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EVALUATION OF POLLEN SOURCE ON SEXUAL COMPATIBILITY AND FRUIT SET IN CERTAIN APRICOT CULTIVARS UNDER THE CLIMATE OF ASSIUT GOVERNORATE

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ABSTRACT: This study examined the self-compatibility and cross-compatibility or incompatibility among three four-year-old apricot tree cultivars (Amal, Haid, and Solitaire) grafted onto Nemguard peach rootstock. The research was conducted under the climatic conditions of Assiut, on sandy soil at a private farm in Assiut Governorate, Egypt, over two successful seasons (2021/2022 and 2022/2023). Initial and final fruit set percentages were nearly different in the three studied cultivars, but Haid was superior to the others. Generally, it could be concluded that Haid, Amal apricot cultivars, respectively, could be the higher-yielding cultivars under Egyptian conditions, due to most of the pollen grains germinating on the stigma surface one day afterward pollination, and they grew to the end of ⅓ of the style 4 days after pollination, resulting in high fruit set. On the other hand, the Solitaire cultivar is reported to be self-incompatible. So, we can nominate Haid’s cv. As a self-compatible and good pollenizer for Amal and Solitaire apricot cvs.

Keywords: Apricot, Cultivars, Pollen, Compatibility, and Fruit Set.

INTRODUCTION

Apricot cultivars have traditionally been categorized into six eco-geographical groups based on their geographical origin: Central Asian, East Chinese, North Chinese, Dzhungar-Zailij, Irano-Caucasian, and European. The apricot cultivars from Central Asia, which are the oldest and most diverse, primarily belong to the Dzhungar-Zailij and Iranian-Caucasian groups and are mostly self-incompatible. The commercially cultivated varieties in Europe, North America, South Africa, and Australia are primarily self-compatible and fall under the European group (Herrera *et al*., 2019).

In Egypt, one of the significant cultivated deciduous fruit trees is the apricot (*Prunus armeniaca L*.). It encompasses a large area of stone fruits. The cultivated area totalled 13,076 feddan, producing 74,067 tons (Food and Agriculture Organization of the United Nations, 2023). Many varieties of apricot have been developed during their cultivation. However, there remains a need for new genotypes and the development of new varieties that possess a combination of economically valuable traits, are adapted to local growing conditions, and are free from viral pathogens (Fideghelli and Della Strada, 2008; Gorina, 2014).

For the global production of various crops that rely on pollinators, especially those of great economic value, insect-mediated pollination is essential. With the rise in agricultural demand and the ongoing decrease in pollinators, finding targeted management strategies to protect pollination services has become (Osterman *et al.*, 2024)

Apricot (*Prunus armeniaca L.*) is highly vulnerable to issues during pollination, making pollen production and viability crucial for successful fertilization. Low parameters can result in fruit set failure, consequently leading to diminished yield. A large number of apricot cultivars are frequently introduced, accompanied by scant details regarding flower quality. The phenomenon known as ‘pistil abortion’ occurs frequently and at a high rate in apricots, having a significant impact on the fruit production of this species. Over the past two decades, a few studies have looked into the production of apricot pollen (Gallotta *et al*., 2014).

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Many varieties of apricot need cross-pollination to produce fruit; therefore, verifying the quantity and quality of pollen is crucial from a horticultural standpoint. Choosing the right pollinizers could be crucial for fruit production, with specific cultivars being favored over others based on compatibility and the quantity and quality of pollen grains. The acetic carmine stain method is very rapid and straightforward, unaffected by the operator. It can be applied to large samples of pollen grains, enabling the differentiation of usually developed grains with cytoplasm (stainable grains) from those that are underdeveloped and immature (Szabo *et al*., 1999). This study aimed to evaluate the degree of self- and cross-compatibility among various apricot cultivars and to address the occurrence of heterostyly, a distinct form of polymorphism and herkogamy in flowers, which manifested in a 65% fruit set and a 35% fruit drop for the Solitaire cultivar.

MATERIALS AND METHODS

The experiment was conducted over two seasons (2021/2022 and 2022/2023) to investigate self- and cross-compatibility, as well as incompatibility, between them under the climate conditions of Assiut Governorate on three apricot tree cultivars (Amal, Haid, and Solitaire), four years old, grafted on Nemguard peach rootstock at 4×5 m apart (210 trees/feddan) on sandy soil at a private farm. Each treatment in this study consisted of five replicates, and each replicate contained three trees, as uniform in size and vigor as possible. Also, the experiment design was arranged in a split-plot design Analysis.

Pollen viability:

The viability of pollen grains was tested using a staining solution of 2,3,5-triphenyl tetrazolium chloride (T.T.C) following the methods described by Oberle and Watson (1952). The solution consisted of one part of 10% T.T.C. and ten parts of 60% sucrose.

Pollination procedure and sample collection:

Flowers of various studied types were gathered at the balloon stage, just one day before anthesis. In the Lab., at room temperature, the flowers were placed on paper. After a day, pollen grains from each variety were collected into separate glass tubes and considered as pollinizers for that particular variety. In all cross-pollination cases, hand forceps were used to emasculate flowers of the mother variety at the balloon stage. Cross-pollination was then performed immediately using a fine paintbrush to apply the appropriate pollen to the receptive stigmas.

Three hundred flowers on each shoot were put in a Pergamon bag after they were pollinated to prevent insects from spreading the pollen. During the balloon stage, various shoots and spurs were bagged to perform self-pollination treatments. For open pollination, 10 shoots and spurs per replicate were marked without any treatment. Flower samples were collected from all treatments to examine pollen germination, pollen tube development, and fertilization. These samples were taken after pollination in cross-pollination cases and after a thesis in both open and self-pollination. Samples were collected daily for a week following pollination and after a thesis. All samples were promptly fixed in 70% F.P.A. (formalin: propionic acid: 70% ethanol in a 5:5:90% volume ratio) immediately following collection, as per Gerlach (1984).

Pollen germination and pollen tube development in vitro:

The fixed flowers underwent washing in flowing water for 24 hours, followed by softening in 8 N NaOH for 2 hours. Then, the samples were cleaned for a night in running tap water. Finally, the flowers were treated with a staining solution of 0.1% aniline blue in 0.1N K3PO4 (Kho and Baer, 1968; Preil, 1970). The styles and stigmas were detached from the ovary and gently squashed under a pressing coverslip to monitor pollen germination and pollen tube growth. A Leica fluorescence microscope was used to examine the slides.

Impact of various pollination methods on initial and final fruit set percentages:

Fruit set data were gathered about different pollination treatments, including open, self-, and cross-pollination. The flowers were counted during the anthesis and balloon stages to determine their total number. The counted set fruits were evaluated 3 weeks after pollination or initial fruit set and 2 weeks after final fruit set.

Initial and final fruit sets, % of different pollination treatments were calculated:

Initial fruit set% = (Number of fruitlets/total number of flowers) x 100.

Final fruit set% = (Number of set fruits / total number of flowers) x 100

Statistical analysis:

The analysis of variance (ANOVA) in the MSTAT-C software package (Freed *et al.*, 1989) was done for each season separately as a split-plot design, The differences between mean values of treatments were compared by the least significant difference (LSD) test at a 0.05 level of probability according to procedures reported by Gomez and Gomez (1984).

RESULTS AND DISCUSSION

Pollen viability:

Table 1 presents that the pollen viability is significantly affected by different apricot cultivars under study in both seasons. Pollens from all apricot cultivars demonstrated a high viability rate after 72 hours in T.T.C., as well as after the other two time intervals (24 and 48 hours) across all tested seasons. In addition, the viability range of pollen grains from Haid was greater than that of Amal or Solitaire, which gave the lowest value. Milatović et al. (2016) reported that temperatures ranging from 15 to 25°C were optimal for the development of pollen tubes and the germination of pollen in several apricot varieties. This data is consistent with the previous findings. Pollen germination rates at 5 °C were higher in early-flowering varieties ('Laycot', 'Sylred', and 'Goldrich') than in late-flowering varieties ('Novosadska Rodna', 'Hungarian Best', and 'Pisana'). Consequently, these cultivars may be better adapted to the low temperatures encountered during flowering. The length of the pollen tube at 15°C and 25°C was six to twelve times greater than at 5°C. This leads to the conclusion that a temperature of 5°C, although sufficient for pollen germination, does not create optimal conditions for the growth of the pollen tube.

Table (1): Effect of pollen grains' viability in T.T.C. of the three apricot cultivars during (2021/2022 -2022/2023).

Cultivars	2021/2022			2022/2023		
	Time applications					
	24h.	48h.	72h.	24h.	48h.	72h.
Amal	68.59 b	72.21ab	78.83a	68.66 b	75.10 ab	79.85a
Haid	72.03 a	79.19a	80.85a	73.52 a	82.40 a	85.67a
Solitaire	58.46 c	66.51b	73.57a	59.85 c	69.66 b	76.63 a

The effect of the Self and cross compatibility and/or incompatibility between some apricot varieties:

The data presented in Figure 1 shows that partial cross-incompatibility occurred when flowers of the Amal apricot were pollinated with Solitaire pollen. Pollen tubes contained terminal plugs, but these were unable to penetrate the stigma surface of the Amal cultivar. Seven days post-pollination, a significant amount of non-germinated Solitaire pollen was noted on the stigma surface of Amal (Fig. 1, A). No growth of pollen tubes was observed along the style. The stigma surface of Amal exhibited numerous pollen grains. After pollination, some of this pollen germinated, and they developed to reach the first third of the style length within three days. Some pollen grains sprouted on the surface of the stigma; In 3 to 4 days, the pollen tube extended to ⅔ of the style's length and penetrated the ovules 6 days post-pollination (Fig. 1, B).

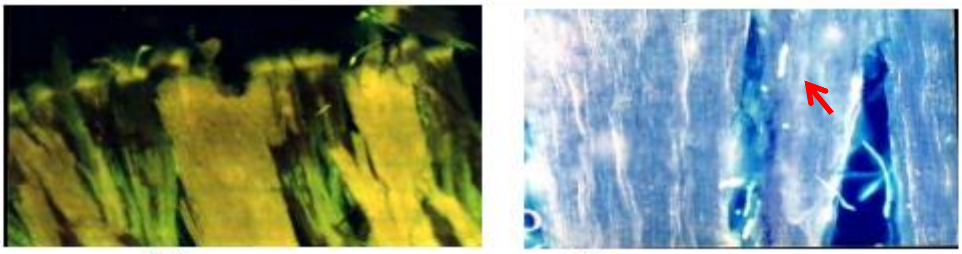

Fig. (1): Self and cross compatibility and/or incompatibility action between Amal and Solitaire apricot cultivars, Plugs in the upper part of the style.

In the Haid cultivar, most pollen grains germinated on the stigma surface one day after pollination, and four days after pollination, they grew to the end of ⅓ length of the style (Fig. 2, A). One day post-pollination, the stigma of Amal exhibited no germinated pollen (Fig. 2, B). In the case of Haid self-pollination and/or Amal apricot flowers pollinated with Haid pollens, a significantly elevated quantity of germinated pollens was noted in comparison to other examined pollinations 4 days post-pollination. They arrived at the base of the style; Different degrees of growth in selfed pollen tubes were revealed through microscopic examination. Three days post-po

During pollination, the pollen tubes extended to one-third of the style's length. Seven days post-pollination, they reached the inferior section of the style (Fig. 2, C, D).

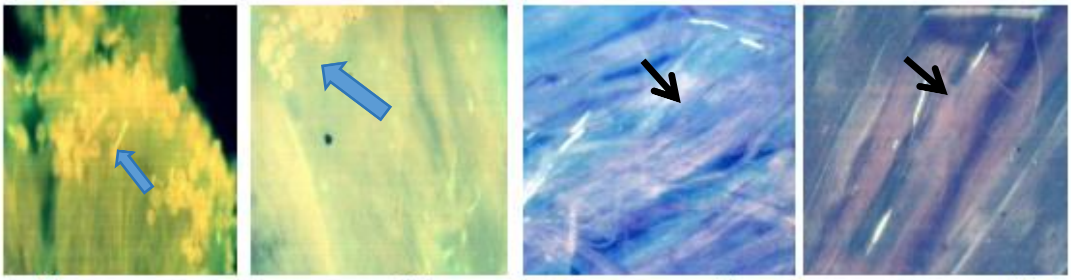

Fig. (2): Compatibility and or incompatibility between Amal and Haid apricot cultivars, with a high number of non-germinated pollen on the stigma surface.

Germinated pollens were found to be extremely poor on the stigma surface of the Solitaire cultivar. However, a few of the pollen tubes developed slightly and reached ⅔ of the style's length six days after pollination. Five days post-pollination, most pollen tubes contained callose plugs along their length and at their ends, as shown in the upper part of the style (Fig. 3, A). The majority of the pollen grains that germinated on the stigma surface of the Solitaire pollen tube exhibited slow growth, with a complete cessation occurring 7 days post-pollination. Several of the styles were displayed with regular growth of the pollen tube. Severe pollen deposition occurred on the ovule six days post-pollination. Fig. (3, B, C). Most of the pollen tubes that developed through the styles, however, showed signs of abnormal development. Thus, Solitaire selfing appeared to be a combination of non-self pollination.

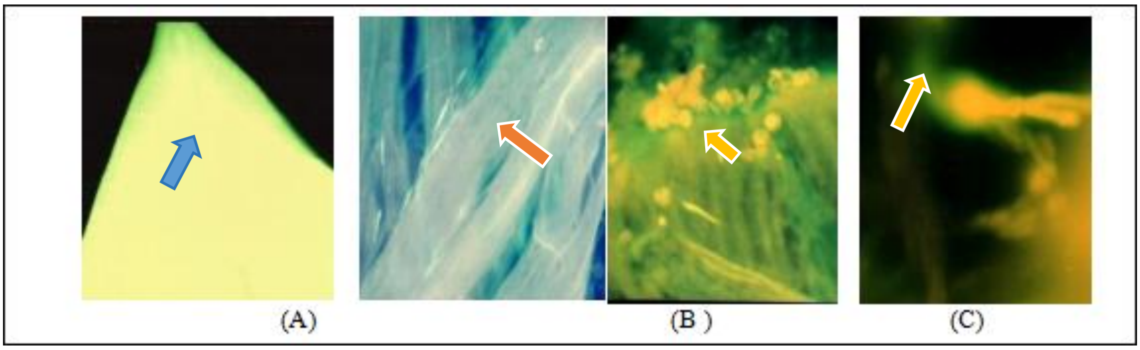

Fig. (3): Compatibility and or incompatibility between the three apricot cultivars. Frequently, callose plugs were observed along and at the end of the tubes - pollen tubes grew to about ⅓ the length of the style 6 days after pollination.

Ibrahim *et al.* (2005) studied the pollination of Canino, Amal, and Perfection apricot cultivars' characters. The pollination treatments used in this study were as follows: Open pollination, bagging only, and Crosspollination as follows:-(A) Perfection (♀) X Canino (♂). (B) Perfection (♀) X Amal (♂). (C) Perfection (♀) X {Canino+Amal} (♂). They found that Canino and Amal are regarded as self-compatible (fertile) cultivars. The cultivar known as Perfection is regarded as self-incompatible (sterile). With open pollination, the Canino and Amal cultivars exhibited higher fruit set percentages compared to those resulting from the bagging-only treatment. Moreover, using Canino pollen grains for cross-pollination of the Perfection cultivar resulted in a greater fruit set percentage. However, the cross-pollination by Amal pollen grains resulted in a lower fruit set percentage compared to the other pollination treatments in both seasons of the study. The pollen-pistil interactions in mango cv. Amrapali was studied as a result of self-, open-, and cross-pollination with cv. Sensation. It was noted that self-pollination in Amrapali led to a quicker drop in fruit retention compared to open- and cross-pollination (Amrapali × Sensation) within 20 days after pollination (DAP). After 25 days of pollination, only three fruits (0.26%) were noted from the 1,133 flowers that underwent self-pollination. they found that 'Amrapali' does not favor self-pollination. Nonetheless, cross-pollination leads to a considerable fruit set (Srivastav *et al.*, 2014).

Effect of different pollination treatments on the percentage of initial fruit set and retained fruits:

It is pretty evident, as shown in Table 2, that Haid selfing and open pollination have the highest initial fruit set and the highest retained fruits compared to the other cultivars under study. Moreover, the Haid cultivar is considered self-compatible. The combinations of Haid × Amal cultivar exhibited the greater percentage of initial and retained fruit set in both seasons, followed by Haid, Amal, and open pollination under study, and Amal selfing in the first and second seasons. On the contrary, the combination of Solitaire × Amal gave the lowest percentage of initial and retained fruit sets in the first and second seasons, respectively. Concerning the Solitaire cultivar, open pollination gave the lowest range of initial and retained fruit sets in both seasons. On the other hand, the combinations of Solitaire × Haid gave the greater initial and retained fruit sets compared to Solitaire × Amal or Solitaire selfing pollinations. Herrera *et al*. (2019) reported that pollination needs a collection of 92 apricot cultivars, comprising both traditional and newly released cultivars from various breeding programs and countries. The establishment of self-compatibility was achieved through microscopic observation of pollen tube behavior in flowers that underwent self-pollination. Incompatibility relationships among cultivars were determined through the identification of S-alleles using PCR analysis. This report presents the self-(in) compatibility of 68 cultivars and the S-RNase genotype of 74 cultivars for the first time. About half of the cultivars (47) demonstrated self-compatibility, while the other 45 showed self-incompatibility. The identification of S-alleles in self-incompatible cultivars enabled their allocation to 11 incompatibility groups, six of which are reported here for the first time. It is beneficial for the proper selection of apricot cultivars in commercial orchards and parental genotypes in breeding programs to determine pollination requirements and incompatibility relationships between cultivars. Abdel-Rahman *et al.* (2024) reported that during the flower study, the heterostyly phenomenon was noted in the Solitaire cv., suggesting that this cultivar should be crossbred with another apricot cultivar. To address this issue, either bloom simultaneously or distribute beehives for cross-pollination.

Table (2): Effect of the different pollinizers on initial fruit set % and retained fruits of three apricot cultivars during (2021/2022-2022/2023).

Pollination treatment		2021/2022		2022/2023	
Female	**Male**	**Initial fruit set (%)**	**retained fruit (%)**	**Initial fruit set (%)**	**retained fruit (%)**
Amal	**Selfing**	28.60 ef	18.55 g	29.20 ef	18.77 g
	Open	30.33 e	25.62 f	33.87 de	26.20 f
	Haid	45.70 bc	37.83 d	55.60 a	50.30 ab
	Solitaire	15.11 gh	6.17 j	15.17 gh	8.11 i
Haid	**Selfing**	50.53 ab	47.56 ab	63.60 a	58.80 a
	Open	46.83 b	43.35 c	50.55 ab	47.64 b
	Amal	46.46 b	43.30 c	50.00 ab	46.77 b
	Solitaire	27.60 f	16.55 g	30.20 e	17.77 g
Solitaire	**Selfing**	10.23 i	6.30 j	13.33 h	8.00 i
	Open	13.12 h	8.88 i	15.10 gh	9.12 i
	Amal	16.00 g	9.00 i	16.22 gh	9.88 i
	Haid	26.63 f	15.56	28.27 ef	16.74

Conclusion

The study found that Haid apricot had a greater viability range than Amal or Solitaire, which had the lowest value. The flowers of the Amal apricot were pollinated with Solitaire pollen, but the terminal plugs could not penetrate the stigma surface. Haid pollen grains germinated on the stigma surface one day after pollination and grew to the end of ⅓ of the length of the style. Haid self-pollination and Amal apricot flowers pollinated with Haid pollens had a significantly higher quantity of germinated pollens compared to other pollinations. Solitaire pollen was found to be very poor on the stigma surface. Haid open pollination had the highest initial fruit set and retained fruits compared to other cultivars. Haid selfing gave the maximum value of initial fruit set and retained fruits. The combination of Haid and Amal apricots showed the highest percentage of initial and retained fruit set in both seasons.

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تقييم مصادر حبوب اللقاح على التوافق الجنسى وعقد الثمار في بعض أصناف المشمش تحت الظروف المناخية لمحافظة أسيوط

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الملخص العربي

اجريت هذه الدراسة على ثلاثة أصناف من أشجار المشمش (أمل، حايد، وسوليتير) عمرها خمس سنوات، مطعمة على أصل خوخ نيمجارد، وتم التحقق من التوافق الذاتي والخلطي و/أو عدم التوافق بينها في ظل الظروف المناخية المصرية المزروعة في تربة رملية في مزرعة خاصة بمحافظة أسيوط، مصر خلال موسمين ناجحين (٢٠٢١/ ٢٠٢٢ و ٢٠٢٢/٢٠٢٣) كانت نسب عقد الثمار الأولية والنهائية مختلفه تقريبًا في الأصناف الثلاثة تحت الدراسه، لكن صنف الحايد كان متفوقًا على الأصناف الأخرى .وبشكل عام، يمكن الاستنتاج أن أصناف المشمش حايد، أمل، على التوالي هي الأصناف ذات المحصول الأعلى في ظل الظروف المصرية، وذلك بسبب إنبات معظم حبوب اللقاح على سطح الميسم بعد يوم واحد من التلقيح، ونمت حتى نهاية ⅓ من القلم بعد ٤ أيام من التلقيح، مما أدى إلى ارتفاع نسبة عقد الثمار . ومن ناحية أخرى، وجد ان صنف سوليتير غير متوافق ذاتيا. لذلك يمكننا ترشيح صنف المشمش حايد كصنف خصب ذاتيا وملقح جيد لكل من صنفي أمل وسوليتير.