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Correlation of biometrical characteristics of fruit and seed with twinning and vigor of *Prunus persica* rootstocks[1]

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ABSTRACT – This study aimed at determining and correlating the main morphometric characteristics of fruits and seeds with the germination potential and vigor of eight peach rootstocks. The experimental design was completely randomized, with four replications of 50 seeds per treatment. The analyzed variables were: length, width, thickness, fresh mass of fruit and seeds, moisture content of seeds, percentage of seeds attacked by fungi and intact seeds, germination percentage, germination speed index (GSI) and mean germination time (MGT). The Capdeboscq, Aldrighi and Tsukuba 1 cultivars showed higher values of length, width, thickness and fresh mass of seeds compared to other cultivars, presenting a relationship with the later period of fruit ripening. Regardless of the fruit ripening time, the germination percentage was high, ranging between 93% and 100% for all cultivars. In addition, the fresh mass of fruits has showed a high positive correlation with the fresh mass of seeds for the eight evaluated peach rootstocks. The GSI and MGT have a high relationship with the fresh mass of seeds.

Index terms: Rosaceae, morphobiometry, seeds, cultivars, peach.

Correlação de características biométricas de frutos e sementes com a germinação e vigor de porta-enxertos de *Prunus persica*

RESUMO - Objetivou-se determinar e correlacionar as principais características morfométricas de frutos e sementes sobre o potencial de germinação e o vigor de oito porta-enxertos de pessegueiro. O delineamento experimental foi inteiramente casualizado com quatro repetições de 50 sementes por tratamento. As variáveis analisadas foram: comprimento, largura, espessura, massa fresca dos frutos e sementes, grau de umidade de sementes, porcentagem de sementes intactas e infectadas por fungos, porcentagem de germinação, índice de velocidade de germinação (IVG) e tempo médio de germinação (TMG). As cultivares Capdeboscq, Aldrighi e Tsukuba 1 apresentaram maiores valores médios de comprimento, largura, espessura e massa fresca de sementes comparados com as demais cultivares, apresentando relação com o período mais tardio de maturação dos frutos. Independente da época de maturação dos frutos, a porcentagem de germinação foi alta variando entre 93% e 100% para todas as cultivares. Somado a isso, a massa fresca do fruto apresenta alta correlação positiva com a massa fresca das sementes dos oito porta-enxertos de pessegueiro avaliados. O IVG e o TMG possuem alta relação com a massa fresca das sementes.

Termos para indexação: Rosaceae, morfobiometria, sementes, cultivares, pessegueiro.

Introduction

According to the Food and Agriculture Organization of the United Nations, the production of peaches and nectarines occupied the eighth place in the world fruit ranking in 2013, providing 21 million tons that were produced in an area of 1.5 million hectares, three times the volume produced in 1983 (seven million tons), representing a 300% growth.

With a 232 thousand ton production of peaches and nectarines, Brazil occupies the 13th position in the world ranking, with 1.1% total production of these fruits (FAO, 2016); Rio Grande do Sul state is the biggest peach production center (127.936 tons), followed by the states of São Paulo (31.209 tons), Santa Catarina (20.963 tons) and Minas Gerais (19.912 tons) (IBGE, 2016).

Despite the fact that stone fruit production is very important in Brazil, the country still presents relatively low orchard average productivity, mainly in Rio Grande do Sul state (Mayer et al., 2014). Among factors associated to this low productivity, there are pest incidence, diseases, and climate.

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Another factor is the low quality of the plants produced, mainly in the form of rootstock production; in general, they are obtained from seeds discarded by the canning industry and, therefore, with no genetic identity. This type of plant material is not proper to be used as a rootstock (Picolotto et al., 2012), due to the susceptibility to various biotic and abiotic stressful factors, which occur into the soil and may affect the longevity and productivity of plants.

On the other side, the production of rootstocks by seeds may be feasible if proper selected genotypes are used (Bianchi et al., 2014). In this context, the selection of rootstocks and the study of the productive characteristics and quality of fruit and seeds help identifying cultivars with desirable characteristics, such as seed sanitary and physiological quality, resistance to pests and soil diseases; such characteristics will influence the lifespan of the future orchard.

Peach fruits, both from cultivars and rootstocks, are fleshy drupes with a fine pericarp, a fleshy mesocarp and a hardened endocarp with variable size, flat and ovoidal shape and corrugated surface, with or without pulp adherence; on the inside, one or two seeds may be found (Bacvonkral et al., 2014). The size of fruit and seed is determined by the genetic characteristics of each cultivar and is also influenced by plant management (İmrak et al., 2015).

Biometric studies of fruits and seeds and their phenotypical correlations allow the quantitative evaluation of a character's relevance in relation to another (Felizardo et al., 2015); they may help the best interpretation of seed germination tests in laboratory, since they analyze a simultaneous set of characters. Therefore, the proper methods to analyze stone fruit seeds have been of great interest, especially when they aim at obtaining information expressing their physical and physiological quality (Wang et al., 2016; Thakur, 2015; Szymajda and Żurawicz, 2014; Moreira et al., 2012; Rahemi et al., 2011).

In this context, it is necessary to conduct researches about the biometric and morphological characteristics of fruit and seeds, aiming at the maximum germination capacity and seed vigor of promising cultivars to be used as rootstocks for peach, nectarine and plum cultures, and spread such information to nurserymen and peach producers.

Considering the aforementioned, this work aimed at determining and correlating the main morphometric characteristics of fruits and seeds on the germination potential and vigor of eight peach rootstocks.

Material and Methods

In order to conduct the study, fruits of peach rootstocks were harvested during the crop year 2014-2015, coming from clonal mother plants aged 10, from the Aldrighi, Capdeboscq, Flordaguard, Tsukuba 1, Tsukuba 2, Tsukuba 3, Okinawa roxo and the Selection UFPel 0402 (Selection obtained by open pollination of cv. Okinawa) cultivars, kept in the *Prunus* rootstock Collection of the UFPel, with 31º 52' 00'' S; 52º 21' 24'' W coordinates and 40 m altitude.

After harvesting, fruits were taken to the Plant Molecular Physiology Laboratory, Botanic Department (UFPel), where they were washed under tap running water, discarding the damaged ones. For the biometric characterization, 200 fruits were asystematically chosen from each cultivar; measures of length (cm), width (mm) and thickness (mm), using a digital pachymeter with 0.01mm precision, and fresh fruit mass (g) were registered. Subsequently, the fruits pulp were removed from the endocarps, which were washed under tap running water in order to remove pulp residues and then submitted to an immersion treatment into fungicide solution (Orthocid® 500 – 12 mg.L^{-1}) for twelve hours.

The endocarps were dried in the shadow, where they were kept for two weeks, until being used. In order to obtain seeds, 200 endocarps from each cultivar were flambéed with 70% alcohol, for eight seconds, and then broken with a manual lathe in order to remove seeds. After that, morphobiometric and seed quality evaluations were performed, recording the variables: percentage of intact seeds, percentage of seeds with fungi incidence, percentage of endocarps with double seeds, length, width, thickness and individual fresh mass of seeds. The moisture degree of seeds from the eight peach rootstocks was also determined, using 50 seeds from each cultivar, by oven method (105 ± 3 °C), for 24 hours, with results expressed in percentage (wet basis) (Brasil, 2009).

The experimental design used for all analyzed variables was completely randomized with four replications of 50 seeds, totalizing 200 seeds per treatment.

For the germination test, seeds were submitted to a disinfestation process with 2% hypochlorite solution, for five minutes, and were later washed three times in distilled water. Thus, 25 seeds were placed in each Petri dish (90x15 mm), containing filter paper that was moistened with 4 mL of fungicide solution (Orthocid® 500 – 12 mg.L^{-1}). Petri dishes were closed, sealed with parafilm and taken to a BOD (Biochemical Oxygen Demand) chamber at a temperature of 7 ± 0.5 °C for seed stratification, in the absence of light, where they remained for 23 days, enough time for the germination of more than 90% seeds of all evaluated rootstocks.

During the stratification period, seeds were observed every two days in order to check the beginning of radicle emission, to determine germination speed index (GSI) and mean germination time (MGT), which were estimated by the formulas proposed by Maguire (1962) and Labouriau (1983), respectively.

The averages of the evaluated variables were submitted to analysis of variance and average comparison by Tukey's test at 5% probability, using the Sisvar software (Ferreira, 2011). In order to calculate the Pearson correlation among the characteristics of fruits, seeds and germination the R software was used (2008). Homogeneity and normality of the sample distribution were analyzed by Bartlett's and Shapiro-Wilks' tests.

Results and Discussion

Among the eight evaluated cultivars, 'Aldrighi', 'Capdeboscq' and 'Tsukuba 1' presented higher average values of length, width, thickness and fruit fresh mass, in relation to fruits from the other evaluated rootstocks (Table 1). Fruits from each cultivar were collected from plants cloned by grafting, so the differences in the biometric characters obtained from fruits of different peach rootstocks are associated mainly to the genetic characteristic of each genotype and the interaction with environmental factors during the fruit flowering and development, which influence the period between flowering and fruit ripening.

Table 1. Mean values of length, width, thickness and fresh mass of fruits from eight *Prunus persica* cultivars.

Cultivars	Length (mm)	Width (mm)	Thickness (mm)	Mass (g)
Aldrighi	49.28 a*	44.45 a	44.59 a	50.88 a
Capdeboscq	49.10 a	44.67 a	44.02 a	51.61 a
Flordaguard	42.09 b	41.77 b	40.81 b	42.08 b
Sel. UFPel 0402**	42.07 b	41.67 b	40.02 b	42.61 b
Okinawa roxo	42.73 b	42.35 b	41.59 b	43.59 b
Tsukuba 1	48.60 a	44.67 a	44.02 a	50.39 a
Tsukuba 2	41.17 b	40.93 b	40.94 b	42.18 b
Tsukuba 3	41.80 b	41.25 b	40.05 b	41.60 b
Average	44.61	42.72	42.01	45.62
CV.%	2.84	2.88	2.19	6.13
DMS	2.94	2.88	2.14	6.38

*In the column, averages followed by the same letter do not differ among themselves by Tukey's test ($p \leq 5\%$).
** Selection obtained by free pollination of cv. Okinawa

Considering that the existing knowledge on stone fruit, especially cultivars, has been successfully achieved by many research institutions hosted in the Southern and Southeastern regions of Brazil, it is possible to observe that there is still a need for efforts that may allow the continuity of documented scientific information generation, about data regarding the morphobiometrics of fruits and seeds of selected genotypes to be used as rootstocks. Such information is essential to know the use and germination potential of each genotype, serving as an indicator in obtaining seedlings with better quality, as well as helping in the recommendation and selection of new cultivars of rootstocks with improved characteristics.

Researches conducted with fruit and seeds of species from different botanical families such as majestic heaven lotus (*Gustavia augusta* L., Lecythidaceae Family) by Silva et al. (2014), cucumber tree (*Averrhoa bilimbi* L., Oxalidaceae Family) by Santos et al. (2015), and *Campomanesia adamantium* (Myrtaceae Family) by Dresch et al. (2013), indicated that longer and wider fruits presented higher values of fruit mass and seeds. Similar results were obtained in the present study, where the three rootstocks presenting higher values of fruit mass (Table 1) were also the ones presenting seeds with bigger dimension and seed fresh mass (Table 3).

It is known that there is a relation between fruit mass and fruit development cycle (Silva et al., 2013), where cultivars such as 'Capdeboscq', 'Aldrighi', 'Tsukuba 1', 'Tsukuba 2' and 'Tsukuba 3' (Table 2) that normally have a longer cycle; this factor is associated to the longer period of time for the seed growth, development and embryo maturation.

'Capdeboscq' and 'Aldrighi' seeds have been used for a long time to obtain rootstocks, due to the good climate adaptation to Brazilian conditions and seedling vigor (Fachinello et al., 2005). On the other side, these cultivars, in addition to being susceptible to soil phytonemotoids (Gomes and Campos, 2003), have fruits with endocarp-adherent pulp, characteristic that hinders its cleaning.

In turn, the Tsukuba 1 cultivar presented fruit and seed characteristics that did not differ from 'Aldrighi' and 'Capdeboscq' (Table 2). Such characteristics may be associated to the ripeness period of fruits (Table 2); the seedlings has good vigor, consisting in a good indicator of the potencial form use of this rootstock, since this cultivar has the additional advantage of *Meloidogyne* spp. resistance (data in publishing stage), and fruits have non-adherent to the endocarp pulp.

After breaking endocarps, the 'Selection UFPel 0402' and 'Okinawa roxo' presented higher percentage of endocarps with double seeds; the lowest percentages of double seeds were found in 'Capdeboscq', 'Tsukuba 1' and 2' (Table 2). Knowing the quantity of seeds inside the endocarp allows calculating the necessary quantity of seeds to obtain a determined number of desired rootstocks.

Table 2. Fruit ripening period, percentage of intact seeds (IS), seeds with fungi (SF), double seeds (DS) obtained in eight peach cultivars.

Cultivars	Ripeness period	IS (%)	SF (%)	DS
Aldrighi	01/05 to 01/25	77.11 c	17.89 a	5.00 b
Capdeboscq	01/05 to 01/25	79.00 c	18.22 a	2.78 c
Flordaguard	12/25 to 01/10	93.78 a	2.22 c	5.00 b
Sel. UFPel 0402	11/01 to 12/20	83.00 b	10.0 b	8.00 b
Okinawa roxo	11/01 to 12/20	86.70 b	0.00 c	13.30 a
Tsukuba 1	01/01 to 01/10	84.29 b	1.11 c	14.60 a
Tsukuba 2	01/01 to 01/10	87.67 b	10.33 b	2.00 c
Tsukuba 3	01/01 to 01/10	94.50 a	4.11 c	1.39 c
Average		85.76	7.99	6.51
CV.%		5.09	12.36	9.56
DMS		4.77	4.78	2.12

*In the column, averages followed by the same letter do not differ among themselves by Tukey's test (p≤5%).

In endocarps presenting double seeds, the size and mass of these seeds is normally lower, compared to the ones coming from endocarps with a single seed. 'Flordaguard' and 'Tsukuba 3' were the cultivars with the highest percentage of intact seeds and, together with 'Okinawa roxo' and 'Tsukuba 1' presented the lowest percentage of fungi attack [normally by *Monilinia fructicola* (Gl Wint.) Honey].

On the other hand, 'Aldrighi' and 'Capdeboscq' were the cultivars presenting the lowest percentage of intact seeds and the highest percentage of seeds with fungi attack (Table 2). One of the factors contributing to these results is the fact that these two cultivars have endocarp-adherent pulp, hindering its cleaning and increasing the development of fungi attacking the seed.

In the traditional method of peach rootstock seed stratification, during post-harvest, endocarps are kept in the shadow, under tree crowns, for a period varying from 75 to 150 days, so that pulp remains on endocarps rot and/or dry (Mayer et al., 2014). In this stratification method, it is normally possible to obtain percentages of field germination close to 30% (Mayer and Antunes, 2010), which is due to conditions that support the incidence of phytopathogens and the rotting of peach rootstock seeds, or to the fact that it reduces the physiologic quality of seeds, expressing lower germination percentages, emergence and seedling vigor, low stand and yield in the production of seedlings.

One of the alternatives to circumvent this problem is using rootstocks that produce fruits with non-adherent pulp to the endocarp, which helps cleaning and a lower incidence of fungi attack. Such characteristic is present in the 'Flordaguard', 'Selection UFPel 0402', 'Okinawa roxo', 'Tsukuba 1', 'Tsukuba 2' and 'Tsukuba 3' rootstocks, which also presented higher average values of intact seeds and lower values of seeds attacked by fungi (Table 2). In addition to this, another decisive factor is the proper endocarp management during post-harvest; they have to be cleaned, dried in the shadow and kept in a ventilated area during the storage period, until they undergo stratification. In this process, treating endocarps with a recommended fungicide for the culture also helps reducing the inoculum source of fungi that affect seed feasibility.

The process of seed germination is one of the most important steps to establish seedlings. For example, the quicker seed germination is, the least time they undergo adverse conditions, increasing the possibilities of seedling establishment (Lopes and Franke, 2011).

Table 3. Mean values of length (mm), width (mm), thickness (mm), seed fresh mass (g), germination (G, %), germination speed index (GSI) and mean germination time (MGT) obtained in seeds from eight peach rootstock cultivars.

Cultivars	Length	Width	Thickness	Mass	G	GSI	MGT
Aldrighi	16.99 a	7.86 a	5.48 a	0.40 a	100 a	10.58 a	12.1 a
Capdeboscq	17.37 a	7.66 a	5.43 a	0.41 a	100 a	12.15 a	12.4 a
Flordaguard	15.01 b	6.88 b	4.60 b	0.32 b	93 a	7.82 c	15.1 b
Okinawa roxo	15.56 b	6.71 b	4.39 b	0.28 b	100 a	10.36 a	11.6 a
Sel. UFPel 0402	15.04 b	6.62 b	3.09 c	0.24 c	100 a	11.89 a	11.9 a
Tsukuba 1	16.87 a	7.99 a	5.37 a	0.39 a	100 a	9.17 b	18.5 c
Tsukuba 2	15.02 b	6.78 b	4.51 b	0.30 b	93 a	5.41 d	23.0 d
Tsukuba 3	15.10 b	6.84 b	4.55 b	0.28 b	93 a	5.49 d	23.0 d
Average	15.87	7.16	4.67	0.32	97.37	9.11	15.9
CV.%	1.73	2.59	2.14	4.37	4.35	6.21	5.8
DMS	1.11	0.74	0.75	0.04	9.92	2.38	1.66

*In the column, averages followed by the same letter do not differ among themselves by Tukey's test (p≤5%).

Seeds of ‘Capdeboscq’, ‘Aldrighi’, ‘Selection UPFel 0402’ and ‘Okinawa roxo’ presented lower mean germination time and higher germination speed index, whereas seeds from ‘Flordaguard’, ‘Tsukuba 2’ and ’Tsukuba 3’ rootstocks presented higher germination average time and lower germination speed index (Table 3). Such characteristics are related to the chilling requirement to overcome the physiological seed dormancy, which is higher in these last three cultivars, compared to the others.

The determination of seed water content at the beginning of the evaluation is important to obtain consistent results in the evaluation of the physiological potential, which allow making comparisons between different genotypes (Ataide et al., 2016). In this study, the water content of eight analyzed peach rootstock seeds was determined, and whose values were 7.1% for cv. Okinawa roxo, 10.8% for ‘Selection UPFel 0402’, 10.6% for ‘Aldrighi’, 10.8% for ‘Tsukuba 1’, 10.1% for ‘Tsukuba 2’ and ‘Tsukuba 3’, 10.6% for ‘Flordaguard’ and 11.1% for ‘Capdeboscq’. The average contents of cultivar seed moisture are within the amplitude range of maximum accepted variation, which is from 3 to 4 percentage points (Nakagawa, 1999). However, it is necessary to consider that in this research, comparisons were made between seeds from genetically different cultivars, and not between seeds from the same cultivar.

According to results from Table 3, Capdeboscq, Aldrighi and Tsukuba 1 cultivars presented higher average values of length, width, thickness and seed fresh mass, compared to the other cultivars. For the germination percentage, 62.5% rootstocks presented 100% germination, whereas 37.5% presented 93% with average time varying from 12 to 23 days (Table 3); they are highly satisfactory results for all evaluated genotypes, when compared to the germination rate related by Mayer and Antunes (2010), in the traditional method of rootstock production directly on the field, whose germination rate and emergence is close to 30%.

In this study, fruit fresh mass presented positive and strong correlation with the seed fresh mass, which presented strong correlation with GSI and MGT (Table 4). According to Santos’ criterion (2010), correlation is considered high when it presents a $0.8 \leq r < 1.0$ correlation coefficient. Positive and moderate correlations were obtained between the fruit length and seed length variables (0.55) and between fruit width and seed thickness (0.57) (Table 4); it is considered moderate correlation when the correlation coefficient varies from $0.5 \leq r < 0.7$ (Santos, 2010). This result is similar to the one reported by Dresch et al. (2013), who observed a positive and moderate correlation between the fruit width of *Campomanesia adamantium* (Cambess.) O. Berg (Mirtaceae), with the seed thickness.

Table 4. Pearson correlation coefficient matrix*[ns] between fruit length (FL), fruit width (FW), fruit thickness (FT), fruit fresh mass (FM), seed length (SL), seed width (SW), seed thickness (ST), seed fresh mass (SM), germination speed index (GSI) and mean germination time (MGT) obtained in eight peach rootstock cultivars.

	FW	FT	FM	SL	SW	ST	SM	GSI	MGT
FL	0.70*	0.90*	0.68*	0.55*	0.21 ns	0.12 ns	0.69*	0.88*	0.64*
FW	-	0.80*	0.74*	0.56*	0.43ns	0.57*	0.52*	0.36 ns	0.12 ns
FT		-	0.55*	0.12 ns	0.20 ns	0.62*	0.34 ns	0.18 ns	0.02 ns
FM			-	0.85*	0.74*	0.72*	0.97*	0.94*	0.81*
SL				-	0.83*	0.55*	0.62*	0.85*	0.67*
SW					-	0.79*	0.64*	0.21 ns	0.07 ns
ST						-	0.43 ns	0.24 ns	0.00 ns
SM							-	0.97*	0.91*
GSI								-	0.03 ns

*, ns= Significant and non significant at 5% probability by Bartlett Shapiro-Wilks’ test.

Knowing the existing correlations between fruit and seed characteristics allows knowing the behavior of a variable through the analysis of another. In this case, with measures of width and thickness and, mainly, of fruit fresh mass, it is possible to estimate the width, thickness and fresh mass of seeds, that is, using bigger fruits, there is a guarantee of bigger seeds and, therefore, with higher germination potential and vigor.

According to Colombo et al. (2015), seeds with bigger mass present better germination capacity, standardization and seedling emergence, which is a result of the evaluated biometric characteristics; the fresh mass of fruits with the fresh mass of seeds had a direct and strong relation with the germination average time (Table 4).

Based on the results obtained in this research, it is possible to deduce that the bigger the fruit fresh mass is, the bigger the seed mass will be. Such characteristics are useful to help the selection of new genotypes to be used as rootstocks, which, in addition to the standardization of fruit lots with higher fresh mass, allows obtaining seed lots with higher mass and with the maximum germination power, implying in the reduction of MGT, and improving the homogeneity of seedling lots.

Conclusions

The biometric measures of fruits and seeds from Aldrighi, Capdeboscq and Tsukuba 1 cultivars presented positive and high correlation.

The fruit fresh mass presents high positive correlation with the seed fresh mass of eight evaluated peach rootstocks.

The germination speed index and the germination average time have high relation with seed fresh mass.

In the evaluated cultivars whose fruits have endocarp-adherent pulp, the percentage of phytopathogen infected seeds is higher, in relation to the ones whose pulp does not adhere to the endocarp.

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