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Germination and emergence of passion fruit (*Passiflora edulis*) seeds obtained by self- and open-pollination

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ABSTRACT. Seed dormancy is an important adaptive mechanism in many species and is generally lost during plant domestication because of selection that occurs through the collection and planting of seeds. We compared germination and seedling emergence in selfed and open-pollinated progenies obtained from eight passion fruit vines (genotypes). Self-pollination was performed at the button stage to overcome self-incompatibility. The experiment was a randomized block design in a factorial scheme (2 x 8; type of progeny x genotypes) with four replicates and 50 seeds per experimental unit. At 14, 21 and 28 days after sowing, the germination percentage and the emergence speed index were analyzed. The total length of seedlings (cm), shoot length (cm), radicle length (cm) and total dry matter of seedlings (g) were evaluated 28 days after sowing. The mass of 100 seeds (g) was determined before sowing. Differences were noted between genotypes and progeny types with respect to germination and emergence speed. In general, seeds obtained by selfing exhibited earlier germination and a higher emergence speed. It was concluded that seed dormancy is associated with the genotype of the embryo and is most likely conditioned by a dominant genetic effect.

Keywork: Passiflora edulis, dormancy, selection, dominance.

Germinação e emergência de sementes de maracujá-azedo (*Passiflora edulis*) obtidos por polinização aberta e autopolinização

RESUMO. A dormência de sementes é um importante mecanismo de adaptação das espécies e, geralmente, perdida durante a domesticação de plantas, devido à seleção através da coleta e plantio de sementes. Objetivou comparar a germinação e emergência de plântulas provenientes de sementes originadas de progênies autofecundadas e de polinização aberta de oito genótipos de maracujazeiro. As autopolinizações foram realizadas na fase de botão com intuito de superar a auto-incompatibilidade, e progênies de polinização aberta originadas do cruzamento natural com controle somente do genitor feminino. O experimento foi instalado no delineamento em blocos casualizados, em esquema fatorial 2 x 8 (tipo de progênies x genótipos), com quatro repetições e 50 sementes por unidade experimental. Aos 14, 21 e 28 dias da semeadura foram analisadas a porcentagem de germinação e o índice de velocidade de emergência. O comprimento total das plântulas (cm), comprimento da parte aérea (cm), comprimento da raiz (cm) e matéria seca total de plântulas (g) foram avaliados aos 28 dias de semeadura. A massa de 100 sementes (g) foi avaliada antes da semeadura. Há diferenças entre genótipo e tipo de progênie na germinação e velocidade de emergência. Verificou-se início de germinação adiantado e maior velocidade de emergência em sementes obtidas por autopolinização. Conclui-se que a dormência da semente está associada com o genótipo do embrião e que provavelmente é condicionado por um efeito genético dominante.

Palavras-chave: Passiflora edulis, dormência, seleção, dominância.

Introduction

Seed dormancy is an important mechanism of species adaptation that ensures a distribution of germination over time and prevents germination during adverse periods, which could cause extinction risks. Seed dormancy tends to be lost during domestication through successive selection during the collection and planting of seeds (GROSS;

OLSEN, 2010). Seed dormancy can be classified as physiological, morphological, or physical. Physiological dormancy is caused by endogenous factors and responds to heat or hormonal treatment. Morphological dormancy is caused by immature embryos, and physical dormancy is caused by a layer that is impermeable to water. These processes can function together to delay seed germination (BASKIN; BASKIN, 2004).

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Dormancy is regulated by genetic environmental components. The genetic control of dormancy has been studied in several species. Monogenic and polygenic inheritance were found to be controlled by dominant or recessive genes. Issa et al. (2010) concluded that dormancy in peanut is controlled by the action of a dominant gene. According to Andreoli et al. (2006), dormancy in wheat is determined by two recessive genes. Gu et al. (2004) found that rice seed dormancy is governed by multiple loci and epistasis. Genetic control of dormancy can be provided by the maternal genotype, the embryo or a combination of both genotypes (BASKIN; BASKIN, 2004). An understanding of the inheritance of dormancy is important for improving the selection of non-dormant genotypes in some species, including those that are not domesticated, and ensuring a more rapid germination rate. Maia et al. (2011) found that common bean seed qualities such as germination, emergence and seedling vigor can be controlled genetically. Genetic and environmental factors acting during seed production and seed drying can affect the permeability of the integument, thus determining the percentage and intensity of dormancy (NAKAGAWA et al., 2005; SAMARAH et al., 2004).

The germination of Passiflora seeds has mainly been studied via removal of the aryl and other techniques to stimulate germination, as reviewed by Alexandre et al. (2009). The presence of a hard coat that prevents water absorption and consequently inhibits germination was reported by Maciel et al. (1997) and Alexandre et al. (2004a), among other authors. The objective of this work was to compare germination among genotypes and between self- and open-pollinated progenies of passion fruit (*Passiflora edulis* Sims) to verify the effects of plant and embryo genotype at seed germination.

Material and methods

This work was conducted at the Department of Plant Science, Universidade Federal de Viçosa, Viçosa, Minas Gerais State, Brazil. Seeds were collected from eight plants (genotypes) after selfing and open pollination. Self-pollination was performed at the bud stage to overcome self-incompatibility as described by Bruckner et al. (1995). The open pollinated progenies resulted from natural pollination of the flowers.

Seeds were extracted from mature fruits and separated from the mucilage and aryl via friction with quicklime in a fine mesh sieve. The removed seeds were washed, placed on a paper towel and allowed to dry in the shade for three days. The seeds were sown at a depth of $0.5~\rm cm$ and spaced at $2~\rm x~2~cm$ in plastic

boxes ($40 \times 27 \times 10$ cm) containing fine washed sand as a substrate. The boxes were placed on a bench inside the greenhouse. The experiment was outlined as a randomized block design in a factorial scheme (2×8 ; type of progeny x genotype) with four replications and 50 seeds per experimental unit.

The measured traits were germination percentage (%) and emergence speed index (ESI) (MAGUIRE, 1962), and measurements were performed at 14, 21 and 28 days after sowing. The total seedling length (cm), seedling height (cm), length of the primary root (cm) and total dry mass of the seedlings (g) were evaluated on the 28th day. The weight of 100 seeds was quantified using a semi-analytical balance (0.001 g) before sowing.

The ESI was established based on daily assessments of seedling emergence beginning from the emergence of the first normal seedlings (11 days after sowing) until the 28th day. The shoot, root and total lengths of the seedlings were measured with a ruler graduated in centimeters after the seedlings were removed from the sand and washed. Subsequently, to determine the total dry matter mass, all the seedlings from each plot were placed in paper bags and incubated in a 60°C oven with air circulation for 72 hours until they reached a constant weight. The average air temperature inside the greenhouse was 22.6°C, and the minimum and maximum average temperatures were 17.3 and 27.9°C, respectively.

The data were subjected to analysis of varance, and the means were compared using the Tukey test (p \leq 0.05). The germination percentage data were transformed to arcsine (x 100^{-1})⁻², and the other data were not transformed.

Results and discussion

Interaction effects between genotype and type of progeny were found for germination percentage on the 14th day and for emergence speed index (ESI) on all evaluation days (p \leq 0.01). The interaction and type of progeny effects on seed germination disappeared at the subsequent evaluations (21st and 28th days). The influence of genotype on germination and emergence speed became strongly evident as previously reported by Alexandre et al. (2004b), who observed that genotype had a significant effect on germination percentage and ESI, suggesting that these traits could be utilized for the improvement of sour passion fruit. Melo et al. (2000) also recommended the selection of plants with enhanced germination rates for the genetic improvement of Passiflora spp. The absence of interaction and progeny effects on germination at 21 and 28 days indicates that because dormancy was overcome, the effect of progeny type disappeared. Interaction effects between genotype and type of progeny on the ESI were noted during all evaluations because this trait was influenced by the initial rapid germination.

Selfed progenies of six of the eight genotypes exhibited increased germination at 14 days after sowing (Table 1). The effect of progeny type indicates that the control of germination may be related to the genetic constitution of the embryo because the maternal tissues of the seeds had the same origin. According to Foley and Fennimore (1998), dormancy is usually a quantitative trait influenced by environmental factors, and its genetic control may be dominant or recessive depending on the species and the access. The results of this work are consistent with the hypothesis that dormancy is under dominant genetic control because the self-pollinated seed exhibited a higher germination percentage initially. A higher proportion of recessive homozygotes is expected in seeds resulting from self-pollination. Seeds from natural pollination are derived from crosses, and the passion fruit vine is self-incompatible (SUASSUNA et al., 2003); therefore, passion fruit seeds would be predominantly heterozygous. The absence of the same phenomenon in two genotypes (e.g., 4 and 5) is quite feasible because it would be unlikely that all the original plants were heterozygous at potential dormancy gene loci. The observed germination percentage was similar in magnitude to that described by Lima et al. (2006) and Wagner Júnior et al. (2006).

Seed dormancy can be conditioned by the peripheral tissues (pericarp, seed coat or endosperm) or by the embryo itself (BASKIN; BASKIN, 2004). The previously discussed data provide evidence for the effect of the embryo. Gu et al. (2004) reported that seed dormancy is more pronounced in natural rice accessions than in cultivars, indicating that

domestication and inbreeding reduce seed dormancy. In this study, the more rapid germination of seeds obtained by selfing supports the hypothesis that the absence of dormancy is a recessive trait. The elucidation of the number of loci controlling dormancy in *Passiflora edulis* and their inheritance will depend on future studies. In rice, Gu et al. (2004) found evidence of multiple dormancy loci and epistatic control. Dormancy is recessive in the wild oat (FOLEY; FENNIMORE, 1998). The use of self-pollination to assist the selection of germination uniformity and increased emergence speed in passion fruit should be a goal of future studies.

In four genotypes (1, 3, 7 and 8), ESI was influenced by progeny type at all three evaluation times (Table 2). In the genotypes that exhibited no difference in germination at the 14th day (4 and 5), there was no difference in ESI between the two types of progeny. This was also the case for genotype 2, which did not exhibit a progeny effect but showed very low germination (Table 1). Genotype 6 showed a difference in ESI only on the first evaluation day. Although a progeny effect on germination was observed in this genotype, the difference was less pronounced as that observed in the other genotypes (Table 1). Therefore, the ESI value was influenced by germination in the first evaluations.

Seedling growth traits were less affected by both genotype and type of progeny. For total dry matter mass (DM), there was significant interaction ($p \le 0.05$) between genotype and type of progeny and also a significant genotype effect. The DM means differed among genotypes; however, they only differed between type of progeny in genotype 8 (Table 3). The shoot length was affected by the genotype and the type of progeny ($p \le 0.05$), although no differences were noted by the Tukey test. There was no significant effect on the total length of seedlings or the length of the radicle ($p \le 0.05$).

Table 1. Germination of seeds from eight genotypes of *Passiflora edulis* originating from self (S0) and open pollination (OP) at 14 days (Ger 14), 21 days (Ger 21) and 28 days (Ger 28).

Genotype	Ger 14 (%)		Ger 21 (%)		Ger 28 (%)		
	Progeny						
	OP	S0	OP	S0	OP	S0	
1*	6.0Bde	55.0Acd	90.5Aab	95.0Aab	94.0Aab	98.0Aa	
2	0.5Bc	8.0Ae	61.5Ac	60.0Ac	81.0Ab	80.5Ab	
3	11.0Bcd	68.0Abcd	85.0Bb	95.0Aab	94.5Aab	98.0Aa	
1	23.0Abc	19.0Ae	86.5Ab	87.5Aab	92.0Aab	94.0Aab	
5	41.5Aab	49.0Ad	93.5Aab	86.0Ab	98.0Aa	90.0Bab	
6	50.5Ba	86.5Aab	97.0Aab	98.5Aa	98.5Aa	98.5Aa	
7	21.0Bbcd	92.5Aa	94.0Aab	95.5Aab	96.5Aa	96.0Aa	
8	28.5Babc	76.5Aabc	98.0Aa	95.5Aab	99.5Aa	97.5Aa	

Means followed by the same capital letter (in rows) and the same lowercase letter (in columns) are not significantly different based on the Tukey test ($p \le 0.05$). *, Identification of each passion fruit genotype.

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Table 2. Index of emergence speed at 14 days (ESI 14), 21 days (ESI 21) and 28 days (ESI 28) for *Passiflora edulis* seeds of eight genotypes originating from self (S0) and open pollination (OP).

Genotype	ES	I 14	ES	I 21	ES	I 28	
	Progeny						
	OP	S0	OP	S0	OP	S0	
*	0.221Bcd	2.085Ac	2.701Bbc	3.342Abcd	2.769Bc	3.397Abc	
<u>)</u>	0.020Ad	0.317Ad	1.563Ad	1.740Ae	2.118Ad	2.202Ae	
}	0.425Bcd	2.759Abc	2.596Bc	3.584Aabc	2.820Bbc	3.639Aabc	
	0.969Abc	0.709Ad	2.971Aabc	2.729Ad	3.109Aabc	2.862Ad	
	1.677Aab	1.948Ac	3.276Aab	3.039Acd	3.382Aa	3.159Acd	
•	2.000Ba	3.397Aab	3.429Aa	3.767Aab	3.483Aa	3.767Aab	
	0.792Bcd	3.781Aa	3.055Babc	3.876Aab	3.108Babc	3.887Aab	
3	1.043Bbc	3.371Aab	3.251Bab	3.999Aa	3.278Bab	4.046Aa	

Means followed by the same capital letter (in rows) and lowercase letter (in columns) are not significantly different based on the Tukey test ($p \le 0.05$). *, Identification of each passion fruit generates

Table 3. Shoot length (SL) and dry matter mass (DM) of seedlings and mass of 100 seeds (M/100S) from the progenies of eight *Passiflora edulis* genotypes originating from self (S0) and open pollination (OP).

Genotype	S	L		DM	M/1	.00S	
	Progeny						
	OP	S0	OP	S0	OP	S0	
1*	3.454Aa	3.301Aa	0.025Aab	0.023Abcd	1.10Acd	1.10Ab	
2	2.801Aa	2.861Aa	0.018Ab	0.021Acd	1.00Ad	1.10Ab	
3	3.232Aa	3.595Aa	0.020Ab	0.026Aabcd	1.40Aa	1.30Aa	
4	3.325Aa	3.948Aa	0.020Ab	0.019Ad	1.00Ad	0.90Ac	
5	3.488Aa	3.600Aa	0.030Aa	0.028Aabc	1.30Aab	1.10Bb	
5	3.537Aa	3.504Aa	0.026Aab	0.025Aabcd	1.00Ad	0.90Ac	
7	3.529Aa	3.968Aa	0.026Aab	0.030Aab	1.00Ad	1.00Abc	
8	3.382Aa	3.910Aa	0.026Bab	0.032Aa	1.20Abc	1.10Ab	

Means followed by the same capital letter (in rows) and the same lowercase letter (in columns) are not significantly different based on the Tukey test ($p \le 0.05$). *, Identification of each passion fruit genotype.

The mass of 100 seeds was numerically higher in seeds derived from open pollination in most genotypes; however, a significant difference was noted only in genotype 5 (Table 3). This trend may be related to inbreeding depression, which can occur with self-pollination in allogamous plants.

Conclusion

In this work, seed dormancy was related to the genotype of the embryo.

The seed dormancy in *Passiflora edulis* is most likely conditioned by a dominant genetic effect.

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