Post-harvest Management of *Prunus persica* Stones and the Effects on Seed and Seedling Quality

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Summary

The aim of this study was to evaluate the effect of two forms of post-harvest management of 'Capdeboscq' peach stones (Farmer-Management and UFPel-Management) on the quality of seeds and seedlings to be used as rootstock. The experimental design was completely randomised with four replications of 50 seeds. The variables to be analysed were fresh seed weight, seed moisture content, percentage of intact and fungus-infected seeds, germination percentage, germination speed index (GSI) and seedling height up to 24 days after planting (DAP). At 24 DAP, the stem diameter, Soil Plant Analysis Development (SPAD index), chlorophyll index, nitrogen balance index, shoot dry weight, root dry weight, total dry weight and the Dickson quality index (DQI) were evaluated. The seeds from traditional farmer management showed a high rate of infestation by phytopathogens (36%) and only 64% of intact seeds. The seeds from the Federal University of Pelotas (UFPel) management showed a rate of germination 1.63 times greater than those from the farmer management. It was concluded that proper post-harvest management of the stones, removing the pulp and treating the stones with fungicide, had a direct effect on maintaining the physiological quality of seed intended for the production of rootstock, making it possible to obtain increased vigour and higher initial seedling quality.

Key words

Rosaceae, Dickson quality index, peach tree, vigour, germination

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Introduction

In Brazil, commercial seedlings of stone-fruit species are generally obtained by grafting scions of interest onto ungrafted, seed-derived rootstock from any late-maturing peach scion, which are usually obtained from the canning industry (Fischer et al., 2016).

This propagation material is not suitable for use in the production of rootstock, due to a mixture of seedlings of different cultivars, there being no guarantee of genetic identity, which results in the formation of heterogeneous seedlings of low quality (Bianchi et al., 2014).

To produce quality seedlings, some nurseries have their own 'Capdeboscq' peach orchards, with the aim of using seeds of this cultivar for the production of rootstock. Among the main characteristics of the cultivar, most important is the late cycle, which allows for adequate embryo development (Bianchi et al., 2014), resulting in the formation of seeds of greater germination potential and seedling vigour (Souza et al., 2019; Souza et al., 2017; Souza et al., 2016).

On the other hand, there is a lack of specifically suitable methods for analysing the physiological quality of the seeds of stone fruit, as well as lack of definition concerning the post-harvest management of stones and seeds, making it difficult to predict the quality and success of using batches of *Prunus* spp. seed for rootstock production.

In the case of *Prunus*, all species of commercial interest have seeds with physiological and physical dormancy, the latter determined by the presence of a highly lignified endocarp (Fischer et al., 2016). Although overcoming this barrier is indispensable for obtaining a high percentage of germinated seeds (Souza et al., 2017), the success of germination also depends on the proper handling and processing of the stones (endocarp) and seeds after the fruit is harvested, in order to maintain not only physiological but also phytosanitary quality so that the seeds express maximum vigour.

In the southern region of Brazil, which, according to Mayer et al. (2014), is the main production area for seedlings of *Prunus persica*, the traditional method of producing rootstock for stone fruit still predominates, so the post-harvest processing of seeds consists in keeping the stones in the shade under the tree canopy for a period ranging from 75 to 150 days, so that pulp residue adhering to the stones rots and/or dries, and the physiological dormancy of the seeds is broken.

With this method of stratification, seed germination percentages of around 30% have been obtained in the field (Mayer and Antunes, 2010), resulting in low stands and low seedling production. This result is due in part to the variability of seed dormancy, the physical barrier determined by the endocarp, as well as a higher incidence of phytopathogens and seed rot caused by the phytosanitary conditions under which the stones are stratified.

The cultivation of stone fruit in Brazil, especially the peach crop, is socially and economically very attractive. However, there is still a lack of information and the need to introduce new techniques that might be of aid, not only in seed management but also in establishing morphophysiological standards for estimating the quality of the rootstock being produced.

Souza et al. (2015) suggest shoot height, stem diameter near the collar and the Dickson quality index as the main morphological parameters for determining seedling quality in peach rootstock.

In view of the above, the aim of this study was to evaluate the post-harvest management of stones of the Capdeboscq cultivar on the quality of seeds and seedlings to be used as rootstock.

Material and methods

Plant material used in the experiment

The plant material used were stones harvested during the first two weeks of January 2015 from ten-year-old clonal matrix plants of the Capdeboscq cultivar, grown on private property in the district of Pelotas, located at 31º32'03" S and 52º23' W at an altitude of 102m. To meet the aim of the study, two methods for post-harvest handling of the Capdeboscq cultivar were used.

Post-harvest management of the fruit stones

Traditional management adopted by the producer (Farmer-Management)

After harvesting, the fruit was taken to the local canning factory for the pulp to be removed. The stones discarded in the process were sent to the producer's property and arranged in a 5-6 cm layer and kept in the shade under the tree canopy for 75 days to allow the natural removal and/or drying of any pulp residue adhered to the stones. After this period, the stones were packed in raffia bags and stored in a shed for 45 days at average temperatures ranging from 7°C to 18°C.

Management adopted at UFPel (UFPel-Management)

Under the UFPel Management, after harvesting, the fruits were taken to the Laboratory of Plant Molecular Physiology, Department of Botany (UFPel), where the pulp was removed from the stones (endocarp), and washed in running water to remove any remaining pulp and then treated by immersion in a fungicide solution (Orthocid® 500-12 mg L^{-1}) for 12 hours. The stones were dried in the shade and stored in paper bags for four months as per Souza et al. (2016).

Processing the stones and seeds for germination

On 15 May 2015, after post-harvest management, the stones from both storage sites were processed by flaming with 70% alcohol for eight seconds; the endocarp was then broken on a manual lathe to remove the seeds.

A sample of 50 seeds from each batch or type of management (Farmer-Management or UFPel-Management) was used to determine the degree of moisture by means of the greenhouse method (105 ± 3°C) for 24 hours, with the results expressed as percentage moisture (Brazil, 2009). A morphometric evaluation and seed quality analysis were carried out when the following variables were evaluated: percentage of intact seeds, percentage of seeds with incidence of fungus, and fresh seed weight (g).

The experimental design was completely randomized, with two treatments (Farmer-Management and UFPel-Management) and four replications per treatment, each replication comprising 50 seeds.

After the morphometric evaluation, the seeds from both treatments were sterilised with sodium hypochlorite solution (1.5%) for 10 minutes, followed by triple-washing in distilled water. Twenty-five seeds were then placed in each Petri dish (90x15mm) containing a sheet of filter paper, which was moistened with 4 mL of fungicidal solution (Orthocid® 500-12 mg L^{-1}).

The Petri dishes were sealed with parafilm and taken to a Biochemical Oxygen Demand (BOD) chamber at 7 ± 0.5°C for stratification in the absence of light, where they remained for up to 23 days, the time required for at least one of the treatments to achieve 100% germination. During the cold moist stratification, the seeds were evaluated every two days in order to obtain the germination speed index (GSI), as proposed by Maguire (1962). At the end of the stratification period (23 days), the total percentage of germinated seeds was recorded for each treatment.

Initial growth and morphological characteristics of the seedlings

After 23 days of cold stratification, the seeds were planted in polystyrene trays (114 cm^3 per cell) containing a substrate composed of 25% soil + 25% vermiculite + 25% medium sand + 25% Bioplant® commercial substrate. The physical and chemical analysis of the substrate used for planting (Table 1) was carried out by the Plant Substrate Analysis Laboratory (LASPP) of the Federal University of Lavras.

At this stage of the experiment, the experimental design was completely randomised, consisting of two treatments (Farmer-Management and UFPel-Management) with five replications of five seedlings per plot. The seedlings were kept in a greenhouse at an average temperature ranging from 18°C to 36°C, night/day respectively, and manually irrigated as necessary, using three irrigations per week of the nutrient solution proposed by Souza et al. (2015).

Seedling height was evaluated using a millimetre rule after seedling emergence (2 cm above the substrate), and recorded at intervals of two days until 25 days after planting (DAP), by which time at least 75% of the seedlings were ready for transplanting (15 cm in height, from the collar to the apex), as recommended by Souza et al. (2011a).

At 24 DAP, the following parameters were determined: the diameter of the stem (mm) two cm above the level of the substrate; the SPAD index, by means of a SPAD-502 chlorophyll meter (Minolta, Osaka, Japan); the chlorophyll index (CHLI) and the nitrogen balance index (NBI), using a Dualex chlorophyll meter. The measurements were taken between 09:00 to 11:00 am, on two completely expanded leaves located on the top third of the plant, in 25 plants of each treatment.

Other variables to be evaluated were the root and shoot dry weight. After uprooting, the roots and shoots were separately packed in paper bags, remaining in a drying oven at 70°C for 72 hours. After drying, the samples were weighed on a 0.01 g precision balance to determine the shoot dry weight (SDW), root dry weight (RDW), and by summing these, the total dry weight of the plant (TDW). To measure the quality of the peach seedlings for use as rootstock, the Dickson quality index (Dickson et al., 1960) was used.

Statistical analysis of the data

All the data were tested for homogeneity of variance (Bartlett) and normality (Shapiro-Wilk). The mean values of the evaluated variables were then submitted to analysis of variance, and when significant, were compared by Tukey's test at 5% probability. A Pearson correlation analysis was performed between the variables GSI and plant height. Regression analysis was also performed for the variable seedling height, with the aim of establishing a growth curve over time. All statistical analyses were carried out with the aid of the R (2014) software.

Results and discussion

After four months post-harvesting, the mean moisture content of the 'Capdeboscq' seeds from the UFPel-Management was 10.9%, while from the Farmer-Management, the content was 16% (Table 2).

According to Marcos Filho (2005), recalcitrant seeds with a moisture content between 15% and 20% show greater respiratory activity in relation to seeds with a moisture content between 10% and 12%. This higher metabolic activity is insufficient to induce germination, but contributes to the rapid consumption of seed reserves and the greater activity of microorganisms, accelerating the loss of physiological quality in the seeds.

This corroborates the results seen in the seeds from the Farmer-Management, which displayed a high percentage (36%) of infestation by microorganisms (phytopathogens), and where only 64% were intact. The low quality of the Farmer-Management seeds contributes to a smaller economic return on the process of seedling production, due to the need for a larger quantity of seeds per metre of row or when sowing, to obtain a minimum number of seedlings per unit of cultivated area in the nursery.

Table 1. Physical and chemical analysis of the substrate used when planting to obtain seedlings of the Capdeboscq cultivar

Substrate	OM	V	Al	H+Al	SB	CTC	P	K	Ca	Mg	Zn	Fe	Mn
				$cmol_c\ dm^{-3}$				$mg\ dm^{-3}$				$mg\ dm^{-3}$	
SP*	2.12	78.00	0.01	1.00	1.83	1.78	1.89	7.80	0.42	0.10	1.62	5.12	3.12

SP = Substrate used when planting. SB: Sum of bases; T: Cation exchange capacity (CTC); V: Base saturation; OM: Organic matter.

Table 2. Mean values of quality seeds *P. persica* 'Capdeboscq' from two systems of post-harvest management

Management	Intact seeds (%)	Seeds with the incidence of fungus	Fresh seed weight	Moisture content after four months of storage	Germination of intact seeds (%)	Rate of germination
Farmer	64.0 b	36.0 a	0.54 a	16.0 b	70.0 b	7.7 b
UFPel	98.0 a	2.0 b	0.40 b	10.9 a	100.0 a	12.6 a
CV%	2.8	14.2	6.30	4.43	2.2	11.4

Mean values followed by the same lowercase letter in a column do not differ by Tukey's test ($p \leq 5\%$).

On the other hand, the seeds of the Capdeboscq cultivar from the UFPel-Management showed a high percentage of intact seeds (98%) (Table 3). This was attributed to proper post-harvest management, where processing consisted of the complete removal of the pulp from the stones immediately after harvest, followed by fungicide treatment and drying in the shade.

From the analyses and inferences made in this work, it was possible to demonstrate that the UFPel Management is effective in maintaining physiological quality and vigour in the seeds of *P. persica*, and can be recommended for use on a commercial scale, allowing more-vigorous seedlings to be established (Figure 1). In addition, the seeds had a germination speed 1.63 times greater (12.6/7.7) than those from the Farmer-Management (Table 2).

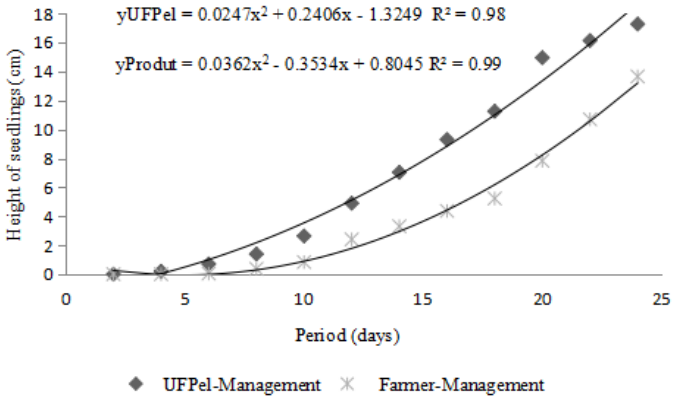

Figure 1. Growth in height of 'Capdeboscq' seedlings, from seeds submitted to UFPel-Management and to Farmer-Management, grown in a greenhouse up to 25 days after planting.

Initial seedling height showed a direct relationship with the GSI, with a Pearson correlation coefficient (r) of 0.97; the plant material from the UFPel-Management showed greater seedling vigour, and was ready for pricking out at 24 DAP under greenhouse conditions (Figure 1). This confirms that seeds that are properly packed post-harvest maintain their physical and physiological quality for longer, resulting in a high percentage of germination and emergence, and high seedling vigour.

The mean values for seedling height agree with those obtained by Souza et al. (2017), who worked with 'Capdeboscq' rootstock under the same conditions as the present study and achieved the pricking-out stage 25 days after planting. In turn, Picolotto et al. (2007), under experimental conditions similar to the present study, obtained seedlings of the Capdeboscq cultivar with a mean height of 15.0 cm and a diameter of 1.90 mm by 35 DAP. Schmitz et al. (2014), evaluating the growth of 'Capdeboscq' rootstock, obtained a mean height of 15.30 cm at 58 DAP.

In this study, the morphological variables and the Dickson quality index obtained with the 'Capdeboscq' seedlings from seeds of the UFPel-Management showed the greatest mean values at 25 DAP, when compared to the morphological characteristics of the seedlings from seeds of the Farmer-Management.

Such morphological characteristics are important for reaching the grafting stage quickly, serving as a differential parameter to ensure better yield and quality in the produced seedlings.

Determining the most suitable post-harvest management for seeds of *P. persica* is one of the factors that contribute to the rapid growth and development of the seedling (Souza et al., 2017). However, for good seedling performance, especially during the growth phase, the plant requires an adequate supply of nutrients, especially Nitrogen (N) (Smiderle et al., 2017).

Proper nutrient management and the use of tools which allow the nutritional status of plants to be determined quickly and at low cost, especially N, have been proposed by Souza et al. (2011b) and Souza et al. (2018).

In the present study, it was possible to see variations in both the chlorophyll index (CHLI) and the Nitrogen Balance Index (NBI) between seedlings of the Capdeboscq cultivar that came from seeds of the UFPel-Management and those from the Farmer-Management (Table 3). This demonstrates a significant difference in N dynamics by the 'Capdeboscq' seedlings, since 50% to 70% of the total N in the leaves makes up part of the enzymes and pigments which are associated with the chloroplasts (Souza et al., 2019).

Souza et al. (2017), evaluating nutritional performance in 'Capdeboscq' seedlings using the same nutrient solution as in the present research, obtained mean values for N (22.50) and the SPAD index (38.8) similar to those recorded in the present study in seedlings from seeds of the UFPel-Management (Table 3). This nutritional and physiological similarity is important, since little is known about the morphophysiological and nutritional aspects of *P. persica* seedlings produced in a greenhouse.

Considering the economic potential of cultivating stone fruit in Brazil, especially the peach, there is still a lack of knowledge related to the management and study of the post-harvest physiology of the seed of this species. Therefore, the improvement in post-harvest seed management, together with the tests for germination, vigour and seedling growth presented in this study are of great importance for Brazilian peach growers and nurserymen, with

Table 3. Mean values for stem diameter (SD, mm), root dry weight (RDW, g), shoot dry weight (SDW, g), total dry weight (TDW), Dickson quality index (DQI), N balance index (NBI), Chlorophyll index (CHLI) and SPAD index, in *P. persica* 'Capdeboscq' seedlings from seeds submitted to Farmer-Management and to UFPel-Management at 24 DAP in a greenhouse

Management	SD	RDW	SDW	TDW	DQI	NBI	CHLI	SPAD index
Farmer	2.14 b	0.174 b	0.204 b	0.376 b	1.24 b	17.45 b	18.90 b	30.08 b
UFPel	2.97 a	0.192 a	0.286 a	0.456 a	1.58 a	23.45 a	22.07 a	37.29 a
CV%	0.79	2.73	2.24	2.31	2.39	3.51	2.04	1.43

Mean values followed by the same lowercase letter do not differ by Tukey's test ($p \leq 5\%$).

the aim of producing plants of better physiological quality. On the other hand, the ring nematode *Mesocriconema xenoplax* (Raski) Loof, has been associated with Peach Tree Short Life Syndrome (PTSL), whose symptoms are usually identified at the end of the dormancy period, and are characterised by a reduction or paralysation of growth and a reduction or lack of bud sprouting and flowering. Parasitism by *M. xenoplax* in the peach tree also causes darkening of the root system, followed by destruction of the tissue and atrophy of the roots (Kuhn et al., 2015).

Conclusions

The proper post-harvest processing of the stones has a direct influence on maintaining the physiological quality of seeds intended for the production of rootstock, resulting in greater vigour and initial seedling quality.

The traditional management system for peach stones intended for the production of rootstock, compromises the physiological quality of the seeds, reducing the percentage of germination, the vigour and final quality of the seedlings.

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