



# Adventitious rooting of cultivars and clonal selections of *Prunus* spp. rootstock under intermittent mist system<sup>1</sup>

Newton Alex Mayer<sup>2\*</sup>, Jéssica Luz Lopes<sup>3</sup>, Bernardo Ueno<sup>2</sup>

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## ABSTRACT

To reduce peach tree losses due the Peach Tree Short Life (PTSL), the main research line in Brazil is to select tolerant rootstock and perpetuate it by cloning methods. The objective was to evaluate adventitious rooting capacity of softwood cuttings of three cultivars and several clonal rootstock selections of *Prunus* spp., under intermittent mist system. Two trials were conducted and ‘Capdeboscq’, ‘Okinawa’ (*P. persica*) and ‘Sharpe’ [‘Chickasaw’ (*P. angustifolia* Marsh.) x *Prunus* spp.] were used as reference cultivars. After 53 days (trial 1) and 72 days (trial 2) from the cutting setting, we conclude that softwood cuttings under intermittent mist system are a feasible way to propagate peach rootstock selections of 2010/2011 PTSL selection cycle. Root quality was satisfactory in most selections, with high percentages of cuttings suitable for transplanting and excellent root number and root length. OS-GRA-10-17, VEH-GRA-10-31, VEH-GRA-10-32, VEH-GRA-10-37 and VEH-GRA-10-38 selections had the highest mortality and the lower percentages of live rooted cuttings. The ease of vegetative propagation must be taken into account in more promising rootstock.

**Keywords:** Rosaceae; softwood cutting; cloning; PTSL syndrome.

## INTRODUCTION

Among main agronomic problems that affect peach in Rio Grande do Sul State, Brazil, there are the absence of clonal rootstocks adapted to the edaphoclimatic conditions of the state’s peach area and which present tolerance to the Peach Tree Short Life (PTSL). This syndrome has affected commercial peach and plum orchards, especially in Pelotas area since late 1970s, which greatly reduces orchard life (Mayer *et al.*, 2009; Mayer & Ueno, 2012; Ueno *et al.*, 2019; Mayer & Ueno, 2021).

The syndrome is characterized by a collapse of peach tree scion at winter, with decreasing or standstill growth, weak and/or uneven sprouts, late or nonexistent flowering, and shoot death, parts or whole scion death, but the rootstock usually remains alive (Mayer *et al.*, 2009; Mayer & Ueno, 2012; Campos *et al.*, 2014; Ueno *et al.*, 2019; Alba *et al.*, 2019). In Southeast of the United States

(South Carolina, North Carolina and Georgia), the Peach Tree Short Life (PTSL) manifests characteristics and symptoms very similar to those observed in South Brazil (Brittain & Miller, 1978; Okie *et al.*, 1994; Beckman *et al.*, 2008; Beckman *et al.*, 2012; Mayer & Ueno, 2012; Campos *et al.*, 2014; Ueno *et al.*, 2019). With the technological advances obtained through of breeding programs for rootstocks, there was a significant reduction in tree losses caused by the PTSL syndrome in the United States, which culminated in the releasing of tolerant rootstock ‘Guardian<sup>®</sup>’, ‘Sharpe’ and ‘MP-29<sup>®</sup>’ (Okie *et al.*, 1994; Okie & Nyczepir, 2004; Beckman *et al.*, 2008; Beckman *et al.*, 2012). ‘Sharpe’ is in the public domain and was introduced in Brazil by Embrapa Clima Temperado (Mayer & Ueno, 2015).

In Brazil, the PTSL syndrome is restricted to the peach areas of Rio Grande do Sul (Mayer & Ueno, 2012; Campos

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<sup>2</sup> Embrapa Clima Temperado, BR 392, km 78, Pelotas, Rio Grande do Sul, Brazil. alex.mayer@embrapa.br; bernardo.ueno@embrapa.br

<sup>3</sup> Instituto Federal de Educação, Ciência e Tecnologia Sul-riograndense, Campus Visconde da Graça, Pelotas, Rio Grande do Sul, Brazil. jessicalopesgestaoambiental@gmail.com

\*Corresponding author: alex.mayer@embrapa.br

*et al.*, 2014; Ueno *et al.*, 2019; Mayer & Ueno, 2021). The rootstock used, in most of the peach and plum orchards in the State, are formed from seed germination and cause genetic variability, usually coming from canning industry (pits as residue of peach processing). This rootstock propagation method produces trees with genetically heterogeneous root systems, does not allow preservation of original rootstock's genetic identity, prevents varietal identification and manifests differences in vigor, yield, fruit quality, different reactions to biotic and abiotic factors and also to the PTSL syndrome (Mayer *et al.*, 2009; Mayer & Ueno, 2012; Mayer *et al.*, 2014a; Menegatti *et al.*, 2019; Mayer & Ueno, 2021). If the genetic variability of rootstock is undesirable in commercial orchards, on the other hand it is essential in a rootstock selection at field with PTSL history (Mayer *et al.*, 2009).

In view of this situation, researchers of the Embrapa Clima Temperado (Pelotas, Brazil) started a peach rootstock selection focusing in genotypes tolerant to the PTSL. After seven cycles of selection and cloning, 146 genotypes were rescued, which are maintaining in the "Prunus rootstock collection" (Mayer & Ueno, 2021). Among the desirable characteristics of a good rootstock, in addition to scion graft compatibility and tolerance to biotic and/or abiotic soil problems, vegetative propagation is one of main selection criteria (Okie & Nyczepir, 2004; Beckman *et al.*, 2008; Reighard & Loreti, 2008; Beckman *et al.*, 2012; Warschefsky *et al.*, 2016; Oliveira *et al.*, 2018). Therefore, in order to advance in rootstock selection process, it is necessary to know the propagation potential of selected clones, comparing them with reference cultivars.

As many species within the genus *Prunus* are difficult to root (Anderson *et al.*, 2016) and rooting of softwood cuttings is influenced by genetics (Hartmann *et al.*, 2002; Neêas & Krška, 2013; Mayer *et al.*, 2014b; Mayer *et al.*, 2015a; Anderson *et al.*, 2016; Mayer *et al.*, 2020), our hypothesis is that rootstock selections have a different potential for adventitious rooting of cuttings. The objective was to evaluate adventitious root potential in softwood cuttings from three cultivars and several clonal selections of *Prunus* spp. under intermittent mist system.

## MATERIAL AND METHODS

Mother trees of rootstock selections were originally selected in several commercial peach orchards in the Rio Grande do Sul (Brazil) that contained asymptomatic trees neighboring trees with typical PTSL symptoms in 2010 (Mayer *et al.*, 2009). With the scion cut methodology below the grafting point carried out between July and September, new and exclusive shoots of each rootstock was stimulated, which were collected at beginning of the following year for cloning by softwood cuttings,

completing the biannual selection and rescue cycle (Mayer *et al.*, 2009). Each selected clone received a code, including initial letters of peach grower's name where the selection was made; peach scion cultivar; selection year; and clone number. Trees were established in "Prunus rootstock collection", at Embrapa Clima Temperado (Pelotas, Rio Grande do Sul, Brazil) to constitute mother trees (Mayer & Ueno, 2021).

Were used softwood shoots collected in 7-years-old mother trees from 30 clonal selections belonging to 2010/2011 selection cycle. For operational and infrastructure reasons, two trials (named as 1 and 2) were carried out, each one containing 15 selections. Three cultivars were also used as reference: a) 'Capdeboscq' (*P. persica*), scion peach cultivar released for canning. Its seeds were also widely used as a rootstock until the 1980s, in Southern Brazil, due to ease of obtaining a reasonable seed germination and good adaptation to edaphoclimatic conditions (Mayer *et al.*, 2014a); b) 'Okinawa' (*P. persica*), a rootstock cultivar most used in Southeast Brazil for stone fruit, being normally propagated by seeds obtained from mother trees (Mayer *et al.*, 2014a); c) 'Sharpe' [*P. angustifolia* Marsh. x *Prunus* spp.], rootstock cultivar resistant to *Meloidogyne incognita* and *M. floridensis*; tolerant to *Armillaria tabescens* and PTSL. It is easily propagated by softwood or hardwood cuttings (Beckman *et al.*, 2008; Mayer & Ueno, 2015).

Between August 12<sup>th</sup> and 14<sup>th</sup>, 2018, mother trees were drastically pruned, in order to stimulate vigorous vegetative growth (Figure 1a.). After 129 days of pruning, we proceeded to collect softwood shoots (Figure 1b.) for trial 1, transferring them quickly to intermittent mist system to avoid dehydration. Softwood cuttings were 12cm-long, with distal apical bevel and basal transversal cuts, diameter between 8 and 10 mm, being left two or three distal nodes with all half-leaves to reduce transpiring rate (Figure 1c.). The cutting base was immersed in a hydroalcoholic solution at 50% (hydrated ethyl alcohol 96° GL + distilled water) of 4, 3 - indolylbutyric acid - IBA (C<sub>12</sub>H<sub>13</sub>NO<sub>2</sub>, purity >98%, Vetec®) at 3,000mg.L<sup>-1</sup> by fast immersion of 5 seconds (Figure 1d.) (Hartmann *et al.*, 2002; Mayer *et al.*, 2018; Mayer *et al.*, 2020). Trial 2 was set up on 13<sup>th</sup> and 14<sup>th</sup>, January/2019, with same methodology adopted in trial 1.

Cuttings were placed in perforated plastic boxes (46 x 30 x 10 cm) containing a fine vermiculite and perlite mixture (1:1, v/v). These containers were kept on 1m-high iron benches under intermittent misting system located inside an arch-type greenhouse (24 x 8m, with 3 m high), without air temperature nor air relative humidity control. Mist system was composed of a nozzle's line (Tietze® Violeta-type, equidistant 1 m each other), placed at 70 cm above containers. A 50% reflective screen (Lumineti®) was placed 1.2 m above the mist system, to reduce sun radiation.

Intermittent mist system was programmed by a timer to be activated for 10 seconds each 5 minutes, in the first 20 days of each trial, which allowed maintenance of constant moist on leaf surface (Ruter, 2015; Mayer *et al.*, 2018; Mayer *et al.*, 2020). In the following days, off-period was gradually increased at night until 45<sup>th</sup> and, after that, completely turned-off at night.

Treatments consisted of 15 clonal rootstocks, selected as tolerant to the PTSL, besides three reference cultivars (total of 18 treatments each trial). The trials were setting according to a completely randomized design, with four replications of 10 cuttings per plot. Air temperature (°C) and air relative humidity (%) were recorded every 30 min with a Datalogger (AK172-V2, AKSO), kept in shade 70 cm above the misting nozzles line. Data were summarized in Table 1.

After 53 and 72 days of trial 1 and 2 setting, respectively, the following variables were evaluated: 1) percentage of cuttings with original leaves; 2) percentage of sprouted cuttings. Cuttings were carefully removed from the rooting substrate, immersed in water for cleaning, and were evaluated: 3) percentage of callus cutting; 4) percentage of dead cuttings; 5) percentage of dead rooted

cuttings; 6) percentage of live rooted cuttings. Then, the live rooted cuttings were classified into: 7) percentage of rooted cuttings suitable for transplanting, that is, those with a minimum of four roots per cutting, distributed around the cutting base; 8) percentage of rooted cuttings unsuitable for transplanting, those with three or less roots, located unilaterally at the cutting base (Mayer *et al.*, 2014b). Cuttings classified as live rooted, the following were also evaluated: 9) root number per cutting; 10) root length (cm) of the three largest roots per cutting, using a ruler. Those variables expressed as percentage were transformed to  $\arcsin \sqrt{x/100}$ , and remain variables have not been transformed. Data were subjected to ANOVA by F-test and means compared by Scott-Knott test (Canteri *et al.*, 2001).

## RESULTS

Data of callused cutting percentage were not presented, due to low values obtained. In trial 1, of the fifteen selections tested, only four presented callus, with 5.00% (VHS-SEN-10-07 and OS-JAD-10-13) and 2.50% (VHS-SEN-10-10, OS-GRA-10-20), in addition to 'Capdeboscq' and 'Sharpe' (2.50%). In trial 2, callus were



**Figure 1:** A) Mother trees of peach rootstock selections drastically pruned in August; B) adequate softwood shoots for cutting preparation; C) softwood cuttings ready for IBA treatment; D) IBA treatment by fast immersion. Photos: Newton Alex Mayer.



observed only in JTV-GRA-10-46 (7.50%), JTV-JAD-10-41 (5.00%), VEH-GRA-10-39, JTV-GRA-10-47, ISD-GRA-10-53, VS-ELD-10-59 and 'Capdeboscq' (2.50%). Significant statistical differences were found in eight variables in trial 1 (Tables 2 and 3) and only three variables in trial 2 (Tables 4 and 5).

Three variables (percentages of dead cuttings, of dead rooted cuttings and of rooted cuttings unsuitable for transplanting) of the nine evaluated variables in each trial are undesirable, as they reduce propagation potential (yield) of a selection or cultivar. In trial 1, most selections

(eight) had a high percentage of dead cuttings (between 35.00 and 52.50%), as well as the most (eleven) had high percentages of dead rooted cuttings (between 15.0 and 32.5%), so that total mortality (dead cuttings + dead rooted cuttings) reached 82.50% in two selections (VEH-GRA-10-31 and VEH-GRA-10-32). The three reference cultivars are found in groups with the lowest percentages for these two variables, so that percentages of total mortality were low, with 2.50% ('Capdeboscq'), 22.50% ('Okinawa') and 17.50% ('Sharpe'). Among selections, as positive highlights, VHS-SEN-10-07 and VHS-SEN-10-10 had the

**Table 1:** Summary of environmental data recorded inside greenhouse during trial 1 (from Dec/07/2018 to Jan/28/2019) and trial 2 (from Jan/08/2019 to Mar/19/2019)

Environmental data	Trial 1	Trial 2
Maximum air temperature (°C)	44.8	43.9
Minimum air temperature (°C)	9.9	12.2
Air temperatures > 27.0 °C (% of total time)	35.7	34.4
Air temperatures < 15.0 °C (% of total time)	1.9	1.1
Maximum thermal amplitude per 24 h (°C)	27.2	24.1
Minimum thermal amplitude per 24 h (°C)	4.5	7.0
Maximum air relative humidity (%)	100.0	100.0
Minimum air relative humidity (%)	24.7	27.1
Maximum amplitude per 24 h of air relative humidity (%)	75.3	72.9
Minimum amplitude per 24 h of air relative humidity (%)	0.0	0.0

**Table 2:** Rooting response of softwood cuttings from cultivars and clonal selections of *Prunus* spp. rootstocks under intermittent mist system (trial 1)

Rootstock	Dead cuttings	Dead rooted cuttings	Live rooted cuttings	Suitable rooted cuttings	Unsuitable rooted cuttings
	(%)				
VHS-SEN-10-07	5.00 b	10.00 b	77.50 a	44.38 a	55.63 b
VHS-SEN-10-08	35.00 a	27.50 a	37.50 b	60.00 a	40.00 b
VHS-SEN-10-09	47.50 a	5.00 b	47.50 b	56.67 a	43.33 b
VHS-SEN-10-10	17.50 b	5.00 b	77.00 a	47.92 a	52.08 b
OS-JAD-10-12	40.00 a	20.00 a	40.00 b	34.38 a	40.63 b
OS-JAD-10-13	20.00 b	25.00 a	50.00 b	48.75 a	51.23 b
OS-GRA-10-16	12.50 b	32.50 a	55.00 b	62.38 a	37.62 b
OS-GRA-10-17	52.50 a	25.00 a	22.50 c	66.67 a	33.33 b
OS-GRA-10-18	47.50 a	5.00 b	47.50 b	7.14 a	92.86 a
OS-GRA-10-20	35.00 a	32.50 a	30.00 c	77.50 a	22.50 b
EM-PRE-10-21	25.00 b	20.00 a	55.00 b	46.23 a	53.77 b
EM-SEN-10-25	10.00 b	15.00 a	75.00 a	53.18 a	46.83 b
ENP-JAD-10-27	22.50 b	27.50 a	50.00 b	49.11 a	50.89 b
VEH-GRA-10-31	50.00 a	32.50 a	17.50 c	35.00 a	40.00 b
VEH-GRA-10-32	50.00 a	32.50 a	17.50 c	8.33 a	91.67 a
Capdeboscq	2.50 b	0.00 b	90.00 a	85.42 a	14.58 b
Okinawa	17.50 b	5.00 b	77.50 a	81.25 a	18.75 b
Sharpe	12.50 b	5.00 b	80.00 a	69.45 a	30.56 b
F <sub>rootstock</sub>	4.61**	4.17**	5.87**	2.14*	2.07*
CV (%)	42.34	55.42	27.15	52.56	57.67

Means followed by different letters in the column differ from each other by the Scott-knott test. \* Significant at 5% error probability; \*\* significant at 1% error probability.

lowest percentages of total mortality (15.0% and 22.5%, respectively), which were also found in the group with lower percentages of rooted cuttings unsuitable to transplant. In trial 2, there were no statistically significant differences between treatments for percentages of dead cuttings and for dead rooted cuttings. Four selections had percentages of total mortality  $\geq 70.0\%$  (VEH-GRA-10-37, VEH-GRA-10-38, JTV-JAD-10-42 and JTV-ESM-10-50), in addition to 'Okinawa' and 'Sharpe'.

Three selections stood out positively in percentage of live rooted cuttings in trial 1 (VHS-SEN-10-07, VHS-SEN-10-10 and EM-SEN-10-25), with values equal to or greater than 75.0% (Table 2), statistically equal to three reference cultivars. Another ten selections stood out in trial 2, with percentages of live rooted cuttings greater than 37.5%, and statistically higher in relation to three reference cultivars (Table 4). All 13 selections with the highest percentages of live rooted cuttings from both trials are also found in group of those with the highest percentages of rooted cuttings suitable to transplant (Tables 2 and 4).

For root number and root length, statistical differences were detected only in trial 1 (Table 3). Although with these significant differences, all treatments showed, in general, very satisfactory results, in both trials. In trial 1 (Table 3), root number per cutting varied between 5.43 (OS-GRA-

10-18) and 25.77 ('Sharpe'), with a length between 2.72 cm (OS-GRA-10-18) and 8.80cm (VHS-SEN-10-09); in trial 2 (Table 5), root number per cutting ranged between 4.00 (ISD-GRA-10-53) and 18.84 ('Sharpe'), while root length ranged between 1.83cm (JTV-ESM-10-50) and 8.12 cm (JTV-GRA-10-46).

For the percentages of cuttings with original leaves and sprouted cuttings in trial 1, there were statistical differences among treatments for both variables. For the first variable, VHS-SEN-10-07 (72.0%), VHS-SEN-10-10 (67.5%) and EM-SEN-10-25 (62.5%) stood out, which were statistically equal to 'Capdeboscq' (92.5%) and 'Sharpe' (62.5%). The three reference cultivars had the highest percentage of sprouted cuttings, along with six other selections. However, considering both original leaves and the sprouting beginning, only a single selection (VHS-SEN-10-07) stood out in trial 1, besides 'Capdeboscq' and 'Sharpe'. In trial 2, differences were only sprouted cuttings, with the majority (ten selections) being in group with the highest percentages, besides three cultivars.

## DISCUSSION

Cuttings that died early without any adventitious roots (percentage of dead cuttings), were, in general, considered high, in both trials (Tables 2 and 4). Main factor responsible for this cutting mortality is early original leaf fall. Softwood

**Table 3:** Root quality, leaf persistence and sprouting of softwood cuttings from cultivars and clonal selections of *Prunus* spp. rootstocks under intermittent mist system (trial 1)

Rootstock	Root number per cutting	Root length (cm)	Cuttings with original leaves (%)	Sprouted cuttings (%)
VHS-SEN-10-07	8.92 c	8.03 a	72.00 a	32.50 a
VHS-SEN-10-08	8.78 c	5.56 b	35.00 b	7.50 b
VHS-SEN-10-09	10.75 c	8.80 a	35.00 b	37.50 a
VHS-SEN-10-10	9.52 c	6.13 a	67.50 a	7.50 b
OS-JAD-10-12	5.52 c	3.42 b	22.50 b	17.50 b
OS-JAD-10-13	9.40 c	4.67 b	47.50 b	0.00 b
OS-GRA-10-16	15.90 b	6.15 a	30.00 b	35.00 a
OS-GRA-10-17	9.79 c	3.81 b	25.00 b	37.50 a
OS-GRA-10-18	5.43 c	2.72 b	45.00 b	2.50 b
OS-GRA-10-20	13.45 c	4.65 b	12.50 b	5.00 b
EM-PRE-10-21	11.59 c	7.57 a	40.00 b	35.00 a
EM-SEN-10-25	12.96 c	6.75 a	62.50 a	10.00 b
ENP-JAD-10-27	11.86 c	6.35 a	30.00 b	35.00 a
VEH-GRA-10-31	6.00 c	3.99 b	17.50 b	0.00 b
VEH-GRA-10-32	6.67 c	4.74 b	12.50 b	5.00 b
Capdeboscq	18.80 b	8.57 a	92.50 a	25.00 a
Okinawa	21.64 a	4.86 b	37.50 b	57.50 a
Sharpe	25.77 a	6.94 a	62.50 a	65.00 a
F <sub>rootstock</sub>	6.13*	3.52**	4.80**	5.76**
CV (%)	38.36	33.15	37.56	58.83

Means followed by different letters in the column differ from each other by the Scott-knott test. \* Significant at 5% error probability; \*\* significant at 1% error probability.

cuttings without their original leaves do not perform photosynthesis and do not produce cofactors essential to rooting and, with the high summer air temperatures and without having started the rhizogenesis, they die early (Machida *et al.*, 1977; Hartmann *et al.*, 2002; Thomas & Schiefelbein, 2004; Santoro *et al.*, 2010; Tombesi *et al.* 2015; Sanchez *et al.*, 2020). Leaves are essential for softwood cuttings of peach and several other species (Hartmann *et al.*, 2002; Thomas & Schiefelbein, 2004; Santoro *et al.*, 2010; Mayer *et al.*, 2014b; Mayer *et al.*, 2015a; Tombesi *et al.* 2015; Mayer *et al.*, 2018). There are several factors that can contribute to the early original leaf fall on cuttings under intermittent mist system, such as the deficient nutrition of mother tree, inappropriate shoots for cutting preparation (date, type, age, size, diameter, health, etc...), endogenous hormonal balance favorable to leaf senescence, genetics, leaf and petiole morphology, water stress (from the air and/or substrate), inadequate regulation of mist system, thermal stress (especially air temperature differences between day and night), fungal diseases, and inappropriate containers and/or substrates (Hartmann *et al.*, 2002; Osterc *et al.*, 2009; Osterc & Štampar, 2011; Neèas & Krška, 2013; Ruter, 2015; Fragoso *et al.*, 2015; Mayer *et al.*, 2015a; Mayer *et al.*, 2020). Some of these factors usually occur simultaneously,

to a greater or lesser degree, and, in most cases, it is difficult to identify a single factor as the causal agent of early original leaf fall. In the present research, cuttings were not previously disinfected with fungicides. Fluctuations in air temperature and air relative humidity in both trials (Table 1), especially between day and night, may also have contributed to leaf water stress and for the substrate.

The percentage of dead rooted cuttings is another undesirable variable and reveals that adventitious root formation process has started, however, normally due to the water excess in substrate, there is no proper aeration at cutting base, which end up dying. In this respect, the environmental differences between day and night (especially air temperature and air relative humidity) are factors that vary significantly (Table 1). So, that is difficult to adjust mist schedule to maintain leaves adequately moistened during the day (with temperatures greater than 40°C), without causing the substrate to soak during the night, when temperatures fall around 15°C. Additionally, fine vermiculite + perlite mixture (1:1) used as a substrate, combined with plastic boxes characteristics, probably were not sufficient for perfect drainage, and had a similar performance to pure fine vermiculite (Mayer *et al.*, 2020).

According to Mattson & Fulcher (2015), the ability of a substrate to retain water and air is dependent on particles

**Table 4:** Rooting response of softwood cuttings from cultivars and clonal selections of *Prunus* spp. rootstocks under intermittent mist system (trial 2)

Rootstock	Dead cuttings	Dead rooted cuttings	Live rooted cuttings	Suitable rooted cuttings	Unsuitable rooted cuttings
	(%)				
VEH-GRA-10-33	25.00 a	27.50 a	47.50 a	23.81 a	76.19 a
VEH-GRA-10-36	25.00 a	17.50 a	57.50 a	36.61 a	63.39 a
VEH-GRA-10-37	47.50 a	27.50 a	25.00 b	25.00 a	75.00 a
VEH-GRA-10-38	55.00 a	25.00 a	20.00 b	39.58 a	60.42 a
VEH-GRA-10-39	25.00 a	12.50 a	62.50 a	28.47 a	71.53 a
JTV-JAD-10-41	32.50 a	32.50 a	25.00 b	50.00 a	50.00 a
JTV-JAD-10-42	47.50 a	22.50 a	30.00 b	27.08 a	72.92 a
JTV-JAD-10-43	42.50 a	12.50 a	45.00 a	41.67 a	58.33 a
JTV-JAD-10-44	27.50 a	27.50 a	45.00 a	23.75 a	76.25 a
JTV-GRA-10-46	20.00 a	17.50 a	55.00 a	43.16 a	56.85 a
JTV-GRA-10-47	17.50 a	25.00 a	57.50 a	25.83 a	74.17 a
JTV-ESM-10-50	32.50 a	37.50 a	30.00 b	50.00 a	25.00 b
ISD-GRA-10-53	35.00 a	17.50 a	45.00 a	15.00 a	85.00 a
ISD-ELD-10-55	20.00 a	27.50 a	55.00 a	58.45 a	41.55 a
VS-ELD-10-59	42.50 a	17.50 a	37.50 a	16.67 a	83.34 a
Capdeboscq	32.50 a	32.50 a	35.00 b	42.14 a	57.86 a
Okinawa	52.50 a	25.00 a	7.50 b	50.00 a	0.00 b
Sharpe	5.00 a	65.00 a	30.00 b	66.67 a	8.33 b
F <sub>rootstock</sub>	1.83*	1.51 <sup>NS</sup>	2.31*	0.72 <sup>NS</sup>	3.34**
CV (%)	45.57	51.12	38.54	79.21	45.85

Means followed by different letters in the column differ from each other by the Scott-knott test. <sup>NS</sup> Not significant; \* significant at 5% error probability; \*\* significant at 1% error probability.

size. A finer textured substrate, that is, with smaller particles, tends to retain more water in relation to a coarse textured substrate, increasing saturation zone at container bottom and reaching cuttings base. Therefore, alternative substrates should be studied, with a thicker texture and capable of allowing better aeration at the cutting base, as a way of reducing cutting mortality after rhizogenesis.

The percentages of live rooted cuttings in both trials (Tables 2 and 4) were considered satisfactory. The percentage of live rooted cuttings is the key in cutting propagation trials (Hartmann *et al.*, 2002; Mayer *et al.*, 2018; Mayer *et al.*, 2020). The highest values observed in 10 selections (trial 2, Table 4) and statistical equality of three selections (trial 1, Table 2), in relation to the reference cultivars, reveals that selected clones also present satisfactory rooting ability, besides potentially tolerant to the PTSL syndrome. According to the literature review made by Neèas & Krška (2013), a rooting yields of around 30 % can be considered as the limit for difficult to propagate genotypes and 50 % is the limit of successful propagation. The same authors mention that over 60 % success can be considered as economically effective propagation and over 75 % success is highly effective propagation. Thus, considering the 30 rootstock selections tested in the present research and 50 % of live rooted cuttings as a limit, seven and five selections

(trials 1 and 2, respectively) stood out, but VHS-SEN-10-07, VHS-SEN-10-10 and EM-SEN-10-25 can be sorted as highly effective propagation genotypes.

Another important variable is the percentage of rooted cuttings suitable for transplanting, because in addition to being indicative of root quality, it also translates the amount of usable rooted cuttings for the subsequent phase (transplanting and acclimatization). In both trials (Tables 2 and 4), all selections tested were statistically similar to the three reference cultivars, another important characteristic verified in selected clones. The quality of adventitious root system is also expressed by root number and root length. Although statistical differences were detected in both variables in trial 1, and statistical equality in trial 2, results of these variables, in general, were considered quite satisfactory. The number of primary roots that tree will have in the orchard is defined during propagation time under mist system (normally 60 days) (Mayer *et al.*, 2014b; Mayer *et al.*, 2018; Mayer *et al.*, 2020). Consequently, root number and root distribution around cutting will influence the performance of mature tree. With a vigorous and wide root system, there will be greater possibilities for tree adaptation to gravel soils and water deficit, common situations in peach area in Pelotas-RS where PTSL syndrome occurs (Mayer & Ueno, 2012; Mayer *et al.*, 2015b; Alba *et al.*, 2019; Mayer & Ueno, 2021).

**Table 5:** Root quality, leaf persistence and sprouting of softwood cuttings from cultivars and clonal selections of *Prunus* spp. rootstocks under intermittent mist system (trial 2)

Rootstock	Root number per cutting	Root lenght (cm)	Cuttings with original leaves (%)	Sprouted cuttings (%)
VEH-GRA-10-33	13.54 a	6.31 a	25.00 a	57.50 a
VEH-GRA-10-36	9.62 a	6.99 a	45.00 a	30.00 b
VEH-GRA-10-37	7.44 a	5.65 a	5.00 a	40.00 a
VEH-GRA-10-38	5.92 a	3.05 a	10.00 a	5.00 c
VEH-GRA-10-39	11.12 a	7.73 a	45.00 a	45.00 a
JTV-JAD-10-41	17.98 a	6.97 a	15.00 a	50.00 a
JTV-JAD-10-42	7.42 a	5.85 a	25.00 a	27.50 b
JTV-JAD-10-43	9.90 a	6.63 a	27.50 a	42.50 a
JTV-JAD-10-44	15.63 a	5.98 a	22.50 a	47.50 a
JTV-GRA-10-46	6.82 a	8.12 a	50.00 a	52.50 a
JTV-GRA-10-47	8.50 a	5.13 a	37.50 a	52.50 a
JTV-ESM-10-50	7.25 a	1.83 a	27.50 a	22.50 b
ISD-GRA-10-53	4.00 a	5.29 a	30.00 a	75.00 a
ISD-ELD-10-55	16.91 a	7.67 a	42.50 a	55.00 a
VS-ELD-10-59	8.54 a	7.54 a	37.50 a	20.00 b
Capdeboscq	6.45 a	6.90 a	32.50 a	37.50 a
Okinawa	7.75 a	3.45 a	15.00 a	50.00 a
Sharpe	18.84 a	7.88 a	40.00 a	42.50 a
F <sub>rootstock</sub>	1.36 <sup>NS</sup>	1.68 <sup>NS</sup>	1.77 <sup>NS</sup>	9.92 <sup>**</sup>
CV (%)	75.09	45.17	47.14	30.79

Means followed by different letters in the column differ from each other by the Scott-knott test. <sup>NS</sup> Not significant; <sup>\*\*</sup> significant at 1% error probability.

As already discussed, permanence of original leaves to the cutting is essential for photosynthesis and production of cofactors for rhizogenesis in several species, including peach (Machida *et al.*, 1977; Hartmann *et al.*, 2002; Thomas & Schiefelbein, 2004; Santoro *et al.*, 2010; Mayer *et al.*, 2014b; Mayer *et al.*, 2015a; Tombesi *et al.* 2015; Mayer *et al.*, 2018; Sanchez *et al.*, 2020). According to results (Tables 3 and 5), most selections (twelve) in trial 1 and all selections in trial 2 presented percentages  $\leq 50.0\%$  of cutting with original leaves. However, this low data was compensated, at least in part, by the sprouting of cuttings under intermittent mist system (Tables 3 and 5). Only two selections (OS-JAD-10-13 and VEH-GRA-10-31) did not sprout during time under intermittent mist, however, due to the original leaf retention (47.5 and 17.5%, respectively), it was possible to obtain rooting (50.0 and 17.5%, respectively). The photosynthesis and phytohormones production is continuous, if cuttings retain their original leaves and/or new sprouts emerge. Growing tips, leaves and buds are sites indole-acetic acid synthesis, which contributes to adventitious root formation (Hartmann *et al.*, 2002; Fragoso *et al.*, 2015; Mayer *et al.*, 2018). Softwood cuttings of 'Lovell' or semi-hardwood cuttings of 'Guardian®' rootstocks (*P. persica*) did not sprout at rooting phase under a mist system, but still excellent rooting percentages ( $> 85\%$ ) can be obtained (Mayer *et al.*, 2015a). In peach rootstock selections, sprouting percentages of softwood cuttings were usually higher than percentages of cuttings with original leaves, so satisfactory rooting could be obtained (Mayer *et al.*, 2018; Mayer *et al.*, 2020).

Air temperature and its amplitude is another factor that affects rooting of cuttings (Hartmann *et al.*, 2002; Ruter, 2015). The ideal range temperature for rooting cuttings will depend on specie, cutting type, season and rooting facilities (Ruter, 2015). In general, air temperatures between  $15^{\circ}\text{C}$  and  $27^{\circ}\text{C}$  is an ideal range for temperate fruit species (Hartmann *et al.*, 2002). In both trials, it was observed that temperatures  $>27^{\circ}\text{C}$  occurred between 9:00 am and 6:30 pm, or that is, during practically the whole daytime. So, air temperatures  $>27^{\circ}\text{C}$  occurred during a significant part of total rooting time for both trials (Table 1), as well as the thermal amplitude in 24 h was too high, which may have caused stress on cuttings and reduced rooting percentages and root quality. Therefore, the environmental conditions registered in both trials were quite similar, suggesting need some changes in the facilities to reduce maximum air temperatures, in order to promote a longer period of thermal comfort.

## CONCLUSIONS

Softwood cuttings under intermittent mist system are a feasible way to propagate peach rootstock selections of

2010/2011 PTSL selection cycle. Root quality was satisfactory in most of studied selections, with high percentages of cuttings suitable for transplanting and excellent root number and root length. OS-GRA-10-17, VEH-GRA-10-31, VEH-GRA-10-32, VEH-GRA-10-37 and VEH-GRA-10-38 selections had the highest mortality rates and the lowest percentages of live rooted cuttings. Several selections have similar or superior propagation potential in relation to three reference cultivars. The ease of vegetative propagation and root quality must be taken into account, in the search for promising rootstock.

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