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Assessing the Impact of Genotype-Specific Caprifig Fruit Storage on the Pollination Efficacy and Fruit Quality of "Bursa Siyahı" Cultivar: A Multivariate Analysis Approach

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Abstract: Fig types such as "Smyrna" and "San Pedro" require pollination (called caprification in fig cultivation) to produce a commercial crop, based on the crop and pollination characteristics of figs. Caprification is the process of hanging caprifig (male fig) fruits on female fig trees to ensure the transfer of pollen from the female fig to the caprifig by a wasp (*Blastophaga psenes*) that lives within the caprifig. It is necessary to extend the caprification period by using caprifig genotypes that ripen at different times, as female fig fruits ripen gradually. However, as caprifigs may not be continuously available for pollinating female figs, storing suitable caprifigs is necessary. The aim of this study was to assess changes in *Blastophaga psenes*, the duration of *Blastophaga*'s exit, and the viability of pollen from caprifigs of different genotypes (16 08 05, 16 08 09, 16 08 10, 16 09 10, and 16 ZF 08) stored for caprification. These stored caprifig genotypes were subsequently used for pollination three times at 8-day intervals, after which their impact on the set and quality of the edible fig fruits was evaluated. According to the average data, at the end of storage, the least *B. psenes* loss was obtained from the 16 08 05 (61.03%) genotype, and the highest was obtained from the 16 09 10 (67.00%) genotype. Pollen germination tended to increase with the storage of caprifig fruits, but this increase was not linear. After storage, the 16 08 09 and 16 09 10 genotypes exhibited greater pollen germination. The highest fruit set and quality were obtained when the 16 08 09 and 16 09 10 genotypes were used as pollen sources. Furthermore, since the 16 08 10 genotype is the latest ripening caprifig genotype, it has been determined that it can pollinate late-ripening "Bursa Siyahı" fruits. Principal component and path analysis demonstrated that pollen viability and germination rate were crucial in selecting caprifig genotypes for fruit set and quality.

Keywords: pollen source; pollination; caprification; fruit set; caprifig storage; genotype; pollen viability; number of *B. psenes*; xenia

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1. Introduction

Ficus carica L. is one of the 750 species of *Ficus* and includes gynodioecious species with bisexual trees (functional male figs or caprifig) and unisexual female trees [1,2]. Since functional male trees have hermaphrodite flowers, *Ficus carica* L. is usually considered gynodioecious rather than dioecious [3]. Four types of figs are described based on cropping and pollination characteristics. The "common fig" type requires no pollination to set a commercial crop. Botanists use the term "persistent" instead of "parthenocarpic" due to the fact that a fig is not a true fruit. The other two types of edible figs are not persistent and require pollination to set the main crop of figs. These non-persistent types are botanically classified as "cauducous" and are classed as Smyrna and San Pedro types. The San Pedro types are distinguished by setting a persistent early crop, known as "breba" fruit, but require pollination to set the main crop. The fourth type, caprifigs (male figs), serve as the pollen source for commercial plantings of cauducous figs. A male fig plant, known as a caprifig, exhibits distinct male and short-style female flowers. Edible figs contain

only long-style female flowers. A male fig tree bears fruit in three crop cycles during each growing season, occurring in summer (profichi), fall (mammoni), and winter (mamme). The main caprifig crop, known as profichi, coincides with the main summer crop cycle of female trees. While most male fig trees do not produce edible fruit, they play a crucial role as a pollen source for caprification (pollination), which involves the transfer of pollen grains from male trees to female trees through a vector, *B. psenes* L. [3]. *Blastophaga psenes* is a species of wasp in the *Blastophaga* genus that pollinates figs. They develop in caprifigs, which are kept separate from the figs to maximize pollination control. The process of hanging the caprifig fruits placed in bags or baskets on the Smyrna and Sand Pedro-type female trees and enabling the wasp to reach the female fruits is called "caprification". The caprification process needs to be repeated two or three times to achieve an economic yield for "Smyrna" and "San Pedro" figs as the syconia of female fig trees gradually become receptive [4,5]. Therefore, it is essential to identify two or three caprifig cultivars to extend the caprification period. Alternatively, fruits of caprifig cultivars can be stored at 4 °C for 14 days and used for caprification [6].

One of the most important factors affecting caprification success is the selection of caprifigs (a pollen source). When selecting caprifigs, the ripening period of the pollen in the male flowers must coincide with the pollen acceptance period of the female organs [7]. Additionally, factors such as the number of fruits on the shoot, number of *B. psenes*, *B. psenes* emergence and duration, number of male and gall flowers, pollen viability, and germination rate are considered [8,9]. Furthermore, it is desirable for the *Philotrypesis caricae* parasitoid to be scarce in caprifig fruit. This is because the larvae of *P. caricae* feed on the food source of *B. psenes* larvae, and *B. psenes* cannot compete with *P. caricae* larvae, leading to their death [10]. In addition to determining the caprifig's number of *B. psenes* and pollen viability under in vitro conditions, it is essential to test these parameters under in vivo conditions (caprification process) for a suitable pollen source selection [8,9,11].

The choice of pollen source can influence the size and shape of the fruits, their color, and biochemical content, which are collectively known as "xenia" [12]. Different pollen sources have been reported to increase fruit set, fruit size, seed size, and quality in blueberries [13,14], chestnuts [15], and dates [16,17]. Several studies have also investigated the effect of pollen sources on the fruit set, ripening, and fruit quality of edible fig cultivars widely used in Tunisia [5,18–20], Iran [4,21], and Syria [22]. The quality of an edible fig fruit depends on factors such as the size of the fruit flesh, ostiole and fruit cavity, soluble solids content, titratable acidity, and flesh and skin color. Medium-sized fruits with slight ostiole cracking and fruit cavities are considered high-quality fig fruits [23]. Additionally, the timing of ripening, whether early or late, is crucial for extended market presence and increased export potential [24]. However, there is only one published report on determining the effect of pollen sources on fruit set and fruit quality, which is among the Smyrna type figs, "Bursa Siyahı", one of the high-quality fresh fig products preferred in the world market whose export potential is increasing [25].

Recently, the cultivation of "Bursa Siyahı" edible fig orchards to increase Turkey's export capacity has increased. However, variations in fruit yield have been noted, attributed to pollination problems [26]. The main problems causing pollination issues include the use of caprifigs collected from random orchards, whose pollen quality and wasp numbers are unknown, and decreases in wasp emergence and pollen quality in fruits that are not stored in cold conditions [27]. For successful caprification, it is crucial that the genotype used has high *B. psenes* and pollen viability and can preserve them during storage. However, studies on the changes in the number of *B. psenes* and pollen viability in stored caprifig fruits under in vitro conditions are scarce. A previous study revealed that the viability of the wasp and pollen of the Karabulut caprifig genotype could be preserved for 16 days at 4 °C [28]. Also, Anjam et al. [29] observed a decrease of 68.74% in *B. psenes* after 7 days and 86.61% after 14 days in the "Poozdombali" caprifig genotype stored at 4 °C. On the other hand, Zare et al. [30] reported that the "Shanehi", "Gohari", "Kouhi", and "Poozdombali" caprifigs can be stored for 14, 18, 22, and 32 days, respectively. However, there are no

detailed studies determining the wasp and pollen quality of different caprifig genotypes based on the day of harvest or the storage period or assessing the effect of xenia on the "Bursa Siyahı" cultivar using these pollination genotypes.

Therefore, the present study aimed to examine the changes in the wasp and pollen characteristics of caprifig fruit of different genotypes depending on storage and to determine the effect of the use of stored genotypes during pollination on the fruit set and fruit characteristics of the "Bursa Siyahı" cultivar. A secondary objective was to determine the direct and indirect relationships between fruit set and quality and between the traits of the caprifig genotype wasp and pollen using multivariate analysis methods.

2. Materials and Methods

2.1. Plant Materials and Experimental Design

This study was conducted at the Agricultural Application and Research Center of Bursa Uludag University between 2020 and 2022. Data for minimum and maximum temperatures (°C), as well as rainfall (mm), were recorded throughout the pollination and fruit ripening period at the agro-meteorological station during 2020–2022 (METOS® by Pessl Instruments, Bursa, Turkey) (Figure 1). During July, rainfall was concentrated for only a few days, except in 2021, and did not interfere with the caprification or pollen sampling period. In 2022, precipitation was observed during the initial harvest period (228, 237, and 243 JD), whereas in 2020 and 2021, precipitation began to occur as of October (Figure 1). The experimental orchard has typical sandy soils with a pH ranging from 7.1 to 7.5 [31]. Agricultural techniques, including pruning, irrigation, and fertilization, were conducted following standard procedures in the area. In the experiment, the 16 08 05, 16 08 09, 16 08 10, 16 09 10, and 16 ZF 08 genotypes, which were 4 years old and planted at 5 m × 5 m intervals, were used as the male fig (caprifig). The "Bursa Siyahı" cultivar, which is 20 years old and planted at 10 m × 10 m intervals, was used as the female fig.

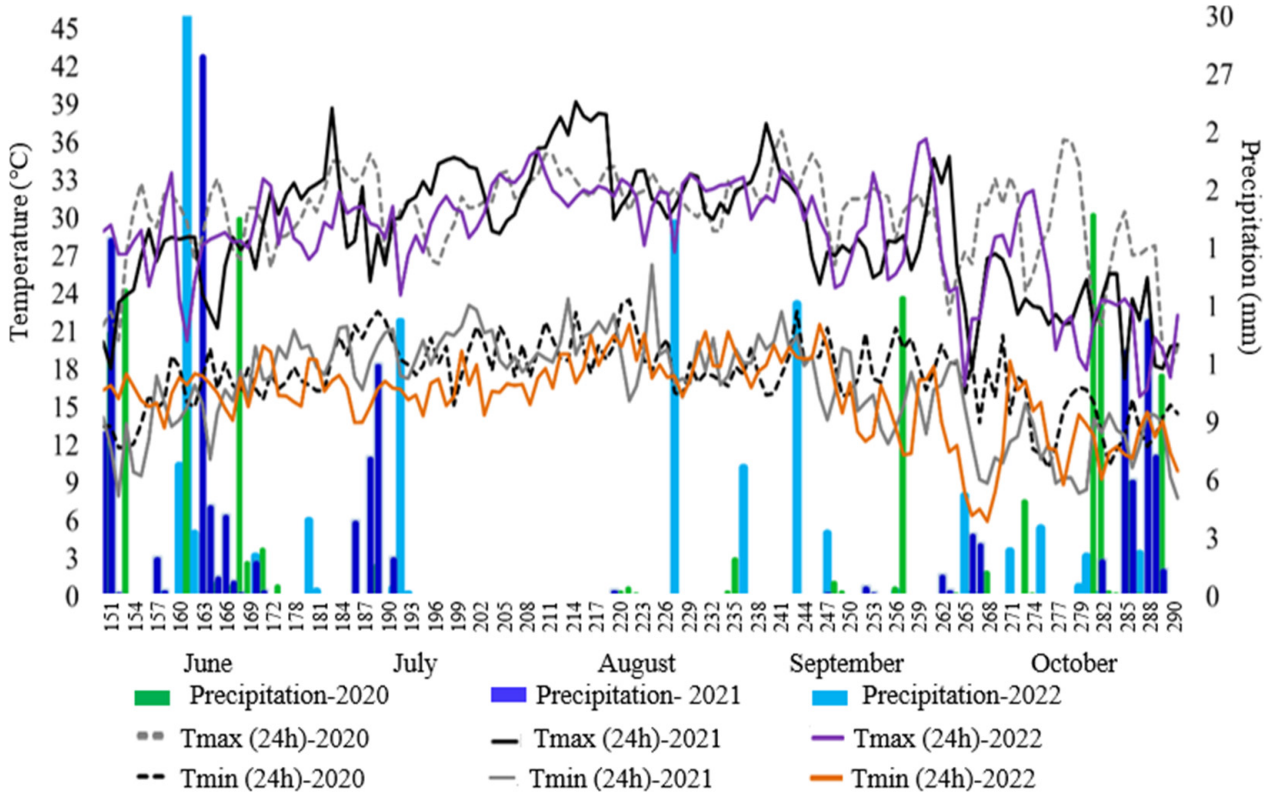

Figure 1. The temperature (max and min) and precipitation from the receptivity period to the harvest period of "Bursa Siyahı" for 2020–2022.

2.2. Flowering Phenology

The phenological observation stages of male and female fig fruits were recorded according to Valdeyron and Lloyd [32] and BBCH (The Biologische Bundesanstalt, Bundessortenamt und Chemische Industrie) scale, respectively [33]. The branches of female and male trees were marked in 3 replicates, with three branches in each replication. The period during which the receptive phase occurred was characterized by a fruit diameter of 11–13 mm, and the conclusion of receptivity was recorded in female fig trees [34]. The male fig trees' receptive period was determined by the exit of *B. psenes* from the fruits; this indicates the pollen maturity stage (Stage D) (Figure 2).

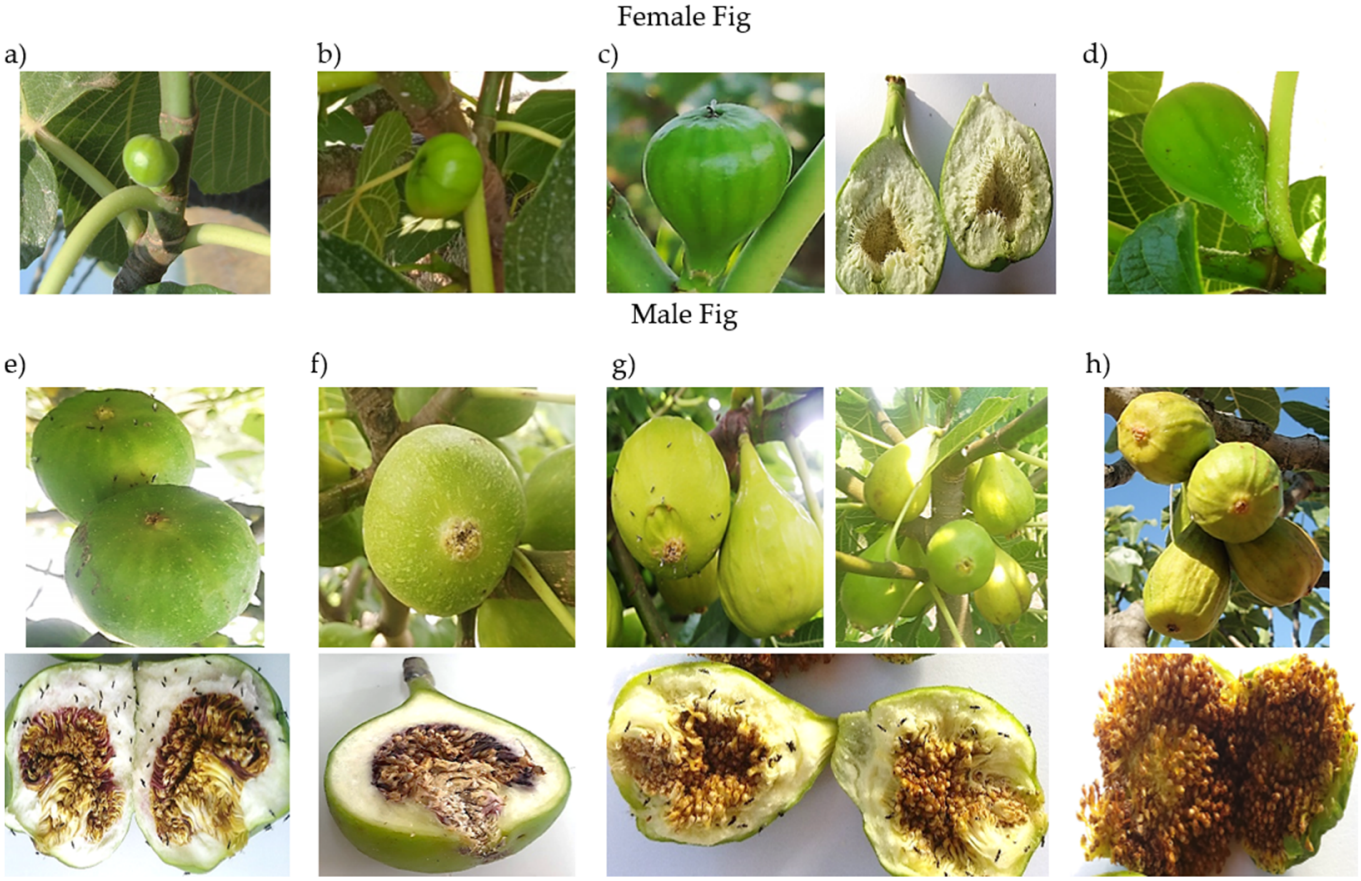

Figure 2. Illustrations of flower development of female and male fig. (**a**,**b**) beginning of female flower receptivity, (**c**) female flower receptivity, (**d**) end of female flower receptivity, (**e**–**g**) receptivity (D stage), and (**h**) end of receptivity (E stage) on male fig.

This stage ends with the yellowing and abscission of the fruits (Stage E). The duration between the transition from stage D to stage E of fruit development determined the ripening period of the genotypes.

2.3. Preparation Storage Conditions

The caprifig fruits of the 16 08 05, 16 08 09, 16 08 10, 16 09 10, and 16 ZF 08 genotypes were harvested when *B. psenes* wasps emerged from the caprifig fruit and the pollen matured in the early morning. These fruits were then stored at 4 ± 0.5 °C under 85–90% humidity for 16 days. The samples were analyzed on the 8th and 16th days of storage, while the caprifig fruits from the control group (0 days) were analyzed immediately after harvest [29,35].

2.4. Number of *B. psenes* and *P. caricae* and Duration of *B. psenes* Exit

To determine the number of *Blastophaga psenes* and *Philotrypesis caricae* in caprifig fruits stored at 4 °C every 8 days, one fruit was placed in a jar (8 × 10 cm) with four replications, and the *B. psenes* emerging from the caprifig fruit over several days were counted. For the control group (0 days), these procedures were conducted in the morning before the wasps emerged on the day of harvest. The mouths of the jars were sealed with mesh to prevent wasps from escaping, and the jars were kept in climate-controlled rooms at 26 °C under a 16 h light/8 h dark cycle [29]. The first exiting wasp was transferred to bottles containing 70% alcohol, and the first and last exit days were recorded to determine the duration of *B. psenes* exit.

2.5. Pollen Viability, Pollen Germination, and Pollen Size

On the 8th and 16th days of storage, as well as the harvest day (0th day), fifteen caprifig fruits were brought to room temperature and longitudinally sliced open. Following this, gentle tapping of the male flowers on the fruit with a glass rod ensured that the anthers remained on the mesh attached to Petri dishes, allowing for pollen to pass through into the dishes [36]. Immediately upon obtaining the pollen, viability and germination tests were conducted. Pollen viability was assessed using 2,3,5-triphenyltetrazolium chloride (TTC) staining, where a bright red color indicated viability and a lack of color indicated nonviability (×40; Leica DC 500, Leica, Germany) [37]. To determine in vitro pollen germination, the Petri dish technique was used [37]. Pollen grains were placed on culture media and then incubated in the dark at 25 °C for 24 h. The culture media comprised 1% agar, 5% sucrose, and 5 ppm H_3BO_3 at pH 5.0 [38]. Pollen germination was evaluated in two random fields of two Petri dishes per treatment, considering that germination occurred when the pollen tube length equaled or exceeded the pollen grain diameter. The pollen sizes of the caprifig genotypes were determined using a Leica DC 500 light microscope, with 15 pollen grains per three replicates examined (×40). The Leica package program was utilized to select pollen, and the equatorial diameter (µm) and polar length (µm) values were recorded.

2.6. Pollination Treatments

Syconia with a diameter of 8–10 mm in the leaf axils were isolated approximately ten days prior to caprification [34]. Before reaching receptivity, uniform branches of "Bursa Siyahı", each about 50 cm long, were carefully chosen. The experiment was established in randomized blocks with three replications, with different genotypes on each tree, one tree in each replication, and four branches were selected from each tree. These branches were then covered with mesh to ensure isolation. Subsequently, one caprifig fruit from each genotype was placed within the isolation mesh, and caprification was conducted three times at 8-day intervals. The caprification process was initiated during the early hours of the morning. After the caprification period, which lasted approximately three weeks, the isolation mesh was removed from each branch.

Fruit Ripening Stage, Fruit Set, and Fruit Characteristics

The fruit ripening stage was determined based on the fruit's skin color change. The ripe fruits were harvested from September to November at 4-day intervals. The fruits were evaluated in four groups based on their ripening times: before 15 September, 15 September–1 October, 1–15 October, and 15 October–1 November. To determine the fruit set rate, the fruits on marked branches were counted during caprification and before harvest. In terms of pomological characteristics, ten fruits were randomly collected from each replicate, and measurements were taken for fruit weight (g), ostiole diameter (mm), flesh thickness (mm), and fruit cavity. The fruits were categorized into four groups based on their weights: 40–60, 60–80, 80–100, and more than 100 g. Ostiole damage was assessed based on the quantity of observed fruit skin damage, using the European grading criteria with four categories: none (no damage), slight damage (ostiole-end splitting covering

less than one-third of the fruit), moderate damage (ostiole-end splitting covering between one-third and two-thirds of the fruit), and severe damage (ostiole-end splitting covering more than two-thirds of the fruit) [39]. The average number of seeds was determined by counting both fertile and sterile seeds in four fruits per caprifig genotype. The ripe fruits were submerged in water to separate the fermented flesh from the seeds. Subsequently, the dried seeds were again immersed in water, where floating seeds were identified as sterile and those at the bottom were identified as fertile [36]. The occurrence of fig endosepsis (internal rot) was assessed based on the presence or absence of damage and expressed as a percentage. The soluble solids content (SSC) (°Brix) was measured using a digital refractometer (PR-101 ATAGO, Norfolk, VA, USA), while the titratable acidity (TA) (g citric acid/100 mL) was determined through the titration of fig juice with 0.1 M NaOH. Fruit skin and flesh colors, in terms of lightness (L), hue (H°), and chroma (C) values, were evaluated using a colorimeter (Chroma Meter CR-400, Minolta, Osaka, Japan).

2.7. Statistical Analysis

The data obtained in the study were subjected to two-factor analysis of variance (ANOVA). The ANOVA included the factors of genotypes (16 08 05, 16 08 09, 16 08 10, 16 09 10, and 16 ZF 08) and storage time (0, 8, and 16 days). Furthermore, the effects of the caprifig genotype on fruit set and fruit quality were analyzed. The mean values were compared using Duncan's multiple range test at $p < 0.05$ and $p < 0.01$. Arcsine root square transformation was applied for binomial data (pollen viability, germination, fruit set, fruit ripening stage, proportion of fruit weight, and ostiole damage). ANOVA was performed using SPSS version 22.0 for Windows (SPSS Inc., Chicago, IL, USA). Principal component analysis (PCA) was performed using the varimax factor rotation method, and biplots were constructed with two principal components displaying the caprifig genotypes and fruit traits. A structural equation model (SEM) was used to elucidate the impact of pollen source traits on fruit set, ripening time, and fruit characteristics. The model was fitted using the maximum likelihood estimation method, and its adequacy was assessed through the χ^2 test and root square mean error of approximation (RMSEA). A nonsignificant χ^2 test ($p < 0.05$) and a lower RMSEA ($p > 0.05$) indicated a satisfactory model fit. The SEM analysis proceeded through several steps: initially, the model was established and checked for its structural fit and the significance of the regression coefficients. Subsequently, iterative adjustments were made until the model fit fell within acceptable standards, with a preference for smaller RMSEA values. Finally, the model results were meticulously analyzed and interpreted. Multivariate analyses were conducted using JMP® Pro 17.0.0 (Copyright © 2023 SAS Institute Inc., Cary, NC, USA), with significance set at $p < 0.05$ and $p < 0.01$.

3. Results and Discussion

3.1. Flowering Phenology

According to the phenological data, the receptive period of "Bursa Siyahı" fruits (173, 174, and 177 JD) started before the male fig genotypes became receptive. In 2020, high temperatures (155, 161, and 166 JD) led to an earlier onset of receptivity in "Bursa Siyahı" fruits (Figures 1 and 3). However, in 2021, due to high temperatures during the receptive period (178, 180, and 184 JD), both the "Bursa Siyahı" cultivar (21 days) and male fig genotypes (8 days) experienced a shorter period of receptivity. Conversely, in 2022, relatively lower temperatures during these periods resulted in delayed and longer duration of receptivity. Moreover, 16 ZF 08 (181, 181, and 182 JD) and 16 08 09 (183, 181, and 180 JD) were recorded as the first genotypes to start being receptive approximately one week after the "Bursa Siyahı" fruits in all years. Furthermore, 16 09 10 (183, 184, and 182 JD) and 16 08 05 (183, 185, and 182 JD) followed these genotypes, with slight variations of 1–2 days. As for 16 08 10, it stood out as the latest ripening genotype (186, 187, and 187 JD) among the genotypes, with a more extended receptivity period (13, 11, and 14 days), and it closed the receptive period of "Bursa Siyahı", in contrast to the other genotypes (Figure 3).

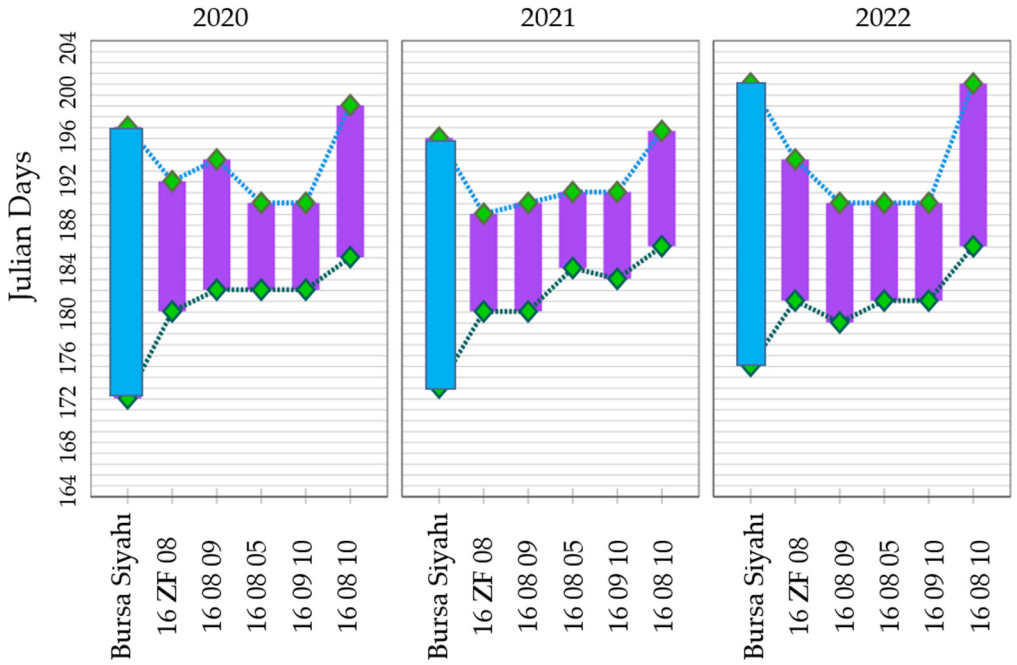

Figure 3. Receptivity period of the "Bursa Siyahı" female fig cultivar (blue bars) and male fig genotype (purple bar). Green dots show the difference in time when receptivity begins for varieties and genotypes, and blue dots show the difference in time when receptivity ends. Green rhombuses indicate the time when receptivity of the cultivar and genotypes begins and ends.

3.2. Number of B. psenes and P. caricae and Duration of B. psenes's Exit

The effects of storage time, genotype, and the interaction between genotype and storage time on the number of *B. psenes* were significant in all years (Table 1, Figure 4, Table S1).

Table 1. Effect of caprifig genotype and storage time on the number of *B. psenes* and *P. caricae* and the duration of *B. psenes*'s exit.

Genotype (G)	Number of *B. psenes* (Number/Fruit)			Duration of *B. psenes*'s Exit (Days)			Number of *P. caricae* (Number/Fruit)		
	2020	**2021**	**2022**	**2020**	**2021**	**2022**	**2020**	**2021**	**2022**
16 08 05	293.00 a	146.55 a	183.55 bc	3.44 a	2.88 a	3.66 a	1.22 b	9.11 a	2.00 b
16 08 09	186.22 bc	86.44 b	264.66 a	2.33 c	1.44 c	2.88 bc	2.22 ab	1.55 c	2.00 b
16 08 10	155.66 c	80.88 b	148.33 c	2.77 b	2.00 b	2.77 c	2.22 ab	2.11 c	1.77 b
16 09 10	287.00 a	98.55 b	168.00 bc	3.22 a	2.00 b	3.22 b	2.77 a	3.88 b	1.44 b
16 ZF 08	217.33 b	109.88 ab	204.66 b	3.11 ab	1.88bc	3.00 bc	2.22 ab	4.11 b	3.88 a
F-value	18.02	5.63	7.01	10.53	10.69	6.54	3.77	23.78	4.36
p-value	<0.01 **	0.01 *	<0.01 **	<0.01 **	<0.01 **	<0.01 **	0.04 *	<0.01 **	<0.01 **
Storage time (ST) (days)									
0	304.66 a	191.93 a	293.80 a	3.80 a	3.20 a	3.86 a	2.33 a	5.13 a	2.40
8	218.80 b	94.40 b	181.40 b	2.73 b	2.06 b	3.13 b	1.33 b	4.26 ab	2.13
16	160.06 c	27.13 c	106.33 c	2.40 c	0.86 c	2.33 c	2.73 a	3.06 b	2.13
F-value	42.95	61.31	52.15	50.03	88.12	51.93	7.58	4.80	0.18
p-value	<0.01 **	<0.01 **	<0.01 **	<0.01 **	<0.01 **	<0.01 **	<0.01 **	0.01 *	0.83 ns

Small letters in the same column indicate a significant difference according to Duncan test. * and ** within a column indicate a significant difference at the $p < 0.05$ and $p < 0.01$ level. (ns: not significant).

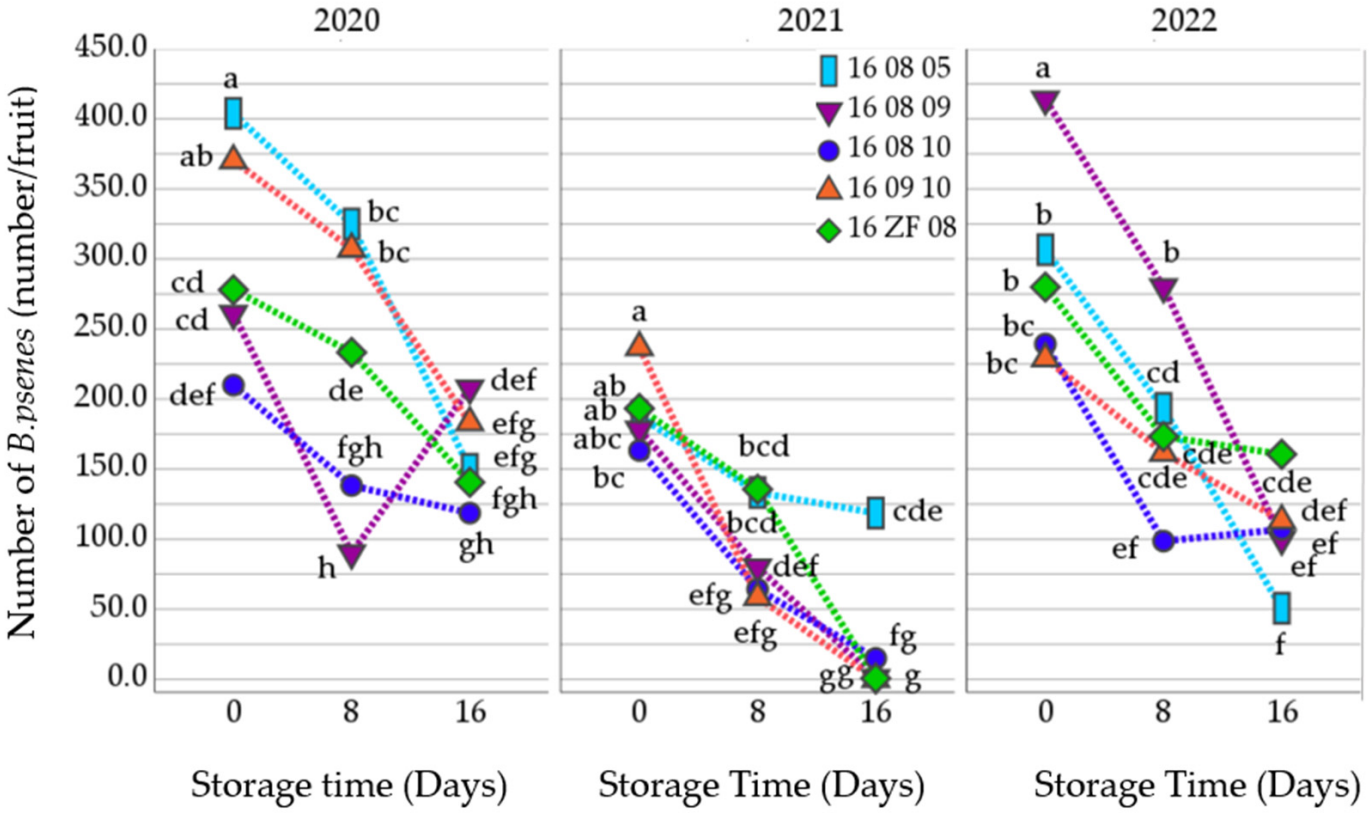

Figure 4. Number of *B. psenes* of caprifig genotypes depending on storage time. Small letters indicate a significant difference according to the Duncan test.

With respect to the storage time, the number of *B. psenes* in the caprifig fruits decreased linearly, but in 2020, the number of *B. psenes* in the caprifig fruits was lower after 8 days of storage (90.0 number/fruit) than after 16 days (207.6 number/fruit) for the 16 08 09 genotype. This difference was related to the variation in fruit size among the sampled fruits. The storage of caprifigs decreased the number of *B. psenes*, with a significant decline, especially at the beginning of 2021, in the initially low *B. psenes* population.

On the 16th day of storage, the percentage of wasps of each genotype decreased by approximately 90–100%. In 2021, due to a decrease in temperature to −10.4 °C, the larvae of *B. psenes* living on mamme fruits were damaged, leading to a reduced transition of *B. psenes* to caprifig fruits. According to the genotypes, in 2020 and 2021, the 16 08 05 (293.0; 146.5 number/fruit) genotype exhibited high *B. psenes* values, whereas in 2022, the 16 08 09 (264.6 number/fruit) genotype yielded high values (Table 1).

The highest number of *B. psenes* was observed on the harvest day for the 16 08 05 (404.0 number/fruit) and 16 09 10 (370.2 number/fruit) genotypes in 2020 and for the 16 09 10 (237.0 number/fruit) and 16 08 09 (414.0 number/fruit) genotypes in 2021 and 2022, respectively (Figure 4, Table S1). Zare et al. [30] reported that the "Shanehi", "Gohari", "Kouhi", and "Poozdombali" caprifig cultivars could be stored at 4 °C for 14, 18, 22, and 32 days. Anjam et al. [29] reported that the percentage of *B. psenes* in the "Poozdombali" caprifig cultivar decreased by 68.74% after 7 days, 86.61% after 14 days, and 90.33% after 21 days. Compared to Anjam et al. [29], average research data showed that less *B. psenes* loss occurred after 16 days of storage (61.03–67.00%).

The duration of *B. psenes'* exit was affected by storage time, genotype, and storage time and genotype interaction in all years (Table 1, Figure 5, Table S1). The lowest duration of *B. psenes* exit was observed at 16 days of storage in all years. The highest duration of *B. psenes* exit was obtained for the 16 08 05 (3.4; 2.8; 3.6 days) genotype. According to the classification by Eroğlu [40], on the day of harvest, the duration of *B. psenes* exit in the 16 08 05 (4.6; 3.6; 5.0 days) genotype was intermediate, while that in the other genotypes was short in all years. Depending on storage, the duration of *B. psenes* exit was short for all the genotypes. Yaman and Çalışkan [41] and Çalışkan et al. [9] reported that the duration of *B. psenes* exit varied between 3 and 7 days on the day of harvest. The results of the present study are similar to those of Yaman and Çalışkan [41] except for the 16 08 10 genotype (2.6 days) in 2021.

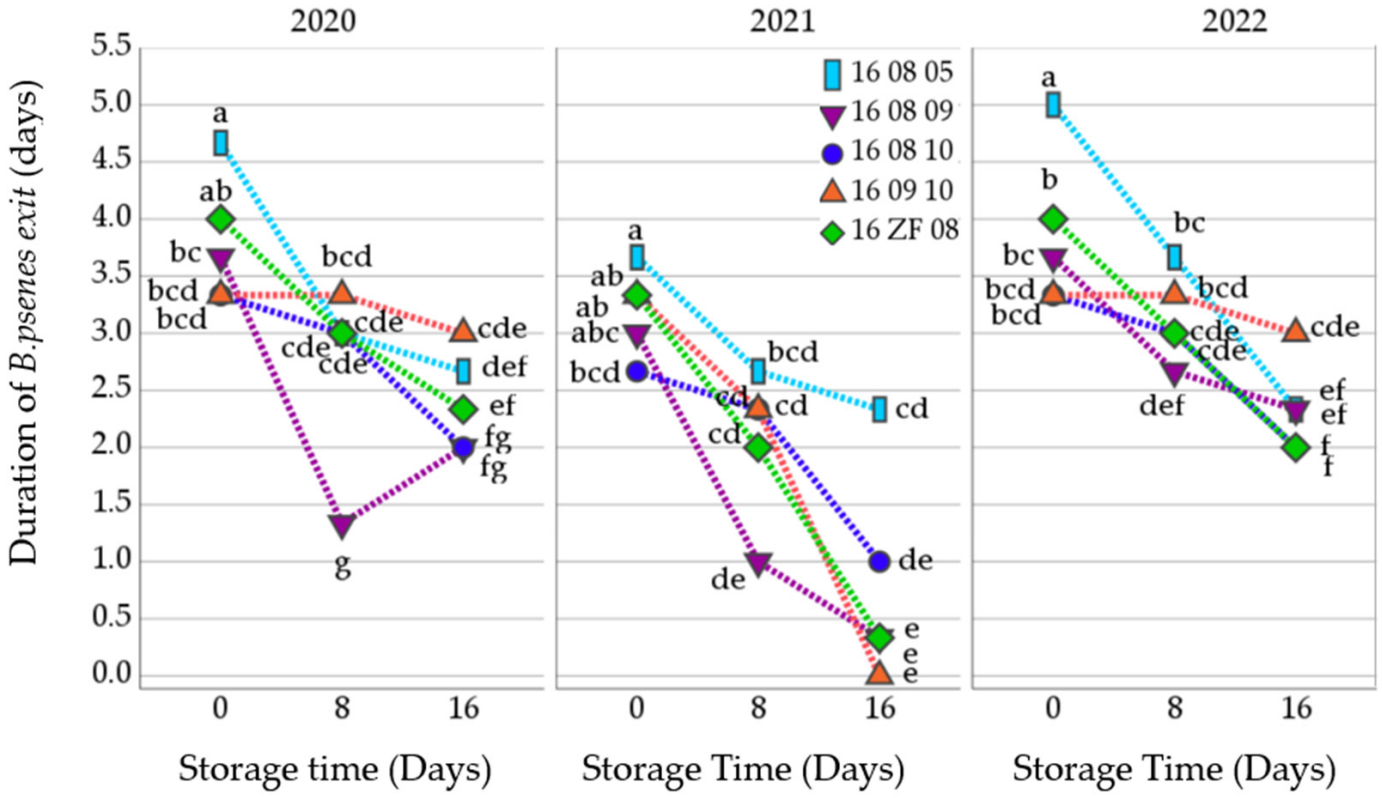

Figure 5. Duration *B. psenes* of caprifig genotypes depending on storage time. Small letters indicate a significant difference according to the Duncan test, $p < 0.05$.

The number of *P. caricae* was significant in 2020 and 2021, depending on the storage time (Table 1). In 2020, the highest number of *P. caricae* was counted on the harvest day (2.11 number/fruit) and 16 days of storage (2.66 number/fruit). In 2021, it was recorded on the harvest day (4.77 number/fruit) and at 8 days of storage (4.61 number/fruit) (Figure 6, Table S1). The highest number of *P. caricae* has been obtained from a different genotype each year. The effect of genotype and storage time interaction was significant in all years. In 2020, the highest number of *P. caricae* was obtained at 16 days of storage (4.60 number/fruit) for the 16 08 10 genotype and on the day of harvest for the 16 09 10 (4.60 number/fruit) genotype. In 2021 and 2022, it was obtained by storing the 16 08 05 (10.60 number/fruit) and 16 ZF 08 (8.20 number /fruit) genotypes for 16 days.

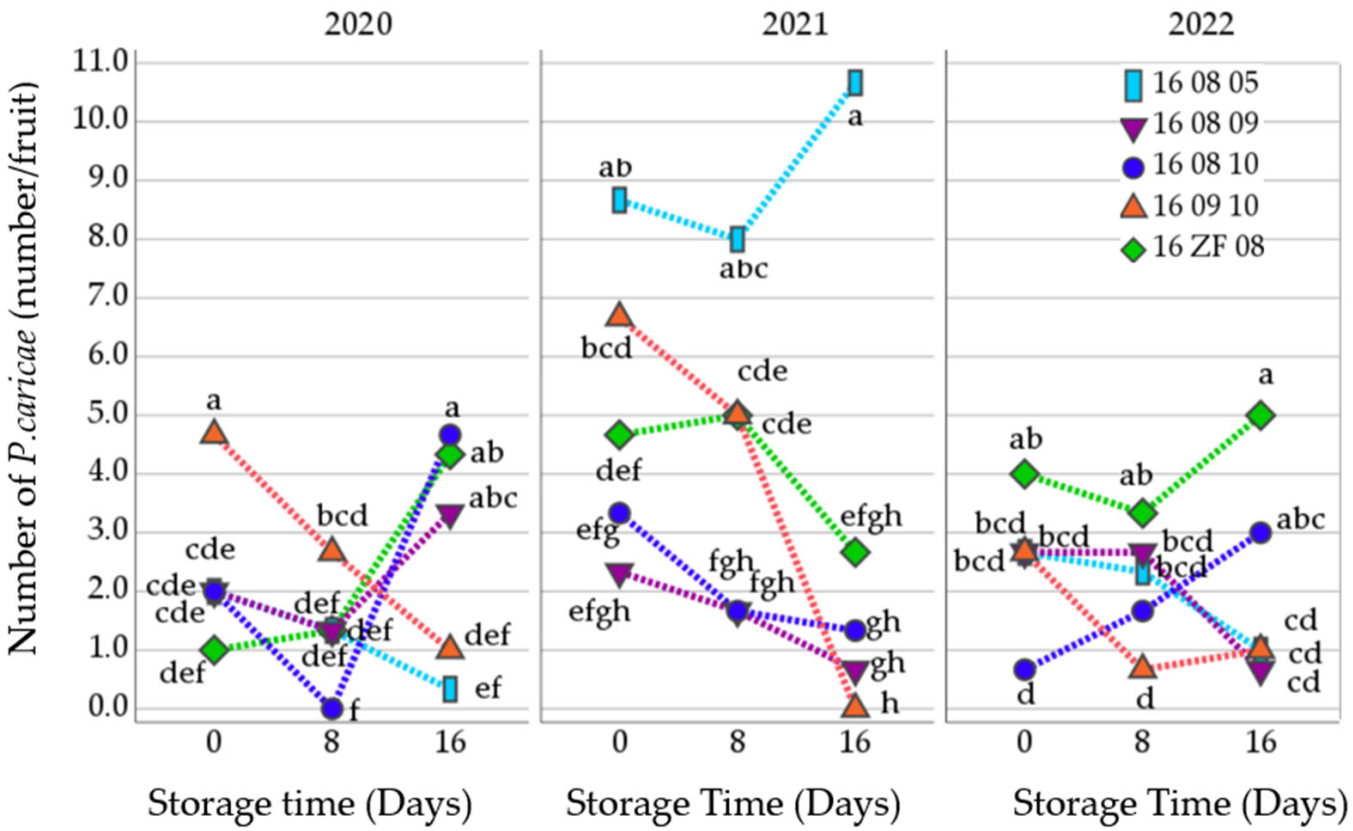

Figure 6. Number of *P. caricae* of caprifig genotypes depending on storage time. Small letters indicate a significant difference according to the Duncan test, $p < 0.05$.

Yaman [42] reported that the number of *P. caricae* varied between 0.0 and 18.0 number/fruit. Caliskan et al. [9] reported that only one genotype was observed, while Ahi Koşar et al. [43] reported that it varied between 0.13 and 6.36 number/fruit. In the studies by Yaman [42] and Ahi Koşar et al. [43], similar *P. caricae* values were obtained, while Çalışkan et al. [9] obtained higher *P. caricae* values. Depending on the years, the highest number of *P. caricae* was obtained in 2021, unlike the number of *B. psenes*. It is known that *F. carica*'s pollinator wasp. *B. psenes* is damaged at low temperatures, whereas the tolerance of *P. caricae* to low temperatures is unknown. Chen et al. [44] reported that the population of *F. racemosa* fig pollinators decreased significantly at low temperatures in China, and the temperature tolerance of parasitoids reportedly increased. Since the highest *P. caricae* values were obtained in 2021 and the number of parasitoids did not decrease linearly with storage, *P. caricae* may be tolerant to low temperatures.

In addition, since *B. psenes* larvae and *P. caricae* cleptoparasitic larvae compete [10], the low number of *B. psenes* in 2021 may have caused competition to end, in favor of *P. caricae*.

3.3. Pollen Viability, Pollen Germination, and Pollen Size

Pollen viability was affected by storage time and genotype in all years (Table 2). While pollen viability increased as the storage time increased in 2020, it decreased in other years. However, there was no linear decrease or increase in pollen viability depending on the storage time. Since ripe caprifig fruits were stored randomly, whether the anthers inside the stored fruits burst before or continue to burst during storage was unknown.

Table 2. Effects of the caprifig genotype and storage time on pollen viability, germination, equatorial diameter, and polar length.

Genotype (G)	Pollen Viability (%)			Pollen Germination (%)			Equatorial Diameter (µm)			Polar Length (µm)		
	2020	2021	2022	2020	2021	2022	2020	2021	2022	2020	2021	2022
16 08 05	75.0 b	80.6 a	88.9 a	26.3 c	46.1 b	45.3 a	9.64 b	9.42 d	9.98 b	9.36 b	9.31 b	9.56 bc
16 08 09	76.9 ab	82.4 a	87.1 a	38.9 a	44.3 b	40.6 b	10.44 a	10.34 ab	10.39 a	9.70 ab	9.56 b	9.98 a
16 08 10	79.6 ab	83.3 a	82.4 b	26.0 c	35.9 c	23.9 d	9.87 b	9.94 bc	9.74 b	9.59 ab	9.49 b	9.43 bc
16 09 10	81.2 a	84.1 a	86.1 a	33.7 b	53.9 a	39.3 b	10.30 a	10.43 a	10.38 a	10.07 a	10.12 a	10.11 a
16 ZF 08	67.2 c	65.0 b	89.3 a	28.2 c	33.2 c	32.2 c	9.90 b	9.79 cd	9.78 b	9.10 b	9.48 b	9.30 c
F-value	10.96	48.76	6.54	24.50	23.63	43.85	11.82	13.23	25.24	5.29	24.28	21.35
p-value	<0.01 **	<0.01 **	0.02 *	<0.01 **	<0.01 **	<0.01 **	<0.01 **	<0.01 **	<0.01 **	<0.01 **	<0.01 **	<0.01 **
Storage time (ST) (days)												
0	74.2 b	85.6 a	88.1 a	28.9	41.7 b	30.4 b	10.04	9.98	10.06	9.62	9.60	9.70
8	75.1 ab	73.1 c	88.6 a	31.4	40.4 b	40.1 a	10.04	9.95	10.04	9.41	9.61	9.68
16	78.6 a	78.4 b	84.2 b	31.6	45.9 a	38.6 a	10.01	9.99	10.03	9.66	9.56	9.63
F-value	5.23	51.69	11.31	2.80	4.73	27.10	0.94	0.72	0.62	1.23	0.87	0.65
p-value	0.04 *	<0.01 **	<0.01 **	0.07 ns	0.01 *	<0.01 **	0.06 ns	0.99 ns	0.71 ns	0.30 ns	0.75 ns	0.86 ns

Small letters in the same column indicate a significant difference according to Duncan test. * and ** within a column indicate a significant difference at the $p < 0.05$ and $p < 0.01$ level. (ns: not significant).

Consistent with the present study, Gaudet et al. [45] reported that the most suitable phenological period for the highest pollen viability in *Cannabis sativa* is the middle of the flowering period, while pollen is still present in the anthers. The highest pollen viability was obtained for the 16 08 09 (76.9%; 82.4%; 87.1%) and 16 09 10 (81.2%; 84.1%; 86.1%) genotypes in all years. Pollen viability was affected by the interaction between genotype and storage time in 2020 and 2021. The viability test performed on the harvest day with the 16 09 10 (87.4%; 94.0%) genotype gave high values, whereas the lowest viability was obtained on the harvest day (62.8%; 70.4%) and at 8 days of storage (59.8%; 60.3%) with the 16 ZF 08 genotype (Figure 7, Table S2). Dafni and Firmage [46] and Gaaliche et al. [38] reported that pollen can be considered functional if pollen viability is above 50%. According to the findings, viability values of the genotypes were consistently over 50% in all years, depending on the harvest day and storage conditions.

Pollen germination was affected by both storage time and genotype in 2021 and 2022 (Table 2). Pollen germination tended to increase with the storage of caprifig fruits. However, similar to the pollen viability results, the increase was not linear. Similarly, Yuan et al. [47] stated that the germination of *Phalaenopsis* pollen increases proportionally with the progression of the flower's phenological development period. Rosell et al. [48] reported that cherimoya pollen showed the highest germination rate during the bursting of anthers. Wang et al. [49] reported that, in litchi, pollen grains are more abundant in longer filaments before anther bursts. Other researchers have also suggested that pollen germination is influenced by anther bursts and pollen age [50,51].

Pollen germination was affected by genotype and storage time interaction in all years (Figure 8, Table S2). In 2020, the lowest pollen germination was recorded on the harvest day (17.91%) for the 16 08 05 genotype. In 2021 and 2022, the lowest values were observed after 8 days (19.53%) and 16 days (20.48%) of storage for the 16 08 10 genotype (Figure 8).

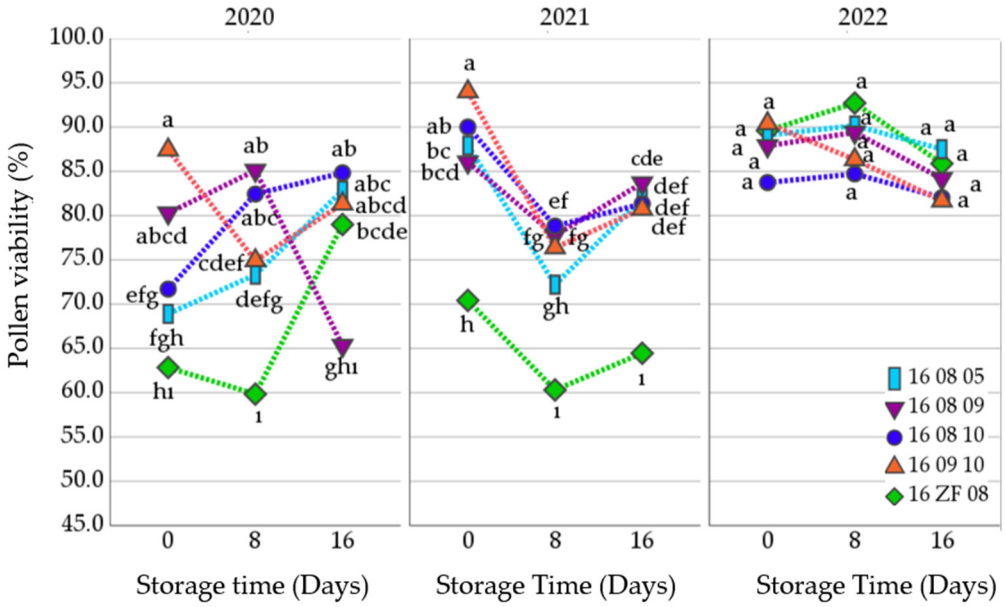

Figure 7. Pollen viability of caprifig genotypes depending on storage time. Small letters indicate a significant difference according to the Duncan test, $p < 0.05$.

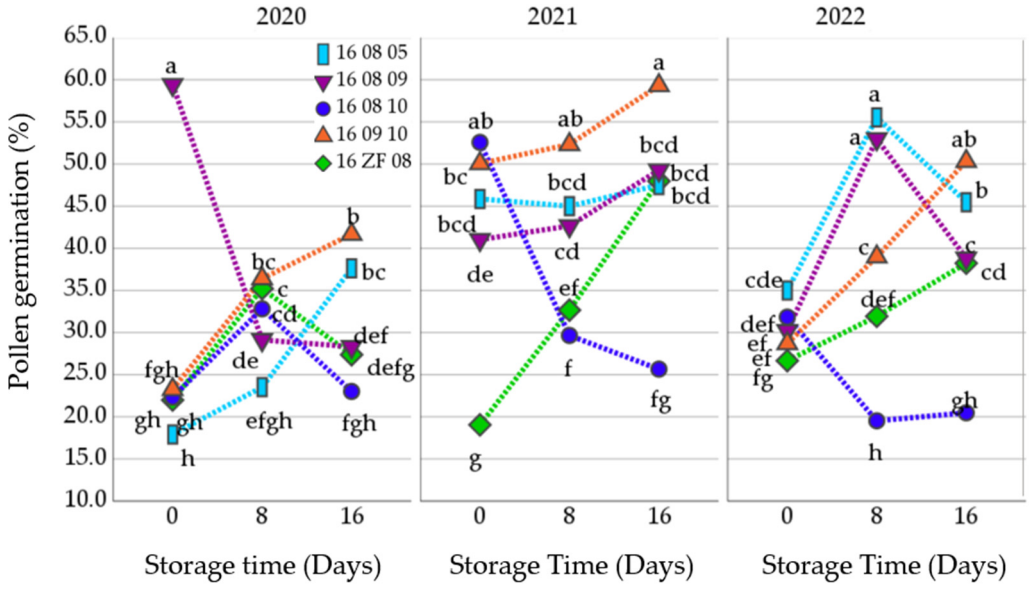

Figure 8. Pollen germination of caprifig genotypes depending on storage time. Small letters indicate a significant difference according to the Duncan test, $p < 0.05$.

Gaaliche et al. [38] suggested that pollen can be functional if germination is above 30%. In the present study, the pollen germination of the 16 ZF 08 genotype on the day of harvest and of the 16 08 10 genotype after 8 and 16 days of storage was less than 30% in all years. Various researchers have reported pollen germination ranging from 2.0% to 43.5% [52], 0.0% to 96.6% [53], 68.32% to 74.08% [8], and 0.0% to 40.74% [42]. In this study, the pollen germination rate was lower than that obtained by Ilgın et al. [8] but close to that obtained by other researchers [42,52,53]. The pollen equatorial diameter and pollen length were affected by only genotype. The highest pollen equatorial diameter and pollen length values were obtained from the 16 08 09 and 16 09 10 genotypes (Table 2). Acarsoy Bilgin et al. [54] reported that the pollen length of caprifig genotypes ranged from 9.60 µm to 11.25 µm, while the pollen equatorial diameter ranged from 11.83 µm to 13.34 µm. Çalışkan et al. [55] determined that the pollen length and equatorial diameter ranged from 9.99 µm to 12.24 µm

and from 8.52 µm to 11.90 µm, respectively. In the present study, the pollen sizes of the genotypes were smaller than those reported by Acarsoy Bilgin et al. [54] but close to those reported by Çalışkan et al. [55].

3.4. Fruit Ripening Stage, Fruit Set, and Fruit Characteristics

Based on three years of data, when the 16 08 05 (39.5%; 30.7%; 20.4%) genotype was used as a pollen source, the rate of ripening of "Bursa Siyahı" fruits before 15 September was higher than that of the other genotypes. The rate of ripening between 15 September and 1 October was high for all the genotypes, although it differed slightly from year to year. The rate of fruit ripening between 15 October and 1 November was significantly higher when the 16 08 10 (35.7%; 37.3%) genotype was used as a pollen source from 2021–2022. The 16 ZF 08 (27.2%; 23.3%) genotype also had a relatively high percentage of fruits during this period (Table 3). The presence of genotypes that may influence the early or late harvest of "Bursa Siyahı" figs is crucial, as it can extend the harvest period of this high-value-added product and keep it on the market for a more extended period. Similarly, Zare [4] reported that "Daneh Sefid" caprifig pollens contribute to early ripening, while "Shak Anjiri" pollens play a crucial role in the early ripening process.

Table 3. Effects of caprifig genotype on the ripening period of "Bursa Siyahı" fruits.

Ripening Period	Genotype						
	16 08 05	**16 08 09**	**16 08 10**	**16 09 10**	**16 ZF 08**	**F Value**	***p*-Value**
				2020			
<15 September	39.50 a	20.30 b	8.16 c	41.22 a	28.53 ab	106.00	<0.01 **
15 September–1 October	28.50 cd	36.28 b	47.26 a	32.77 bc	22.00 d	71.21	<0.01 **
1 October–15 October	20.50 ab	15.20 b	27.57 a	15.92 b	22.07 a	29.84	<0.01 **
15 October–1 November	12.50 c	28.00 a	16.32 b	7.00 d	18.00 b	30.21	<0.01 **
				2021			
<15 September	30.79 a	23.73 ab	11.09 c	20.16 bc	17.40 bc	10.46	<0.01 **
15 September–1 October	46.02	36.76	40.68	42.50	42.42	0.70	0.61 ns
1 October–15 October	21.22 b	24.92 ab	10.54 c	26.99 a	12.94 c	10.89	<0.01 **
15 October–1 November	3.95 c	14.56 bc	35.72 a	13.33 bc	27.20 ab	14.19	<0.01 **
				2022			
<15 September	20.47 ab	24.20 a	16.00 b	18.64 b	25.31 a	4.48	0.03 *
15 September–1 October	40.72 b	38.91 bc	28.69 d	47.22 a	36.06 c	27.30	<0.01 **
1 October–15 October	26.14	14.39	17.45	16.44	15.24	3.74	0.05 ns
15 October–1 November	12.33 b	22.15 ab	37.37 a	19.13 b	23.37 ab	9.22	<0.01 **

Small letters in the same column indicate a significant difference according to Duncan test. * and ** within a column indicate a significant difference at the $p < 0.05$ and $p < 0.01$ level. (ns: not significant).

Al-Khalifah [56] reported that date pollen affects fruit quality and ripening time due to its metaxenia properties. Shahsavar and Shahhosseini [57] reported that some date pollen sources contain relatively high amounts of hormones such as auxin and gibberellins. These hormones are known to be triggered by pollen during different fruit growth and development periods.

The fruit set of the "Bursa Siyahı" cultivar varied depending on genotype. Three years of data showed that a higher fruit set was obtained when the 16 08 09 (81.0%; 76.9%; 85.4%) and 16 09 10 (75.0%; 69.3%; 81.9%) genotypes were used as pollen sources (Table 4). However, caprification performed with the 16 08 10 (66.0%; 61.9; 67.0%) genotype, which exhibited lower pollen germination and a higher number of *B. psenes*, resulted in a lower fruit set. Similarly, a reduced fruit set was observed using the 16 ZF 08 (65.6%; 59.3%) genotype, which showed lower viability and germination rate as pollinators in 2020 and 2021. Deng et al. [58] reported that pollen sources influence fruit sets, and high pollen germination increases the pollen uptake rate of the stigma, ensuring adequate pollination. In the present study, consistent with this, pollen viability and germination of genotypes with high fruit sets were consistently higher in all years. Westwood [59] emphasized that the fruit set should be at least 70% for an adequate fig yield. In this study, the fruit set of the "Bursa Siyahı" cultivar pollinated with the 16 08 10 genotype was less than 70% in all years. Similarly, Zare [4] reported fewer fruit drops to occur when the "Daneh-Sefid" genotype is used in caprification. Marcotuli et al. [60] stated that caprifig genotypes exhibit different characteristics and highlighted that pollination may affect fruit set, yield, and quality.

Table 4. Effects of caprifig genotype on the fruit set and fruit characteristics of the "Bursa Siyahı" cultivar.

Genotype	Fruit Set (%)			Fruit Weight (g)			Ostiole Diameter (mm)			Flesh Thickness (mm)			Fruit Cavity (mm)		
	2020	2021	2022	2020	2021	2022	2020	2021	2022	2020	2021	2022	2020	2021	2022
16 08 05	65.0 b	75.3 a	78.0 ab	75.2 b	81.7 ab	81.6 ab	5.1 ab	5.6 ab	8.1 a	19.4	20.9 a	20.7 b	4.9 b	4.9 a	5.9 ab
16 08 09	81.0 a	76.9 a	85.4 a	86.7 ab	85.3 a	82.1 ab	7.3 a	6.0 ab	7.1 ab	21.9	20.9 a	21.8 ab	1.8 c	4.0 ab	6.9 ab
16 08 10	66.0 b	61.9 ab	67.0 b	83.8 ab	75.5 ab	73.2 b	7.3 a	3.5 b	6.6 b	20.6	19.3 c	21.0 ab	1.1 c	2.1 b	4.0 b
16 09 10	75.0 ab	69.3 ab	81.9 a	91.3 a	84.7 a	80.5 ab	5.3 ab	7.5 a	5.7 b	20.7	20.4 ab	24.4 a	7.0 a	3.4 ab	9.5 a
16 ZF 08	65.6 b	59.3 b	78.8 ab	82.8 ab	70.8 b	77.4 b	3.9 b	3.7 b	6.5 b	19.8	19.8 bc	18.8 b	4.3 b	2.9 b	5.1 b
F-value	11.4	8.4	17.8	14.0	11.00	9.83	2.88	2.44	2.98	3.4	11.5	4.7	18.5	7.0	5.6
p-value	<0.01 **	0.01 *	0.04 *	0.03 *	<0.01 **	<0.01 *	0.01 *	<0.01 **	0.01 *	0.06 ns	<0.01 **	0.04 *	<0.01 **	0.01 *	0.01 *

Small letters in the same column indicate a significant difference according to Duncan test. * and ** within a column indicate a significant difference at the $p < 0.05$ and $p < 0.01$ level. (ns: not significant).

The fruit weight of the "Bursa Siyahı" cultivar varied depending on the genotype (Table 4). According to three years of data, when the 16 09 10 (91.3; 84.7; 80.5 g) and 16 08 09 (86.7; 85.3; 82.1 g) genotypes were used as pollen sources, the fruit weight of the "Bursa Siyahı" cultivar was increased. In 2020 and 2021, relatively higher fruit weight values were obtained when the 16 08 10 genotype (83.8; 75.5 g) used as a pollen source. This may be related to the fact that, when the 16 08 10 genotype was used as a pollen source, lower fruit set values were obtained for the "Bursa Siyahı" cultivar. Consistent with the findings of this study, Gaaliche et al. [18,19], Rahemi and Jafari [21], and Pourghayoumi et al. [5] reported that pollen sources significantly affected fruit weight. The effect of pollen sources on fruit characteristics has been reported for other fruits such as oranges [61], apples [62], blueberries [13], and plums [58].

The weight ratio of "Bursa Siyahı" fruits varied depending on the genotype used for caprification (Table 5). When the 16 ZF 08 genotype (18.3%; 40.5%; 22.0%) was used as a pollen source, the percentage of fruits between 40 and 60 g was higher in all years. When the 16 08 05 (61.2%; 40.9%) genotype was used as a pollen source, the percentage of fruits between 60 and 80 g was higher in 2020 and 2021. The proportion of fruits weighing between 80 and 100 g varied among genotypes in 2021 and 2022. In both years, the percentage of fruits was significantly higher when the 16 08 09 (34.9%; 38.4%) genotype was used as a pollen source. The proportion of fruits weighing more than 100 g was significantly higher when using the 16 09 10 (30.0%; 21.6%) genotype in 2020 and 2021 (Table 5). The size of fig fruits is an important feature that affects the quality of the domestic market and exports [63]. Although larger fruits are preferred in the domestic market for table figs, it is preferred for the exported fruits to have a weight between 60 and 80 g. When the 16 08 09 and 16 09 10 genotypes were used as pollen sources, larger fruits suitable for the domestic market were obtained, whereas the use of the 16 08 05 genotype resulted in an average of 43.47% of the fruits weighing between 60 and 80 g.

Table 5. Effect of caprifig genotype on "Bursa Siyahı" fruit's weight (%).

Fruit Weight Ratio	Genotype						
	16 08 05	**16 08 09**	**16 08 10**	**16 09 10**	**16 ZF 08**	**F-Value**	***p*-Value**
				2020			
40–60 g	7.16 b	15.62 a	17.58 a	8.67 b	18.31 a	6.43	0.01 *
60–80 g	61.25 a	27.72 b	25.75 b	26.28 b	27.37 b	25.07	<0.01 **
80–100 g	24.56	30.73	33.20	35.76	29.92	1.11	0.41 ns
>100 g	7.00 b	24.39 a	25.20 a	30.04 a	23.77 a	6.97	0.01 *
				2021			
40–60 g	11.66 b	13.81 b	23.75 b	16.11 b	40.54 a	19.61	<0.01 **
60–80 g	40.96 a	28.27 b	35.49 ab	28.39 b	39.96 a	11.46	<0.01 **
80–100 g	23.75 a	34.94 a	29.91 a	34.16 a	10.88 b	15.10	<0.01 **
>100 g	23.96 a	23.29 a	11.58 bc	21.66 ab	8.17 c	10.43	<0.01 **
				2022			
40–60 g	23.40 ab	15.56 b	37.30 a	30.60 ab	22.03 ab	4.51	0.03 *
60–80 g	28.20	32.56	27.13	31.10	30.43	0.44	0.77 ns
80–100 g	25.33 b	38.40 a	25.56 b	19.45 b	27.00 ab	6.27	<0.01 **
>100 g	23.03	13.13	10.33	18.43	20.50	2.06	0.17 ns

Small letters in the same column indicate a significant difference according to Duncan test. * and ** within a column indicate a significant difference at the $p < 0.05$ and $p < 0.01$ level. (ns: not significant).

The ostiole diameter of the "Bursa Siyahı" cultivar varied according to genotype in all years. According to three years of data, the ostiole diameter was higher when the 16 08 09 (7.3 mm; 6.0 mm; 7.1 mm) genotype was used as a pollen source (Table 4). The ostiole damage ratio of "Bursa Siyahı" fruits varied depending on the genotype (Table 6). When the 16 08 10 (44.3%; 55.5%; 21.9%) genotype was used as a pollen source, the percentage of fruits without ostiole damage was higher in all years. When the 16 09 10 (30.5%) and 16 08 09 (27.4%) genotypes were used as pollen sources, the percentage of fruits with low and moderate ostiol damage increased in 2020. Additionally, the 16 09 10 (16.6%; 25.4%) genotype caused severe ostiole damage in 2020 and 2021. The 16 08 10 (29.1%; 30.5%) genotype was the other genotype that caused severe ostiole damage in 2020 and 2022. Studies conducted in Iran and Tunisia have reported that ostiole diameter was affected by pollen source [18,19,21]. Contrary to these findings, Pourghayomi et al. [5] reported that the effect of pollen source on ostiole diameter was insignificant. The flesh thickness of the

fruits varied according to genotype in 2021 and 2022. In both years, flesh thickness was higher when the 16 08 09, 16 09 10, and 16 08 05 genotypes were used as pollen sources. Gaaliche et al. [18] and Trad et al. [20] reported that pollen sources significantly affect the flesh thickness of fruits.

Table 6. Effects of caprifig genotype on the ostiole damage ratio of "Bursa Siyahı" fruits.

Ostiole Damage Ratio	Genotype						
	16 0805	**16 08 09**	**16 08 10**	**16 09 10**	**16 ZF 08**	**F-Value**	***p*-Value**
				2020			
None	60.00 a	23.07 b	44.34 ab	44.44 ab	62.58 a	10.43	<0.01 **
Slight	9.89 b	27.04 a	15.04 ab	30.55 a	14.88 ab	7.98	<0.01 **
Moderate	17.40	15.06	11.49	8.33	13.23	3.19	0.07 ns
Severe	12.63 ab	27.72 a	29.12 a	16.66 ab	9.29 b	6.75	0.01 *
				2021			
None	48.51 ab	34.85 b	55.55 a	40.06 b	58.07 a	4.40	0.03 *
Slight	25.53	30.95	24.81	21.31	24.14	1.21	0.37 ns
Moderate	10.24	20.01	15.00	14.85	13.33	2.52	0.12 ns
Severe	15.93 b	15.10 b	4.44 c	25.49 a	9.44 bc	15.26	<0.01 **
				2022			
None	20.41 b	23.25 ab	21.90 ab	37.41 a	18.44 b	5.86	0.02 *
Slight	26.80	25.84	41.90	29.19	26.08	0.40	0.80 ns
Moderate	17.71 b	28.92 ab	24.52 ab	22.98 ab	32.60 a	4.84	0.04 *
Severe	35.07 a	21.98 ab	30.55 ab	10.03 b	22.80 ab	21.26	<0.01 **

Small letters in the same column indicate a significant difference according to Duncan test. * and ** within a column indicate a significant difference at the $p < 0.05$ and $p < 0.01$ level. (ns: not significant).

The fruit cavity was affected by genotypes in all years. Higher fruit cavities were obtained using the 16 09 10 (7.0 mm; 3.4 mm; 9.5 mm) genotype in caprification in all years (Table 4). Obtaining higher fruit cavities using the 16 09 10 genotype was related to the higher rate of fruits weighing more than 100 g. Also, when the 16 08 09 and 16 08 05 genotypes were used as pollen sources, higher fruit cavities were obtained in 2021 and 2022. The fruit cavity is closed by seed germination and seed development resulting from the fertilization of the female flowers [64]. Therefore, the fruit cavity may occur due to increasing fruit size or a lack of fertilization.

The number of fertile seeds was affected by genotype in all years (Table 7). Higher numbers of fertile seeds were obtained in all years when the 16 08 09, 16 09 10, and 16 08 05 genotypes were used for caprification. The number of sterile seeds in the fruits varied according to genotype. Higher sterile seed values were obtained when the 16 ZF 08 (150.3; 111.6; 65.0 number/fruit) genotype was used as a pollen source in all years. Seed weight was affected by genotype, and higher values were obtained when the 16 08 09 (1.6; 1.7; 1.8 mg/fruit) and 16 09 10 (1.5; 1.6; 1.7 mg/fruit) genotypes were used as pollen sources in all years (Table 7).

Table 7. Effects of caprifig genotype on the number of fertile and sterile seeds and single seed weight of "Bursa Siyahı" fruits.

Genotype	Number of Fertile Seeds (Number/Fruit)			Number of Sterile Seeds (Number/Fruit)			Single Seed Weight (mg/Fruit)		
	2020	2021	2022	2020	2021	2022	2020	2021	2022
16 08 05	1107.66 a	1004.22 a	1132.73 ab	65.33 b	67.66 b	78.66 a	1.22 c	1.60 ab	1.89 a
16 08 09	1258.33 a	1125.34 a	1205.83 a	12.00 c	25.66 c	33.66 b	1.60 a	1.70 a	1.86 a
16 08 10	1193.33 a	957.33 ab	977.33 b	14.00 c	52.00 bc	55.00 ab	1.34 bc	1.46 bc	1.46 ab
16 09 10	1073.22 a	1073.22 a	1293.33 a	18.00 c	35.66 c	55.00 ab	1.53 ab	1.66 a	1.70 a
16 ZF 08	812.66 b	702.66 b	1303.00 a	150.33a	111.66 a	65.00 a	1.41 abc	1.40 c	1.20 b
F-value	24.50	5.68	6.50	103.28	60.32	8.67	12.42	10.15	6.87
p-value	<0.01 **	0.04 *	0.04 *	<0.01 **	<0.01 **	0.03 *	0.01 *	<0.01 **	0.03 *

Small letters in the same column indicate a significant difference according to Duncan test. * and ** within a column indicate a significant difference at the $p < 0.05$ and $p < 0.01$ level).

Doi et al. [14] reported that seed weight affects fruit characteristics rather than the number of seeds in blueberries. Similarly, the pollen source affects the size of date [16], almond [65], and hazelnut [66] seeds. The fig endosepsis of fruits varied according to the genotype used for pollination in all years. A lower fig endosepsis rate was obtained when the 16 08 10, 16 09 10, and 16 08 09 genotypes were used as pollen sources, whereas a higher rate was observed when the 16 ZF 08 genotype was used. (Table 8). Michailides and Morgan [67] reported that *Fusarium* species, which trigger fig endosepsis, are carried from caprifig fruits to female fig fruits by *B. psenes*. Consistent with this, the low number of *B. psenes* that entered the caprifig fruits due to frost led to the low number of *B. psenes* that entered the female fig fruit; thus, the rate of fig endosepsis was lower in 2021. There was no significant difference between the genotypes regarding SSC in the three years; however, higher values were obtained when used as 16 08 05 and 16 08 09 genotypes in caprification (Table 8). Rahemi and Jafari [21] and Gaaliche et al. [18,19] reported that the effect of the pollen source used in caprification on SSC was significant, while Zeybekoğlu [52] reported that the pollen source did not affect the SSC of the fruit. The TEA values were greater in 2020 when the 16 09 10 (0.33 g/100 mL) and 16 08 09 (0.25 g/100 mL) genotypes were used as pollen sources (Table 8). Gaaliche et al. [19] reported that the TEA was affected by pollen sources, and using the "Djieba 1" cultivar as a pollinator increased the TEA in the fruit.

Table 8. Effects of caprifig genotype on the SSC and TA of "Bursa Siyahı" fruits.

Genotype	Fig Endosepsis (%)			SSC (°Brix)			TA (g/100 mL)		
	2020	2021	2022	2020	2021	2022	2020	2021	2022
16 08 05	12.33 ab	12.53 ab	17.59 ab	19.76	17.50	16.03	0.17 b	0.26	0.29
16 08 09	6.54 b	8.51 b	18.91 ab	18.93	17.10	16.03	0.25 ab	0.28	0.34
16 08 10	7.16 b	3.26 c	11.95 b	20.60	17.33	14.76	0.23 b	0.32	0.32
16 09 10	5.00 b	4.45 c	24.67 a	20.13	16.63	15.30	0.33 a	0.30	0.31
16 ZF 08	15.33 a	16.17 a	25.16 a	18.23	17.83	15.60	0.23 b	0.30	0.29
F-value	21.90	51.60	5.09	1.48	0.77	3.09	10.35	0.72	0.75
p-value	<0.01 **	<0.01 **	0.02 *	0.29 ns	0.57 ns	0.08 ns	<0.01 **	0.60 ns	0.58 ns

Small letters in the same column indicate a significant difference according to Duncan test. * and ** within a column indicate a significant difference at the $p < 0.05$ and $p < 0.01$ level. (ns: not significant).

The results show that the skin hue angle (H°) was significantly affected by genotype; however, the lightness (L) and chroma (C) were not affected (Figure 9). The highest hue, a lighter skin color, was obtained when the "Bursa Siyahı" cultivar was pollinated with the 16 08 05 and 16 09 10 genotypes. Rahemi and Jafari [21] also reported that pollen sources significantly affect the skin color of fig fruits.

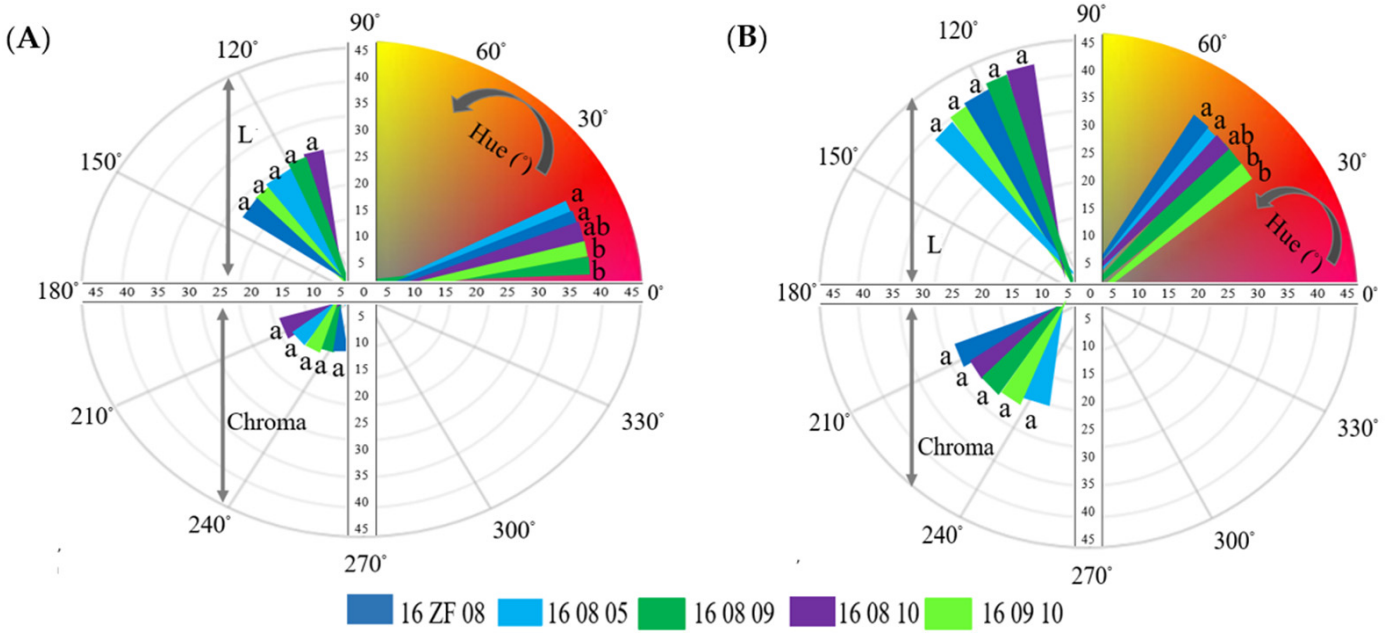

Figure 9. Effects of caprifig genotype on the skin (**A**) and flesh (**B**) color of "Bursa Siyahı" fruit. Small letters indicate a significant difference according to the Duncan test at $p < 0.05$.

Pourghayoumi et al. [5] stated that the H° was considerably affected by pollen sources, while the L and C values were not. The results show that flesh H° was significantly affected by genotype; however, the L and C values were not affected. The highest H° value was obtained when the 16 08 05 genotype was used for caprification. However, it was lower when the 16 09 10 genotype was used for caprification (Figure 9). Şimşek et al. [24] stated that consumers prefer pink and red flesh colors, as shown in the table figures. The H° value decreased when the 16 09 10 genotype was used as a pollen source, and the fruit flesh was a darker red–pink. Pourghayoumi et al. [5] stated that, when the "Avgeizi" pollen source was used on the "Sabz" cultivar, the female cultivar was lighter. Condit [68] reported that pollen sources with dark fruit flesh darkened the flesh color of female fruits. In the present study, contrary to Condit [68], when 16 08 05 and 16 ZF 08 were used as pollen sources, which had darker fruit skin and flesh color, the flesh color of the "Bursa Siyahı" fruit did not darken due to the higher hue value.

3.5. Multivariate Analysis

PCA was utilized to assess the dataset and its structure regarding the most significant variables. The analysis revealed that the first five principal components had eigenvalues exceeding 1.0, with a cumulative explained variance of 88.51% (Table 9).

Table 9. Eigenvalues and cumulative variance for five factors resulting from principal component analysis.

Parameters	Contribution				
	1	2	3	4	5
FW	0.902 *	0.125	0.041	−0.000	0.218
FT	0.894 *	0.093	0.086	−0.000	−0.065
PED	0.793 *	−0.318	0.262	−0.258	−0.084
PV	0.771 *	−0.013	−0.325	0.370	−0.220
OD	0.769 *	0.057	−0.428	0.146	0.205
SSW	0.761 *	0.169	−0.250	0.019	0.434
NSS	−0.758 *	0.136	0.406	0.061	0.066
NFS	0.744 *	0.394	0.036	0.061	−0.275
PPL	0.742 *	0.011	0.387	−0.104	−0.487
PG	0.718 *	0.533	−0.115	−0.118	0.220
FS	0.669 *	0.314	0.153	−0.04	0.493
<15 September	0.221	0.932 *	0.174	−0.123	0.038
15 October–1 November	−0.329	−0.858 *	0.285	0.071	0.074
NBP	0.060	0.872 *	0.127	0.132	0.307
FC	0.447	0.797 *	0.278	0.000	−0.098
NPC	−0.509	0.769 *	−0.095	0.070	0.052
DBPE	−0.390	0.736 *	−0.156	0.312	−0.360
FHue	−0.145	0.112	0.882 *	−0.000	0.000
TA	0.388	−0.387	0.580 *	−0.32	−0.44
SSC	0.080	−0.056	−0.172	0.901 *	0.140
SHue	−0.299	0.275	0.298	0.766 *	−0.178
Eigenvalue	8.133	5.156	2.446	1.659	1.191
% of variance	37.456	24.829	9.761	9.073	7.391
Cumulative variance %	37.456	62.285	72.045	81.119	88.510

* Significant factor loading (values above 0.50). NBP = Number of *B. psenes*, DBPE = Duration of *B. psenes* Exit, NPC = Number of *P. caricae*, PV = Pollen viability, PG = Pollen germination, PED = Pollen equatorial diameter, PPL = Pollen polar length, <15 September = the proportion of fruit ripening before 15 September, 15 October–1 November = the proportion of fruit ripening between 15 October and 1 November, NSS = Number of sterile seeds, FS = Fruit set, NFS = Number of fertile seeds, SSW = Single seed weight, FC = Fruit cavity, FW = Fruit weight, FT = Flesh thickness, OD = Ostiole diameter, SSC = Soluble solids content, TA = Titratable acidity, SHue = Skin Hue value, FHue = Flesh Hue value.

The variability in these components was distributed as follows: 37.45%, 24.82%, 9.76%, 9.07%, and 7.39%, respectively. PC1 accounted for the highest variance in the dataset. It exhibited positive correlations with pollen viability, germination, pollen equatorial diameter, pollen polar length, fruit set, fruit weight, ostiole diameter, fruit thickness, number of fertile seeds, and single seed weight but displayed negative correlations with the number of sterile seeds. PC2 showed positive correlations with the number of *B. psenes* and *P. caricae*, duration of *B. psenes* exit, fruit cavity, and proportion of fruit ripening before 15 September but was negatively associated with the proportion of fruit ripening after 15 October. PC3 demonstrated positive associations with flesh hue value and TA, while PC4 showed a correlation between SSC and skin hue value (Table 9). Based on average data from 2020 to 2022, 16 09 10 and 16 08 09 were positioned in the positive quadrant of PC1 due to their relatively high average fruit weight, fruit set, flesh thickness, pollen viability, and pollen germination value, but the 16 08 09 genotype was in the negative quadrant of PC2 due to the relatively low number of *B. psenes* and duration of *B. psenes* exit. Among them, 16 ZF 08 showed the lowest pollen germination, fruit set, the highest percentage of sterile seeds, and the highest proportion of fruit ripening after 15 October, positioning it furthest from the centroid. In the 16 08 10 genotype, since there were fewer *B. psenes* than in the 16 ZF 08 genotype and the rate of fruit ripening after 15 October was higher, the genotype was included in the negative section of PC1 and PC2 (Figure 10).

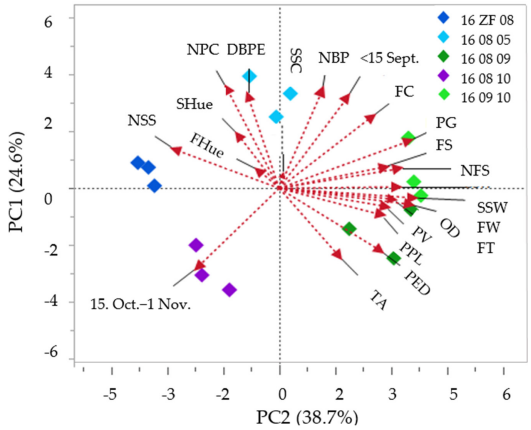

Figure 10. Multivariate analysis of pollen sources according to the variables. Abbreviations are explained in Table 9.

SEM analysis indicated that only pollen viability (0.357) directly affected the fruit set, which explained 12% of the variation in the fruit set (Figure 11). An increased number of *B. psenes* (0.641) in the genotypes increased the proportion of fruits that ripened before September, while an increase in pollen germination (−0.506) reduced the proportion of fruits that ripened after October 15th. The duration of *B. psenes* exit (−0.486) in fruits negatively affected the proportion of fruits that ripened after October 15th. Pollen germination positively influenced seed weight (0.642) and fruit weight (0.244), while pollen viability positively affected ostiole diameter (0.393) and the number of fertile seeds (0.823) but negatively affected the number of sterile seeds (−0.603). Moreover, an increase in sterile seeds negatively affected fruit weight (−0.403) but positively affected the fruit cavity (0.507).

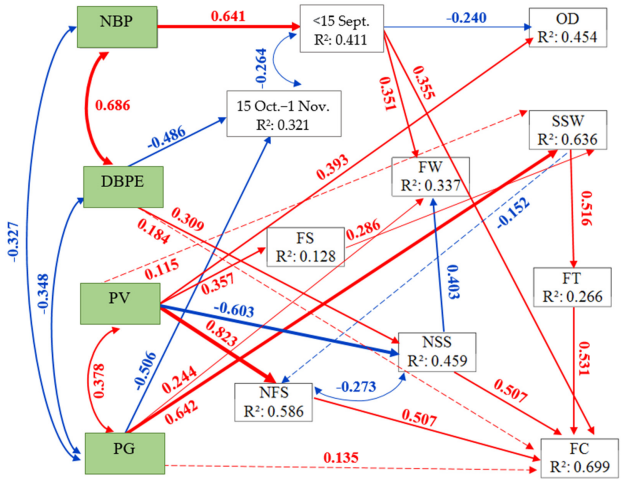

Figure 11. Structural equation model explaining the direct and indirect effects of the number of *B. psenes* (NBP), duration of *B. psenes* exit (DBPE), pollen viability (PV), and pollen germination (PG) on fruit set, ripening time, and fruit characteristics. <15 Sept. = the proportion of fruit ripening before 15 September, 15 Oct.–1 Nov. = the proportion of fruit ripening between 15 Oct. and 1 Nov., NSS = number of sterile seeds, FS = fruit set, NFS = number of fertile seeds, SSW = single seed weight, FC = fruit cavity, FW = fruit weight, FT = flesh thickness, OD = ostiole diameter. Red one-way arrows indicate positive effects, and blue one-way arrows indicate negative effects and dotted arrows indicate nonsignificant relationships. The width of an arrow indicates the relative strength of the causal influence. The numbers associated with each path represent the standardized coefficients. R^2, marked below each response variable, indicates the proportion explained by all the related impact factors. A double-headed arrow indicates a covariance between observed variables.

An increase in fruit seed weight positively affected flesh thickness (0.516), increasing the number of fruit cavities (0.196) (Table 10, Figure 11). In terms of indirect effects, an increase in pollen germination influenced flesh thickness (0.331) by increasing single seed weight and the size of the fruit cavity. An increase in pollen viability affected fruit weight (0.243) by reducing the number of sterile seeds and increasing ostiole diameter. An increase in pollen viability affected single seed weight (0.102) by influencing the fruit set. An increase in the number of *B. psenes* increased fruit weight (0.224) by increasing the proportion of fruits that ripened before 15 September. Moreover, the number of *B. psenes* positively and indirectly affected the number of fruit cavities (0.227) by increasing the proportion of fruits that ripened before 15 September.

Table 10. Effect estimates of the structural equation model.

Direct Effect	Estimate	Standardized Estimate	*p* (|Z| > z)	Indirect Effect	Estimate	Standardized Estimate	*p* (|Z| > z)
NBP→ < 15 Sept.	0.093	0.641	<0.01 **	NBP →FC	0.007	0.227	<0.01 **
DBPE→15 Oct.–1 Nov.	−7.507	−0.486	<0.01 **	NBP →FW	0.020	0.224	<0.01 **
DBPE→NSS	22.162	0.309	<0.01 **	DBPE →FC	0.544	0.156	0.01 *
PV→FS	0.502	0.357	0.01 *	DBPE→FW	−1.230	−0.124	0.02 *
PV→NFS	20.41	0.823	<0.01 **	PV →FW	0.219	0.243	<0.01 **
PV→NSS	−3.945	−0.603	<0.01 **	PV→OD	0.023	0.102	0.03 *
PV→OD	0.08	0.393	<0.01 **	PV→SSW	0.002	0.102	0.04 *
PG→15 Oct.– 1 Nov.	−0.590	−0.506	<0.01 **	PG→FT	0.060	0.331	<0.01 **
PG→SSW	0.014	0.642	<0.01 **	PG→FC	0.033	0.126	0.04 *
PG →FW	0.182	0.244	0.04 *	<15 Sept. →OD	0.024	0.148	0.02 *
<15 Sept. →FC	0.078	0.355	<0.01 **	FS→FT	0.023	0.147	0.01 *
<15 Sept. →FW	0.221	0.351	<0.01 **	NSS→OD	−0.006	−0.170	0.01 *
FS→SSW	0.005	0.286	<0.01 **	SSW→FC	2.296	0.196	0.03 *
FT→FC	0.765	0.531	<0.01 **				
NFS→FC	0.006	0.507	<0.01 *				
NSS→FC	0.024	0.507	<0.01 *				
NSS→FW	−0.055	−0.403	<0.01 *				
FW→OD	0.108	0.424	<0.01 *				
SSW→FT	4.175	0.516	<0.01 *				

* and ** within a column indicate a significant difference at the $p < 0.05$ and $p < 0.01$ level. Goodness-of-fit indices: chi-square = 80.70 ($p < 0.001$), df = 61, RMSEA = 0.084 (90% CI = 0.011; 0.131), CFI = 0.940, → = effect. Abbreviations are explained in Figure 6.

Single seed weight indirectly increased the fruit cavity (0.196) by increasing the flesh thickness. As the number of sterile seeds increased, the ostiole diameter (−0.170) decreased indirectly as the fruit weight decreased (Table 10, Figure 11). Khadivi-Khub and Anjam [69] reported that caprifig genotypes with higher *B. psenes* populations should be selected for pollination performance. According to both the biplot and SEM analyses, the fruit set was directly influenced by pollen characteristics compared to that of *B. psenes*. Multivariate analysis suggested that an excess of fertile seeds and a shortage of sterile seeds within the fruit can affect the cavity. Consistent with this, Karadeniz et al. [70] reported that the fruit cavity of the "Bursa Siyahı" fig cultivar increased due to the larger size of the fruits. Moreover, nonpollinated fig fruits with large fruit cavities and sterile flowers rapidly dehisced from the plant [67]. Furthermore, it has been determined that an increase in seed weight, rather than the number of seeds in the fruit, affects fruit flesh thickness. Similarly, Doi et al. [14] reported that, in blueberries, seed weight may be more appropriate than seed number when assessing the effects of seeds on fruit characteristics.

In this study, pollen characteristics directly affected fruit set, seed characteristics, and some physical characteristics of the fruit and indirectly impacted some physical characteristics of the fruit.

Doi et al. [71] reported that differences in the pollen source only affect the number of seeds included in a berry, and fruit weight and ripening are affected by the number

of seeds. Denney [72] defined this phenomenon as metania, a part of xenia. According to multivariate analysis, increasing the number of *B. psenes* in caprifig fruit increased the proportion of early harvested fruit. This can be explained by the fact that fertilized seeds produce large amounts of auxin which stimulates ethylene production in the tissue [73].

4. Conclusions

In the present study, although a suitable early pollen source could not be identified for the "Bursa Siyahı" cultivar, a later ripening genotype, 16 08 10, has been identified, which could facilitate the pollination of late "Bursa Siyahı" fruit. Additionally, despite the loss of *B. psenes* during the storage of caprifig fruits at 4 °C for 16 days, caprifig performance remained unaffected. It is understood from the multivariate analysis results that this situation is related to the pollen viability and germination rate not falling below critical values during storage. This result is important because the genotypes that ripen in the middle or mid-late period can be stored and used to pollinate late "Bursa Siyahı" fruits. The 16 08 09 and 16 09 10 genotypes stood out for promoting fruit set, obtaining medium-sized fruits, minimizing ostiole damage, and meeting consumer demand regarding fruit flesh and skin color. Since the weight of the exported "Bursa Siyahı" was expected to vary between 60 and 80 g, this expectation was met if the 16 08 05 genotype was used as the pollen source. If 16 08 10 and 16 ZF 08 genotypes were used as pollen sources, the rate of fruits ripening later was found to be high. Multivariate analysis revealed that pollen viability and germination rate, rather than *B. psenes* characteristics, directly affected fruit set and seed characteristics and indirectly affected fruit physical characteristics. The characteristics of *B. psenes* directly affected the harvest time and indirectly affected the physical properties of the fruits. These results are important because they explain which feature should be considered first when choosing the caprifig genotype for pollination.

Supplementary Materials: The following are available online at https://www.mdpi.com/article/10.3390/agronomy14050958/s1, Table S1: Effect of caprifig genotype, storage time and genotype and storage time interaction on the number of *B. psenes* and *P. caricae* and the duration of *B. psenes*'s exit; Table S2: Effects of the caprifig genotype, storage time and genotype and storage time interaction on pollen viability, germination, equatorial diameter, and polar length.

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