**Performance of peach and plum based rootstocks of different vigour on a late
peach cultivar in replant and calcareous conditions**

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Abstract

The field performance of fifteen peach and plum based rootstocks of different vigour
(Adesoto, Evrica, Garnem, GF 677, HM-2, Krymsk[®] 1, PAC 9801-02, PAC 960, PAC
9907-02, PAC 9917-26, PAC-MUT, PADAC 9907-23, ROOTPAC[®] 40, ROOTPAC[®]
70 and Tetra) grafted with ‘Calrico’ cultivar was compared after 7 years of
establishment on an *Armillaria* infested replant site. ‘Calrico’ is a selected clone of
“Calanda” late peach cultivar. Differences in parameters such as tree survival, leaf
chlorophyll content, vigour, yield, cumulative yield, yield efficiency and fruit size were
analyzed among rootstocks. All PAC 9801-02 and Tetra trees survived and the mortality

rate was low in Evrica, PADAC 9907-23 and ROOTPAC® 40. The rest of genotypes showed higher mortality rates. Leaf chlorophyll concentration was higher when grafted on Adesoto and Evrica and lower when grafted on PAC 9907-02 and PAC 9917-26. Garnem and PADAC 9907-23 were the most vigorous rootstocks. The highest yield efficiency was induced by Krymsk® 1 and PAC 9801-02 due to their lower vigour. The highest fruit weight was also induced by Krymsk® 1 but its cumulative yield was low. Other rootstocks that showed high fruit weight and cumulative yields were ROOTPAC® 70 and Tetra. Overall, Evrica, PAC 9801-02, ROOTPAC® 40 and Tetra were among the best adapted to soil sickness and calcareous soil showing a good agronomic performance. The first three rootstocks (Evrica, PAC 9801-02, ROOTPAC® 40) also exhibited a high capacity to control tree vigour associated with high yield efficiency.

Keywords: *Armillaria mellea*, Iron chlorosis, *Prunus*, Replant, Rootstock, Vigour

1. Introduction

“Calanda” peach [*Prunus persica* (L.) Batsch.] is a native Spanish variety highly profitable and very much appreciated in Europe due to the large size, non melting flesh, clingstone and late harvest of fruits (Fernández et al., 2009; Ferrer et al., 2005). Its production is concentrated in the Calanda area (Ebro Valley, Spain), a region that suffers, among other problems, replant disease and iron chlorosis, such as almost all the Mediterranean area.

Replant disease in stone fruits often occurs when trees are grown in a soil that had previously supported the same or similar plant species leading to reductions in plant growth, crop yields and shortening of the productive life of the orchard (Bent et al., 2009; Reighard et al., 2008). Biotic factors, such as soil-borne pathogens, adverse soil

microbiota and decomposition of the remaining roots from the previous crop, as well as abiotic factors, such as soil nutrition, structure and decline in soil organic matter content, have been suggested to additionally contribute to replant disease etiology (Calvet et al., 2000; Manici and Caputo, 2010; Bent et al., 2009). In *Prunus* trees, species of *Armillaria* fungus genus has been implicated in root rot and peach tree mortality (Beekman and Pusey, 2001). Much research has been conducted on soil fumigation as a solution to replant disease in the past decades (Browne et al., 2006; Utkhede, 2006). However, research is needed to breed and evaluate disease resistant or tolerant crops as a more sound and environmental friendly approach (Browne, 2009; Utkhede, 2006). Replant disease is becoming a primary challenge in peach production as the industry moves forward replanting orchards with a newer generation of vigour-controlling rootstocks better adapted to adverse conditions.

Iron chlorosis is mainly the result of the low iron bioavailability in calcareous soils and the difficulty of Fe acquisition by the roots (Hell and Stephan, 2003). Iron is an essential micronutrient for plant growth and development because of its importance in numerous cellular functions (Jiménez 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 (Almaliotis et al., 1995; Álvarez-Fernández et al., 2003). If amended, it increases orchard management costs derived from Fe-quelate treatments (Abadía et al., 2004; Sanz et al., 1992). The genetic approach to prevent iron chlorosis is based on the use of tolerant rootstocks (Gogorcena et al., 2004; Rombolà and Tagliavini, 2006).

The use of new rootstocks that are more resistant to abiotic and biotic stresses as well as inducing adequate growth represents the best solution to increase orchard productivity and efficiency (Reighard et al., 2008). In the Mediterranean area, the almond × peach hybrid GF 677 has been one the most utilized rootstocks in the last

decades. It has high tolerance to iron deficiency, but induces excess of vigour (Cinelli and Loreti, 2004; Zarrouk et al., 2005). Early experimental data indicated that other rootstocks (e.g. Barrier and Cadaman) show equally satisfying, if not actually better, field performance under replanting conditions (Massai and Loreti, 2004). New rootstocks recently released or under selection may improve peach production with reduction of labour costs.

The present study was carried out with ‘Calrico’ late peach cultivar, grafted onto peach and plum based rootstocks of different vigour and grown on heavy and calcareous soil in a replant site. The objective was to evaluate the performance of the rootstocks in these conditions, through tree survival, leaf SPAD value, vegetative growth, yield and fruit size analysis.

2. Materials and methods

2.1. Plant material

Fifteen peach rootstocks were compared in one trial established in February 2002. Rootstock under evaluation included peach and plum hybrids (Table 1). Most rootstocks are experimental genotypes in their late stages of selection or new commercial *Prunus* rootstocks of Spanish, Italian and Russian origin. Some of them are inter-specific hybrids bred specifically for several resistance and tolerance traits. Adesoto and GF 677 rootstocks were used as medium and high vigour reference rootstocks. They were grafted with the late ripening non-melting clingstone ‘Calrico’ peach cultivar in September 2002.

2.2. Field trial

Table
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The trial was carried out in the Experimental Farm of Woody Plants of the Department of Food and Agriculture of the Aragón Government in Alcañiz (Teruel, Spain), in the “Calanda” peach denomination growing area, on a calcareous soil, with 48% total calcium carbonate, 11-12% active lime, pH in water 8.2 and a silt-loam texture. Trees were planted at 6.5 m × 5 m and trained in the central leader system. Trial was established one year after uprooting a 14-year-old peach orchard that was grafted on GF 677. The replant site had a history of high mortality rate among trees caused by the oak root fungus *Armillaria mellea* Vahl P. Kumm. The orchard was drip irrigated and managed following the usual local procedures. The experiment was established in a randomized block design with eight single-tree replications for each rootstock. Guard rows were used to preclude edge effects.

2.3. Chlorophyll analysis

The chlorophyll (Chl) concentration per unit leaf area was estimated in the field, using a SPAD 502 meter (Minolta Co., Osaka, Japan). Thirty leaves per tree, selected from the middle of bearing shoots located all around the tree canopy, were measured with the SPAD to obtain an average leaf Chl concentration representative of the leaves belonging to the outer part of the tree canopy. Measurements were carried out at the beginning and end of the trial evaluation: at 122 and 172 after full bloom (DAFB) in 2005 and 2009, respectively.

2.4. Growth measurement and yield

Trunk girths were measured during the dormant season since 2005 at 20 cm above the graft union, and the trunk cross-sectional area (TCSA, in cm²) was calculated. Dead trees were recorded each year at the time when growth measurements were taken.

Cumulative yield per tree and yield efficiency (cumulative yield in kg per final TCSA) of each rootstock were computed from the harvest data. At each harvest, 50 fruits were sampled from each tree and they were used to determine fruit weight (g).

2.5. Analysis of data

Data were evaluated by two-way variance (ANOVA) analysis with the program SPSS 17.0.0 (SPSS, Inc., Chicago, USA). When the F test was significant, means were separated using Duncan's multiple range test ($P \leq 0.05$). Regression analysis was carried out by Pearson's correlation.

3. Results

3.1. Tree mortality

At the seventh year after grafting, replant conditions generated varying levels of tree mortality, being higher with the peach based rootstocks (Fig. 1). PAC 960 rootstock experienced the highest tree mortality with 75% of dead trees (only 2 trees survived); thereby it was not included in the rest of the study. Other rootstocks that suffered high tree mortality were HM-2 and PAC 9907-02 with 63% and 50% of dead trees, respectively. HM-2 experienced progressive tree mortality with 38 % and 50 % of dead trees in 2007 and 2008, respectively (Fig.1). However, PAC 9907-02 suffered high tree mortality at the end of the experiment with 0% and 13% of dead trees in 2007 and 2008, respectively. At the seventh year after grafting, low mortality was found in Evrica, PADAC 9907-23 and ROOTPAC® 40 with only a single dead tree (13%). In contrast, all trees grafted on the plums PAC 9801-02 and Tetra survived well exhibiting homogenous growth. Isolations from root samples taken from dying trees confirmed the presence of the fungus *A. mellea* in most cases.

3.2. SPAD values

Chlorophyll concentration, as determined by SPAD, was significantly affected by rootstocks the years it was measured (Table 2). In the first bearing year, ‘Calrico’ leaf chlorophyll concentration was significantly lower when grafted on PAC 9907-02 and PAC 9917-26. In 2009, ‘Calrico’ showed a significant higher leaf chlorophyll concentration on Adesoto and Evrica, followed by GF 677 and PAC 9801-02, and again a lower concentration on PAC 9917-26, followed by PAC 9907-02 (Table 2). On the remaining rootstocks chlorophyll concentration was intermediate with SPAD values ranging from 36.9 to 38.0 (Table 2).

Table
2

3.3. Vegetative growth

Tree size, as measured by TCSA, was significantly affected by rootstocks starting from the third year after grafting, 2005 (Fig. 2). In 2009, ‘Calrico’ showed the higher TCSA values on Garnem and PADAC 9907-23 (239.6 and 239.0 cm², respectively) and lower values on Krymsk[®] 1 and PAC 9801-02 (58.7 and 76.5 cm², respectively); on the other rootstocks tree growth was intermediate (Table 3 and Fig. 2).

Fig. 2

Table
3

3.4. Tree yield

In the first bearing year (2005), yield was very low, and there were no significant differences among rootstocks (Table 4). However, after 2005, differences among rootstocks became evident. In the seventh year after grafting (2009), HM-2 and PAC 9917-26 produced the highest yield per tree, followed by Garnem, GF 677, PAC 9907-02, PADAC 9907-23, ROOTPAC[®] 40, ROOTPAC[®] 70 and Tetra (Table 4). The lowest yield was recorded on Krymsk[®] 1, followed by PAC 9801-02. A significant high

correlation ($P \leq 0.001$) was observed between yield and vigour in 2006 and 2009 ($r = 0.709$ and $r = 0.728$, respectively).

By year seventh year after grafting, the cumulative yield was greater on HM-2 and ROOTPAC[®] 70 rootstocks (191.4 and 196.9 kg tree⁻¹, respectively), followed by GF 677, PADAC 9907-23 and Tetra, whereas the highest yield efficiency was recorded on PAC 9801-02 (1.62 kg cm⁻²), followed by Evrica, Krymsk[®] 1 and ROOTPAC[®] 40 (Table 3). In contrast, Garnem induced the lowest yield efficiency (0.66 kg cm⁻²), followed by PAC 9907-02, PAC 9917-26, PAC-MUT and PADAC 9907-23 (Table 3).

3.5. Fruit weight

Fruit weight was affected by rootstock except in 2009 (Table 5). In previous years, Krymsk[®] 1 showed the tendency to induce bigger fruit size, followed by Garnem, ROOTPAC[®] 40, ROOTPAC[®] 70 and Tetra. In contrast, PAC 9907-02 induced the lowest fruit weight, followed by GF 677 and HM-2 (Table 5). Fruit weight was variable over the years. In general, smaller fruits were harvested in 2009 when yield was higher. No significant correlation was found between yield and fruit weight over the 5 years of study.

4. Discussion

The performance of ‘Calrico’ late peach cultivar grafted onto fifteen peach and plum based rootstocks grown on a calcareous replant soil, one of the most important problems of the Mediterranean area, derived from the intensive cropping of pome and stone fruit production, was assessed in the present study.

Prunus species are considered highly sensitive to replant problems (Browne et al., 2006). Mortality rate was very high for some of the rootstocks tested, i.e. PAC 960,

HM-2 and PAC 9907-02, seven years after planting probably due to the replant disease. However, Evrica, PADAC 9907-23, ROOTPAC[®] 40, and especially Tetra and PAC 9801-02 rootstocks (all trees survived) seem to tolerate better replant conditions. In this study, GF 677, a rootstock considered resistant to this anomaly (Loreti and Massai, 2006a; Massai and Loreti, 2004), experienced a 25% mortality rate. From the genetic standpoint, it is noteworthy that the plum based rootstocks (no more of 25% mortality rate) had a better survival rate than the peach based rootstocks.

The indirect measurement of leaf chlorophyll concentration by SPAD readings has been used as an indicator of iron chlorosis tolerance in *Prunus* trees (Jiménez et al., 2008). The high SPAD readings of Adesoto, Evrica and Tetra were previously reported in a plum trial established on a calcareous soil (Zarrouk et al., 2006). Adesoto and other rootstocks that in the present study induced high SPAD values in leaves, i.e. GF 677, were previously classified as tolerant to iron chlorosis according to their capacity to reduce iron from the soil (Jiménez et al., 2008). PAC 9907-02, a rootstock classified as sensitive (Jiménez et al., 2008), showed the lowest SPAD values with clear visual symptoms of iron deficiency.

Tree vigour was affected by rootstocks, being Krymsk[®] 1 and PAC 9801-02 the most vigour-controlling rootstocks. The low vigour of Krymsk[®] 1 has already been reported (Peppelman et al., 2007; Reighard et al., 2008). In contrast, Garnem was the most invigorating rootstock, in agreement with the genotype description (Felipe, 2009) and field evaluation (Zarrouk et al., 2005). Tetra showed a similar vigour in comparison to GF 677. However, in a preliminary study on Japanese plum ('Shiro' cultivar), Tetra resulted in a very low vigour (31 % of GF 677 vigour; Sottile et al., 2007). This could be explained by a better adaptation of Tetra to replanted soils that leads to minimal reductions in plant growth.

The dwarfing rootstock Krymsk[®] 1 also induced the lowest yield, but it was among the most yield efficient rootstocks. On the contrary, Krymsk[®] 1 has shown low yield efficiencies in another peach trial (Reighard et al., 2008). The low vigour and high yield efficiency of PAC 9801-02, Evrica, Krymsk[®] 1 and ROOTPAC[®] 40 make them ideal for high density peach orchards allowing the possibility of establishing pedestrian orchards with the benefits of reducing labour costs, especially at pruning and harvest. The last rootstock, ROOTPAC[®] 40, also induced high yield per tree, not different to more invigorating rootstocks such as Garnem, GF 677, ROOTPAC[®] 70 and Tetra. Unfortunately, the plum dwarfing rootstocks Evrica and Krymsk[®] 1 are incompatible with many peach cultivars (Zarrouk et al., 2006) reducing their potential use as rootstocks for peach and nectarine cultivars.

For decades a more efficient production system has been considered a priority for the peach industry in Spain. Low vigour rootstocks for use with peach do exist commercially, but their use is very limited in warm Mediterranean environments (Loreti and Massai, 2006b). Their main drawbacks are the excessive needs of chill units, lack of compatibility with many peach and nectarine cultivars (in the case of plums and plum hybrids), and susceptibility to iron chlorosis and soil-borne pathogens, such as fungi and root-knot nematodes, so common in many peach growing regions of Spain (Pinochet, 1997). The introduction of PAC 9801-02 and the recently released ROOTPAC[®] 40 open the possibility of establishing more efficient peach production systems in regions where high density orchards were not feasible mainly due to the lack of adequate genetic material.

Regarding fruit quality, Krymsk[®] 1 showed the tendency to induce bigger fruit size, followed by Tetra, ROOTPAC[®] 40, ROOTPAC[®] 70 and Garnem. However, all rootstocks exhibited acceptable fruit weight similar to “Calanda” type peaches harvested

at appropriate maturity stage (Ferrer et al., 2005). ‘Calrico’ cultivar induced higher fruit weight in comparison to other peach cultivars established in similar calcareous soil conditions, such as ‘Catherina’, ‘Flavortop’ (Albás et al., 2004) and other breeding progenies (Cantín et al., 2010).

5. Conclusions

Late peach cultivar trees grafted on Evrica, PAC 9801-02 and ROOTPAC® 40 appear to control tree size and to induce high yield efficiency. Tetra, a more invigorating rootstock, seems to induce high yields. The four rootstocks seem to be well adapted to calcareous and replanted soil conditions and can perform even better than the commonly used genotype GF 677.

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Figure captions

Fig. 1. Tree mortality rate (%) from the third (2005) to the seventh (2009) year after grafting. Percentage values right side of the bars indicate accumulated mortality rate in 2009.

Fig. 2. Effect of rootstock on trunk cross-sectional area (TCSA) of ‘Calrico’ peach cultivar from the third (2005) to the seventh (2009) year after grafting. Vertical lines indicate LSD ($P \leq 0.05$)

Table 1

List of studied rootstocks, description and origin.

Rootstock	Species	Origin ^a
Adesoto	<i>P. insititia</i>	CSIC, Spain
Evrica	<i>(P. besseyi × P. salicina) × P. cerasifera</i>	KEBS, Russia
Garnem	<i>P. persica × P. amygdalus</i>	CITA, Spain
GF 677	<i>P. persica × P. amygdalus</i>	INRA, France
HM-2	<i>(P. persica × P. amygdalus) × (P. persica × P. amygdalus)</i>	AI, Spain
Krymsk [®] 1 ^b	<i>P. tomentosa × P. cerasifera</i>	KEBS, Russia
PAC 9801-02	<i>P. besseyi × P. cerasifera</i>	AI, Spain
PAC 960	<i>P. persica × P. amygdalus</i>	AI, Spain
PAC 9907-02	<i>(P. persica × P. amygdalus) × (P. persica)</i>	AI, Spain
PAC 9917-26	<i>(P. persica × P. amygdalus) × (P. persica)</i>	AI, Spain
PAC-MUT	<i>P. persica × P. amygdalus</i>	AI, Spain
PADAC 9907-23	<i>(P. persica × P. amygdalus) × (P. persica)</i>	CSIC-AI, Spain
ROOTPAC [®] 40	<i>(P. persica × P. amygdalus) × (P. persica × P. amygdalus)</i>	AI, Spain
ROOTPAC [®] 70	<i>(P. persica × P. amygdalus) × (P. persica × P. davidiana)</i>	AI, Spain
Tetra	<i>P. domestica</i>	CRF, Italy

^a AI = Agromillora Iberia, S.L. private nursery, Spain; CITA = Centro de Investigación y Tecnología

Agroalimentaria de Aragón; CRF = Centro di Ricerca per la Frutticoltura; CSIC = Consejo Superior de

Investigaciones Científicas; INRA = Institut National de la Recherche Agronomique; KEBS = Krymsk

Experimental Breeding Station;

^b Formerly known as VVA-1

Table 2

Effect of rootstock on leaf chlorophyll concentration, measured as SPAD values, of ‘Calrico’ peach cultivar, the third (2005) and the seventh (2009) year after grafting.

Rootstock	2005	2009
Adesoto	39.1 bcde	41.0 e
Evrica	40.4 de	41.4 e
Garnem	38.4 bcd	37.1 bcd
GF 677	39.3 cde	39.8 de
HM-2	41.0 e	37.1 bcd
Krymsk [®] 1	37.0 ab	36.9 bc
PAC 9801-02	40.3 de	39.2 cde
PAC 9907-02	35.5 a	36.2 ab
PAC 9917-26	35.5 a	34.2 a
PAC-MUT	40.3 de	37.3 bcd
PADAC 9907-23	37.9 bc	37.0 bcd
ROOTPAC [®] 40	N/A	37.3 bcd
ROOTPAC [®] 70	38.3 bcd	37.8 bcd
Tetra	40.3 de	38.0 bcd

Means within a column followed by the same letter are not significantly different at $P \leq 0.05$ by Duncan's

Multiple Range Test.

N/A: not available.

374 **Table 3**

375 Effect of rootstock on TCSA (trunk cross-sectional area), cumulative yield and yield
 376 efficiency of ‘Calrico’ peach cultivar in the seventh (2009) year after grafting.

Rootstock	TCSA (cm ²)	Cumulative yield (kg tree ⁻¹)	Yield efficiency (kg cm ⁻²)
Adesoto	127.4 cd	149.1 cd	1.18 def
Evrica	91.1 b	124.9 bc	1.39 fgh
Garnem	239.6 h	155.4 cd	0.66 a
GF 677	167.0 ef	166.1 de	0.98 bcd
HM-2	207.5 g	191.4 e	0.93 bc
Krymsk [®] 1	57.8 a	82.4 a	1.44 gh
PAC 9801-02	76.5 ab	123.8 bc	1.62 h
PAC 9907-02	179.9 fg	129.0 bc	0.73 ab
PAC 9917-26	186.7 fg	155.4 cd	0.83 abc
PAC-MUT	144.1 de	100.6 ab	0.76 ab
PADAC 9907-23	239.0 h	175.3 de	0.74 ab
ROOTPAC [®] 40	103.2 bc	149.6 cd	1.46 gh
ROOTPAC [®] 70	157.1 def	196.9 e	1.27 efg
Tetra	174.2 ef	183.1 de	1.06 cde

377 Means within a column followed by the same letter are not significantly different at $P \leq 0.05$ by Duncan’s

378 Multiple Range Test.

Table 4

Effect of rootstock on yield (kg) of ‘Calrico’ peach cultivar, from the third (2005) to the seventh (2009) year after grafting.

Rootstock	2005	2006	2007	2008	2009	Average 2006-2009
Adesoto	8.6 a	27.1 abc	27.5 de	39.8 cde	51.9 cd	36.6 cd
Evrica	7.4 a	27.1 abc	24.7 bcde	29.1 abc	40.9 bc	30.4 bc
Garnem	7.8 a	41.6 de	15.7 ab	29.4 abcd	65.5 de	38.1 cde
GF 677	12.7 a	39.8 cde	18.2 abc	36.7 bcde	65.2 de	39.9 def
HM-2	11.2 a	48.1 e	23.0 bcde	43.5 e	69.3 e	46.0 ef
Krymsk [®] 1	8.5 a	14.8 a	17.4 abc	20.7 a	25.2 a	19.6 a
PAC 9801-02	7.3 a	27.5 abc	24.6 bcde	31.8 abcd	36.2 ab	30.0 bc
PAC 9907-02	6.4 a	23.6 ab	13.4 a	34.2 bcde	54.6 cde	31.4 bc
PAC 9917-26	8.3 a	38.5 cde	13.5 a	30.6 abcd	70.0 e	38.2 cde
PAC-MUT	N/A	15.1 a	10.9 a	27.7 ab	47.0 bc	25.2 ab
PADAC 9907-23	12.9 a	44.5 e	19.5 abcd	39.6 cde	64.4 de	42.0 def
ROOTPAC [®] 40	4.8 a	29.9 bcd	25.9 cde	37.7 bcde	54.0 cde	36.9 cd
ROOTPAC [®] 70	20.4 a	50.0 e	29.0 e	40.5 cde	67.2 de	46.7 f
Tetra	16.6 a	42.0 de	27.4 de	41.0 de	64.5 de	43.7 def

Means within a column followed by the same letter are not significantly different at $P \leq 0.05$ by Duncan's

Multiple Range Test.

N/A: not available.

Table 5

Effect of rootstock on fruit weight (g) of ‘Calrico’ peach cultivar, from the third (2005) to the seventh (2009) year after grafting.

Rootstock	2005	2006	2007	2008	2009	Average 2006-2009
Adesoto	269.5 bc	254.0 bc	289.0 cde	245.8 abc	241.7 a	257.6 bcdef
Evrica	265.3 abc	275.0 cd	271.0 bcd	256.7 bcd	237.0 a	259.9 bcdef
Garnem	241.5 ab	272.6 cd	264.6 bcd	283.8 def	272.6 a	273.4 defgh
GF 677	236.0 ab	245.2 bc	256.7 bc	240.7 ab	246.7 a	247.3 abc
HM-2	243.5 ab	236.0 b	244.0 ab	240.7 ab	243.7 a	241.1 ab
Krymsk [®] 1	263.0 abc	293.8 d	315.0 e	317.2 f	257.8 a	296.0 h
PAC 9801-02	294.0 c	289.4 d	292.4 de	263.6 bcde	236.5 a	270.5 cdefg
PAC 9907-02	223.0 a	209.3 a	225.5 a	232.5 ab	239.3 a	226.6 a
PAC 9917-26	260.0 abc	252.2 bc	261.2 bcd	256.7 bcd	267.0 a	259.3 bcdef
PAC-MUT	N/A	282.7 cb	262.8 bcd	227.2 a	244.2 a	254.2 bcde
PADAC 9907-23	245.3 ab	249.9 bc	247.4 ab	255.0 bcd	248.0 a	250.1 bcd
ROOTPAC [®] 40	360.7 d	298.1 d	282.1 cde	276.0 cdef	266.6 a	280.7 fgh
ROOTPAC [®] 70	255.7 abc	268.2 cd	269.8 bcd	310.0 ef	256.3 a	276.1 efgh
Tetra	244.0 ab	264.3 bcd	291.0 de	312.0 f	278.6 a	286.5 gh

Means within a column followed by the same letter are not significantly different at $P \leq 0.05$ by Duncan’s

Multiple Range Test

N/A: not available.