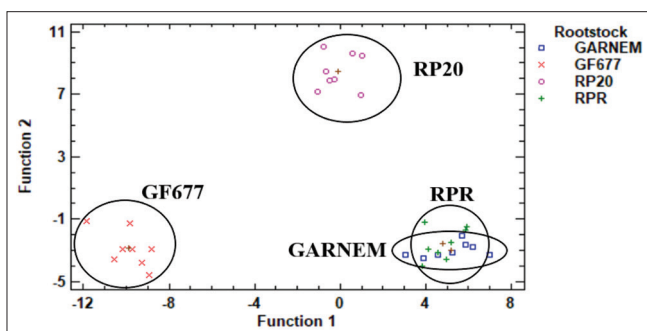


Fig 1. Discriminant analysis of the effects of rootstocks grafted with 'Andross' cultivar.

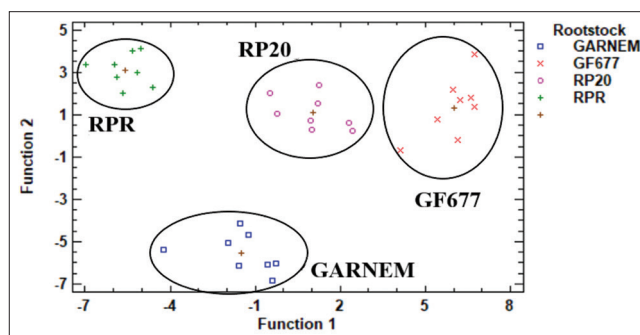


Fig 2. Discriminant analysis of the effects of rootstocks grafted with 'Mercil' cultivar.

from each other (Fig. 2), revealing significant impacts on the cultivar growth and leaf nutrient concentration.

When the rootstocks were examined irrespective of the grafted cultivar, 'RP20' and 'GF 677' were separated from each other and from 'Garnem' and 'RPR', which shared a common area, indicative of the distinct effects of the two former rootstocks on cultivar's growth and nutrient status.

The clustering analysis though revealed some other interesting traits, as the effect of 'Garnem' and 'RP20' seemed not to depend on the cultivar grafted (Fig. 4). Both cultivars grafted on the same rootstock from the two previously mentioned were grouped, based on the agronomical traits and leaf nutrient concentration assayed in the present experiment (Fig. 4). On the other hand, 'GF677' and 'RPR' effect seemed to be dependent on the cultivar grafted, revealing a strong influence of the cultivar on the cultivar-rootstock combination.

DISCUSSION

Based on the discriminant analyses employed, it became obvious that there was a great interaction between scion and rootstock, while 'GF 677' was the only rootstock with distinct differences from the others, in both cultivars tested. On the other hand, when the cultivar was not taken into

account, it became obvious that 'RP20' and 'GF 677' exert distinctly different effects, differing from both 'Garnem' and 'RPR', which exhibited similar behavior. The cluster analysis revealed also the great influence of both 'Garnem' and 'RP20' on the parameters studied in the present trial. In fact, the effect of these rootstocks was so strong, that overpassed any possible influence the grafted cultivar may have on the physiological traits studied here, as both cultivars grafted on the same rootstock were grouped together and far from the other groups, revealing the vigorous and dwarfing nature of 'Garnem' and 'RP20' respectively.

Many researchers have used TCSA as an index of tree growth and yield efficiency (Zarrouk et al., 2005; Jiménez et al., 2011; Font i Forcada et al., 2012; Mestre et al., 2015; Reig et al., 2016; Font i Forcada et al., 2020; Reig et al., 2020). To our knowledge, there is no data on the effect of rootstock and cultivar during the first years after budding. Therefore, we have used the available literature involving young as well as mature trees in full production. In both 2018 and 2019, 'RP20' exhibited the highest TCSA, along with 'Garnem' (in 2018 in 'Mercil' and 2019 in 'Andross'). At the same time the most dwarf rootstock, i.e. 'RP20', presented the lowest tree height in all cultivar-rootstock combinations, while 'Garnem' and 'RPR' the highest, when used as rootstock for 'Mercil', in accordance with the literature (Jiménez et al., 2011; Font i Forcada et al.,

2012). Based on the existing literature so far, there is no conclusive data on the effect of ‘GF 677’ and ‘RPR’ on TCSA, as both significant and non-significant differences have been reported (Mestre et al., 2015; Reig et al., 2016; Font i Forcada et al., 2020; Reig et al., 2020). Nonetheless, the fact that ‘RP20’ presented the highest TCSA during the first years after planting is not a common feature, as in most cases the most dwarf rootstocks are those with the lowest TCSA and overall growth, as has been reported in several almond cultivars grafted on both ‘Garnem’ and ‘RP20’ (Ben Yahmed et al, 2016). As tree height was consistently the lowest in both cultivars on ‘RP20’, it can be assumed that at least during the first years, photosynthates were not used for canopy growth and shoot elongation but rather deposited on other tree parts and especially trunk, increasing thus its diameter. Similar results have been also reported by Opazo et al. (2020). On the other hand, the ‘RP20’ response seemed to depend on the cultivar budded,

as its combination with ‘Mercil’ resulted in the lowest total plant dry weight, while when ‘Andross’ was budded on it, no differences were detected among rootstock-cultivar combinations. It seems that during the first years ‘RP20’ dwarfing properties are mostly visible with ‘Mercil’ than with ‘Andross’, revealing a significant effect of cultivar on tree growth pattern, at least for the first years after budding, as this may change later on (Layne, 1994), similarly to that reported by Opazo et al. (2020).

The yield was generally higher when cultivars were budded onto vigorous rootstocks, such as ‘Garnem’ and ‘GF 677’. Interestingly enough, both cultivars presented lower yield during the third year, when budded onto ‘RPR’, which followed the high yield of the previous year, indicating a tendency of alternate bearing, especially after a relatively warm winter, as has been observed with other rootstocks too (Ben Yahmed et al., 2020). Higher yield efficiency of ‘GF 677’ compared to ‘RPR’ has been reported by other authors too (Mestre et al., 2015; Reig et al., 2016), while no significant differences between the former rootstocks have been also reported (Font i Forcada et al., 2020). Although yield efficiency is expected to be higher in less vigorous rootstocks, due to their reduced growth and TCSA (Zarrouk et al., 2005; Bouhadida et al., 2009; Gogorcena, and Moreno, 2012; Font i Forcada et al., 2016; Mestre et al., 2017), this was not the case in this study, similarly to that reported by Reig et al. (2020) comparing ‘RP20’ and ‘GF 677’, which exhibited similar yield efficiency.

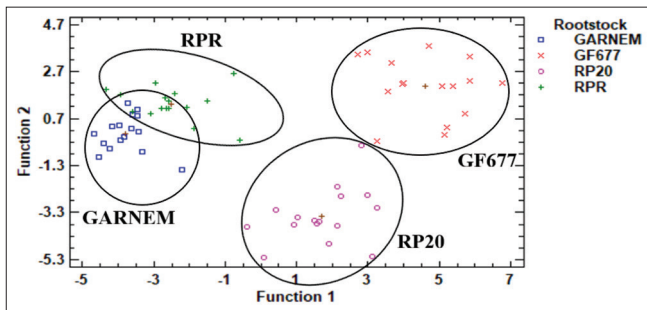


Fig 3. Discriminant analysis of the effects of rootstocks irrespective of the grafted cultivar.

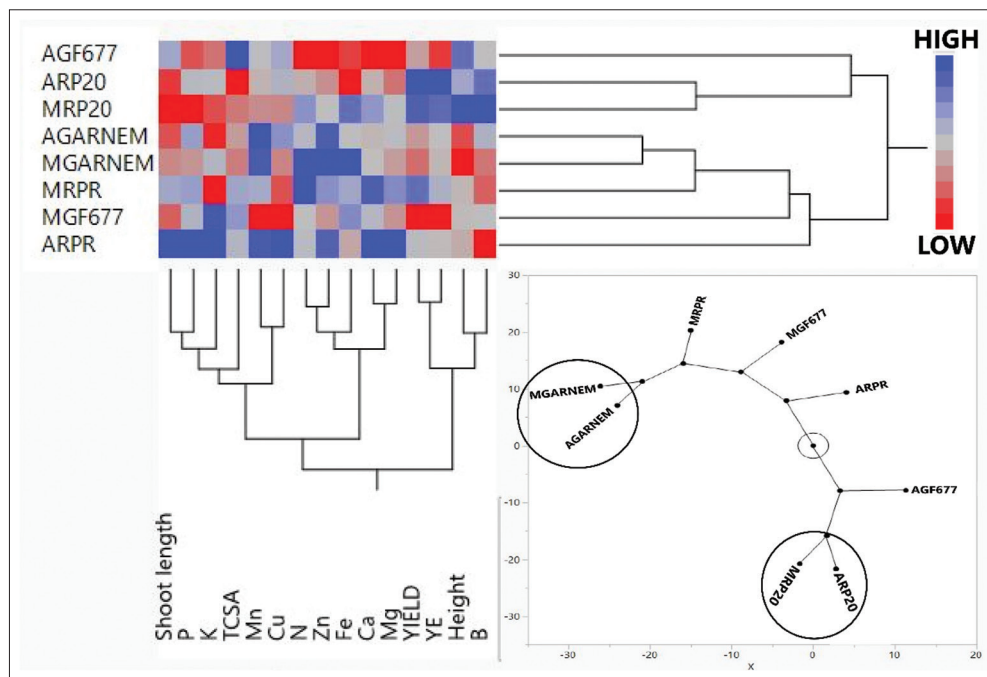


Fig 4. Hierarchical agglomerative cluster analysis and constellation plot for the different rootstock-cultivar combinations, based on agronomical traits and leaf nutrient concentration.

Rootstocks had a significant effect on nutrient concentration found in the leaves of the two cultivars, while the cultivar itself, had also a significant effect, similar to other reports (Caruso et al., 1996; Zarrouk et al., 2005; Remorini et al., 2008; Mestre et al., 2015; Mestre et al., 2017; Font i Forcada et al., 2020). The year-to-year variation has been also reported (Jimenes et al., 2018) as in the present experiment.

The differences observed among rootstocks regarding leaf nutrient concentration of the budded cultivar are attributed to various factors. According to some authors, the final concentration of the nutrient in the leaves depends on the absorption, transportation, redistribution, and use of this nutrient (Jimenes et al., 2018; Shahkoomahally et al., 2020). Since rootstock is the plant part responsible for the absorption and transportation (to some extent) of the nutrient, any rootstock property influencing these two functions (root volume, root morphology, root xylem vessel characteristics), may have a significant effect on nutrient concentration found in the leaves (Meland, 2010; Mestre et al., 2015; Mestre et al., 2017; Ben Yahmed et al., 2020).

It is believed that dwarfing rootstocks are less efficient in absorbing nutrients from the soil, especially some macronutrients, due to their restricted root volume and narrow xylem vessels (Zarrouk et al., 2005; Mestre et al., 2015; Ben Yahmed et al., 2020; Shahkoomahally et al., 2020). In the present experiment 'RP20' was the most dwarfing rootstock, with the rest of the rootstocks are considered invigorating ones. The differences found in the literature among rootstocks have been attributed, among others, to the genetic background of each rootstock (Zarrouk et al., 2005; Ben Yahmed et al., 2020; Shahkoomahally et al., 2021). It seems that peach-almond rootstocks, such as 'GF677' and 'Garnem', respond better to iron deficiency under calcareous soils (Zarrouk et al., 2005; Assimakopoulou et al., 2011; Mestre et al., 2015; Ben Yahmed et al., 2020), as well as some plum rootstocks (Mestre et al., 2015; Ben Yahmed et al., 2020), while peach cultivars grafted on all these rootstocks present high Ca concentration too (Ben Yahmed et al., 2020)

Among the tested rootstocks, it seems that 'Andross' in 2018, as well as both cultivars in 2019, presented high leaf N concentration when budded on 'GF 677' (2018 and 2019 were the years when first fruit production occurred). In addition, 'Andross' on 'RP20' (2019) and 'Mercil' on 'RP20' (2018) presented also high N levels in the leaves, close to that determined on 'GF 677'. Higher N content on trees grafted on the dwarfing 'RP20' compared to those grafted on the invigorating 'GF 677' has been reported by Reig et al. (2020), while 'GF 677' has been found to enhance N concentration compared to the lower vigor rootstock 'Adarcias' (Zarrouk et al., 2005; Mestre et al.,

2015), indicating that its distribution cannot be entirely attributed to the rootstock's vigor.

Leaf P and K concentration seemed to depend on the rootstock used, which was especially evident during the third year, while the cultivar presented a significant influence on their concentration too. This fact indicates that both rootstock and cultivar play a significant role in P and K distribution and translocation. Similar concentrations of P have been determined in 'GF 677' and 'RPR' (Font i Forcada et al., 2020), in 'GF 677' and 'RP20' (Iglesias et al., 2019; Reig et al., 2020), which was the case in the present trial only during 2018. 'GF 677' on the other hand, has been found to induce higher P concentration in the leaves of "Queen Giant" nectarine compared to 'Garnem', similarly to 'Andross' in the present trial. 'RPR' has been found to induce higher K concentration in the leaves of the budded cultivar compared to 'GF 677' (Mestre et al., 2015), which was the case in the present trial concerning 'Mercil'. On the other hand though, 'Andross' presented exactly the opposite results, indicating that K nutrition is being controlled also by the cultivar itself and is not a specific trait of the rootstock, as it has been proposed for plum based rootstocks against peach based ones (Mestre et al., 2015).

Calcium on the other hand was always found in low or even the lowest concentration on the leaves of both cultivars when budded on 'RPR', similar to that reported by Mestre et al. (2015) and Font i Forcada et al. (2020), who attributed this difference on the tendency of plum based rootstocks to present lower efficiency towards Ca. This should be taken into consideration though, since Ca is the nutrient responsible, among others, for the storability and firmness of the fruit, suggesting that a higher Ca supply could be necessary for peaches budded on 'RPR' rootstock.

Magnesium concentration was always high in leaves of both cultivars budded on 'GF 677', while 'RPR' was the rootstock on which both cultivars presented lowest values, indicating that it has a low efficiency towards Mg too, as was also reported by Font i Forcada et al. (2020).

'Andross' trees budded on 'RPR' presented both in 2018 and 2019 the lowest values of Mn, Zn, and Cu but high B concentration. On the other hand both cultivars when budded on 'Garnem' were characterized by low values of Mn and Zn, indicating that micronutrient absorption and translocation is not strictly a genetic trait of the rootstock (plum and peach based rootstocks respectively), similarly to that reported with many other rootstocks (Zarrouk et al., 2005; Font i Forcada et al., 2020). Manganese was found in high concentration in plants budded on 'GF 677' as well as in 'RP20' (only in 2018), in accordance with that

reported by Iglesias et al. (2019) and Reig et al. (2020), who did not find any difference between these two rootstocks and similar to Zarrouk et al. (2005) who found higher Mn concentration on ‘GF 677’.

CONCLUSIONS

Under the environmental conditions that prevailed and the cultivation practices applied in our experiment, ‘GF 677’ followed by ‘Garnem’ could be considered as more suitable rootstocks for both ‘Andross’ and ‘Mercil’ cultivars compared to others tested, based mainly on the yield characteristics. One must have in mind though that as vigorous rootstocks these should be planted at higher distances, forming thus a lower density orchard. When a high-density peach orchard is planned, the dwarf ‘RP20’ is the rootstock of choice, as it offers lower vigor and overall satisfactory yield efficiency, especially if plant spacing using ‘RP20’ can be reduced to half. As both rootstock and cultivar have been found to influence tree growth and yield efficiency, at least during the first years after planting, the fertilization program to be applied should be issued based on both rootstock, cultivar and their interaction properties.

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Author contribution

EN and PAR designed the experiment, EN executed the majority of the trial in the field and laboratory, PAR supervised methods, AA, executed part of the trial in the laboratory, EN, PAR, AA wrote and revised the manuscript.

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