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Article

Influence of Different Liquid Spray Pollination Parameters on Pollen Activity of Fruit Trees—Pear Liquid Spray Pollination as an Example

Limin Liu [1,2,3], Ziyan Liu [1,2,3], Hu Han [1,2,3], Yulin Jiang [1,2,3], Xiongkui He [1,2,3,*], Yajia Liu [1,2,3,*], Dongsheng Wang [4], Xianping Guo [4] and Zhao Liang [5]

1 College of Science, China Agricultural University, Beijing 100193, China
2 Centre for Chemicals Application Technology, China Agricultural University, Beijing 100193, China
3 College of Agricultural Unmanned System, China Agricultural University, Beijing 100193, China
4 Institute of Horticulture, Henan Academy of Agricultural Sciences, Zhengzhou 450002, China
5 School of Information Science and Technology, Taishan University, Tai'an 271000, China
* Correspondence: xiongkui@cau.edu.cn (X.H.); liuyajia@cau.edu.cn (Y.L.)

Abstract: Liquid spray pollination (LSP) is widely used in fruit tree pollination. However, the LSP parameters that affect the pollen activity are still unclear. In this study, three LSP parameters that mainly affect the pollen activity were studied: storage time of pollen suspensions, sprayer parameters and unmanned aerial vehicle sprayer (UAVS) downwash airflow. In addition, sprayer parameters include the recirculation device, pump type, spraying pressure, nozzle size and revolutions per minute (rpm) of the rotary atomizer (RA). The results showed that, with the exception of nozzle size and UAVS downwash airflow, the pollen activity was significantly influenced by LSP parameters. The mean pollen activity decreased by 20.20% when the pollen suspension was stored for 30 min compared to 0 min. The activity of pollen in the tank was dramatically reduced using the recirculation device. The mean pollen activity decreases as the pump production maximum pressure increases. The mean pollen activity decreased from 40.7% to 29.02% when the spraying pressure increased from 0.3 MPa to 2.5 MPa. Additionally, the mean pollen activity decreased from 44.25% to 14.14% as the rpm of RA increased from 3000 rpm to 14,000 rpm. Our study demonstrated that pollen activity would be ensured by appropriate LSP parameters. This study provides a reliable theoretical basis for optimizing and advancing pear LSP technology.

Keywords: downwash airflow; pollination; rotary atomizer; sprayer; spraying pressure; UAVS

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1. Introduction

Pollination plays a crucial role in sexual plant reproduction [1], especially in some plants that have low seed production [2]. Worldwide, insect pollinators enhance approximately 7–8% of the total value of agricultural food production and 35% of fruit yields [3]. This shows that the requirement for insect pollinators is greater in fruit [4]. However, with the growth of intensive agriculture during the past century, the variety and abundance of insect pollinators have been rapidly decreasing [3,5]. There are many reasons for this phenomenon [6], including pesticide abuse [2,7], air pollution [8], climatic conditions [9], parasites [2], habitat loss [10], and a single ecological environment [6]. These can result in pollination limitation and, thus, lower fruit production [2,11].

Therefore, artificial pollination is a valuable tool for crop systems that lack pollinators or are unable to reliably maintain high-quality crop production [11,12]. With the development of artificial pollination, there is a wide variety of artificial pollination techniques, of which the majority (46%) are hand pollinated, 23% are mechanically pollinated (done by automated equipment) and 31% are unspecified methods [12]. Hand pollination is expensive, time-consuming and inefficient but has a high fruit setting rate [11,13]. To pollinate fruit trees, humans are required to climb fruit trees to pollinate them, which undoubtedly increases the risk to their lives and lengthens the pollination time [14,15]. Mechanical pollination has undoubtedly gradually replaced hand pollination in recent years. Numerous studies have confirmed mechanical pollination's effectiveness, lower cost, and higher fruit setting rates [12]. Most mechanical pollination use liquid spray pollination (LSP), which is quick, simple, and can even be done using currently available sprayers [15]. It is extremely convenient to carry out pollination operations using existing sprayers, newly improved sprayers, or even knapsack hand sprayers [4,12].

As a result, LSP is frequently employed to pollinate fruit trees such as kiwifruit, date palm, apple, pear and other fruits [12]. In date palm fruit production, for instance, the use of mechanical pollination reduces labor input by 50% when compared to hand pollination, while the yield of a season is increased by nearly three times [16]. This is mainly achieved by changing the concentration of pollen grains in the pollen suspension. There is evidence that Japanese pear LSP is comparable to hand pollination in terms of fruit set and fruit quality even though it uses one-third or fewer pollen grains compared to hand pollination and takes less time [15]. These focuses on the effect of suspension media and pollen concentration on pollen activity and fruit setting rate [13]. Although the pollen suspensions in the above LSP study on pears were applied using knapsack hand sprayers, the majority of LSP sprayers are large ground sprayers mounted on tractors [11]. On kiwifruit, Shi et al. evaluated an electromechanically regulated mechanical LSP system that had a fruit setting rate comparable to hand pollination [17]. The effect of the new spraying methods on the fruit setting rate and pollination efficiency was studied. The LSP was shown to be effective through the above studies. However, LSP takes a longer time to configure pollen solution and pollen suspension [11,15]. Even they used different sprayers and sprayer parameters. These have the potential to affect the pollen activity. Furthermore, unmanned aerial vehicle sprayers (UAVSs) are developing rapidly and playing an irreplaceable role in East and Southeast Asia [18], where they have the unique advantage of being able to take off and spray on any terrain [19]. Additionally, there are already numerous UAVS service providers in these locations [20]. Several studies have shown that LSP by UAVS is feasible [21,22]. UAVS will undoubtedly be a new platform for LSP. However, compared to other sprayers, UAVSs have a powerful downwash airflow [19]. This has the potential to affect the pollen activity.

In this study, pollen solution refers to a specific solution prepared without pollen. The pollen suspension is a suspension prepared by placing an appropriate pollen grain amount into the pollen solution. Pollen activity is one of the most important factors affecting the final pollination results of pears, and the germination rate of pollen grains is a crucial way to assess pollen activity [13]. Therefore, pollen germination rate of pollen grains is used to indicate pollen activity in this paper.

Although the aforementioned studies have extensively investigated LSP, they have focused on pollen suspension configuration and spraying methods on pollen activity. They have not systematically investigated the effect of LSP parameters on pollen activity, especially from the pollen suspension configuration to being sprayed onto the stigma. Therefore, the aim of this study was to optimize the LSP parameters, assuring a higher activity of pollen sprayed onto the flower stigma... The study evaluated the effect of different LSP parameters on the pollen activity. The influences of the storage time of the pollen suspension, sprayer parameters and UAVS downwash airflow on pollen activity were verified. This study also explored the possible influences of each LSP parameter on pollen activity, which provide a reliable theoretical basis for optimizing and advancing pear LSP technology.

2. Materials and Methods

2.1. Pear Pollen and Pollen Solution Configuration

Xuehuali (Pyrus bretschneiderilia) pollen from Sanmenxia (34.6203° N, 111.6973° E), Henan Province, China, was collected on 22 March 2021, and stored in a refrigerator at −18 °C that night. At the same time, it is always stored in the refrigerator and the appropriate amount of pollen grains are taken out with a spoon during test or pollination. It can remain pollen activity at the temperature for several years. To awaken pollen grains before LSP, the pollen from the refrigerator was deposited in an artificial climate box (RDN-500, Yanghui Technology Co., Ltd., Ningbo, China) of 25 °C with a relative humidity of 90% for six hours. The composition of the pollen solution included xanthan gum, sucrose, calcium gluconate, and boric acid. As an illustration, Table 1 shows the content of various components for the 50 L pollen solution.

Table 1. Composition of pear pollen solution. The table contains the composition, content, and mass (volume) of each component in the pollen solution.

Ingredients	Content (%)	Volume or Mass
Pure water	-	Fixed volume to 50 L
Sucrose	10.00	5.00 kg
Xanthan gum	0.02	10.00 g
Calcium gluconate	0.05	25.00 g
Boric acid	0.01	5.00 g

Pure water was selected when creating the pollen solution. First, 25 L of pure water was added to the 50 L vessel, which was used for heating. After boiling the pure water, heating was stopped. The water was left to stand for 1–2 min. Then, 10.00 g of xanthan gum was put into the vessel and stirred continuously. After the xanthan gum was fully dissolved, 5.0 kg of sucrose was placed into the vessel and stirred continuously. The solution was filtered through gauze into a 50 L fixed-volume tank once the sucrose had likewise fully dissolved. A total of 5.0 g of calcium gluconate and 1.0 g of boric acid were put into a fixed-volume tank and thoroughly dissolved. Finally, when all the ingredients were fully dissolved, pure water was added to the fixed-volume tank to ensure that the volume of the solution was 50 L. The resulting pollen solution had a 24 h shelf life.

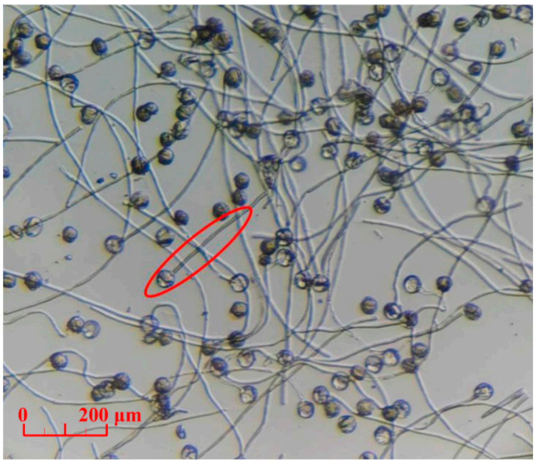
2.2. Pollen Suspension Configuration and Pollen Activity Determination Method

2.2.1. Pollen Suspension Configuration

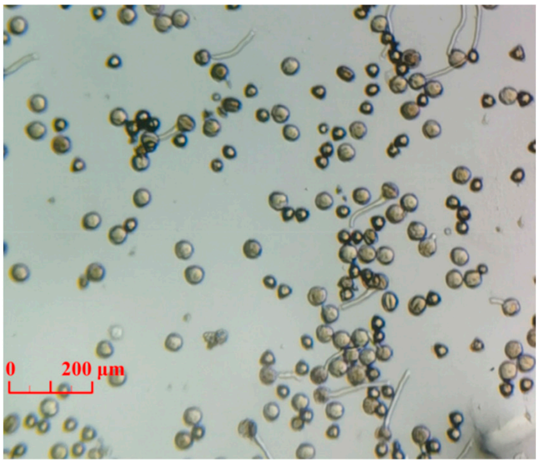
The pollen and pollen solution were measured in accordance with the necessary pollen grain concentration, which was 0.8 $g \cdot L^{-1}$. The required pollen grain amount and two-thirds of the required pollen solution were poured into the mixing tank. Glass rods were used to stir the solution well. A 50 L fixed-volume tank was then filled with the stirred suspension. Next, the remaining pollen solution was poured into the fixed-volume tank to reach the required volume. Finally, the pollen grains and pollen solution were thoroughly stirred to make the pollen suspension.

2.2.2. Pollen Activity Determination Method

The pollen germination rate of pollen grains using liquid culture was observed by microscope (SZM45B1, Shunyu Technology Co., Ltd., Changzhou, China). After the pollen suspension was configured or sprayed, 100 µL of the pollen suspension was placed into a concave slide (size 28 mm × 76 mm) using a pipette gun (Research Plus, Eppendorf Technology Co., Ltd., Hamburg, Germany). Since pollen germination requires a certain humidity, three sheets of moistened filter paper were placed in culture dishes (10 cm diameter). Concave slides were positioned on moistened filter paper at the same time. Culture dishes were placed in the artificial climate box with a relative humidity of 95% and a temperature of 25 °C. The pollen grains were incubated in the artificial climate box for three hours under dark conditions. After the incubation was completed, the pollen grains inside the concave slides were observed and photographed under a microscope. The pollen grains that had a pollen tube length longer than their diameter were considered to be germinated pollen (Figure 1a). Additionally, the pollen grains in the Figure 1b were non-germinated pollen grains. The germination rate of pollen grain (pollen activity) was obtained by the number of total pollen grains and germinated pollen grains (Equation (1)). Three fields of view were randomly selected from each concave slide for observation.



(**a**)



(**b**)

Figure 1. Two different states of pollen grains. The magnification of the picture is 50 times. The germinated pollen was marked by a red oval, which was characterized by the length of the pollen tube being significantly larger than the diameter of the pollen grains. (**b**) was non-germinated pollen, which is characterized by no pollen tube. (**a**) Germinated pollen grains. (**b**) Non-germinated pollen grains.

2.3. Test Sprayers

The pollen grains were applied to the stigma of the pear flowers through sprayers. Eight sprayers were used for this study (Table 2) to study the influence of sprayer parameters on pollen activity. Among them, the first three were air can sprayers of various capacities, which were small air can sprayer (ACP A06L, RuiPu Sprayer Technology Co., Ltd., Taizhou, China), middle air can sprayer (ACP A2L) and large air can sprayer (ACP A5L). When tested, they were filled with half of their own volume of pollen suspension. The knapsack hand sprayer (3WBS-16P, Binda Plastic Co., Ltd., Taizhou, China) has a leather cup-type piston pump. The electric diaphragm pumps are used in a knapsack electric sprayer (3WBES-20SF, Nongzhuangyuan Co., Ltd., Suzhou, China), DJI T20 UAVS (AGRAS T20, SZ DJI Technology Co., Ltd., Shenzhen, China) and homemade orchard sprayer. Additionally, the pump of the homemade orchard sprayer can be replaced with an electric plunger pump.

2.4. Tests of Pollen Suspension Storage Time on Pollen Activity

The pollen grains in the pollen suspension have biological activity. As a result, the pollen suspension was placed in the tank to confirm whether storage time had an influence on pollen activity. It takes some time from the preparation of pollen suspension to spraying it on the stigma. The time was defined as the storage time of the pollen suspension. The test pollen suspension was sampled every 5 min until 30 min, which was pipetted into the concave slide and incubated in the artificial climate box. A microscope was used to detect the number of germinated pollen grains and non-germinated pollen grains in the field of view after the pollen suspension was incubated for three hours. The test was repeated three times. On 25 March 2021, this test was carried out inside at a temperature of 25 °C and a relative humidity of 42%.

Table 2. Types and parameters of sprayers for the test. In this study, eight types of sprayers (three types of pumps) were used for LSP. The parameters of each sprayer were listed, including the type of pumps, recirculation device, revolutions per minute (rpm) of pumps, tank volume, pump pressure, and flow rate.

Types	Small Air Can Sprayer	Middle Air Can Sprayer	Large Air Can Sprayer	Knapsack Hand Sprayer	Knapsack Electric Sprayer	Homemade Orchard Sprayer	DJI T20 Unmanned Aerial Vehicle Sprayer
Picture							
Pump types	Air compression pump	Air compression pump	Air compression pump	leather cup-type piston pump	Diaphragm pump	Diaphragm pump (plunger pump)	Diaphragm pump
Recirculation device	without	without	without	without	without	with (with)	without
rpm of pumps	-	-	-	-	450	550 (960)	400
Tank volume/L	0.6	2	5	16	20	25 (25)	20
Pressure/MPa	0.08–0.12	0.12–0.25	0.32–0.65	0.25–0.45	0.3–0.65	0.35–0.55 (0–3)	0.2–0.4
Flow rate/(L·min^{-1})	0.2–0.6	1.1–1.8	1.9–3.2	2.8–3.8	2.2–4.2	10.0–19.8 (8–12)	4.5–6

Note: The pump of homemade orchard sprayers can be installed diaphragm pump or plunger pump.

2.5. Tests of the Recirculation Device on Pollen Activity

The homemade orchard sprayers have a recirculation device, which consists of a pressure regulator valve and overflow pipeline (Figure 2a). Additionally, recirculation devices are used to keep the pressure and flow stable. The recirculation device would flow the extra liquid back to the tank through the overflow pipeline. The pollen activity is probably impacted, as this extra pollen suspension is recirculated back into the tank via the pump and recirculation device.

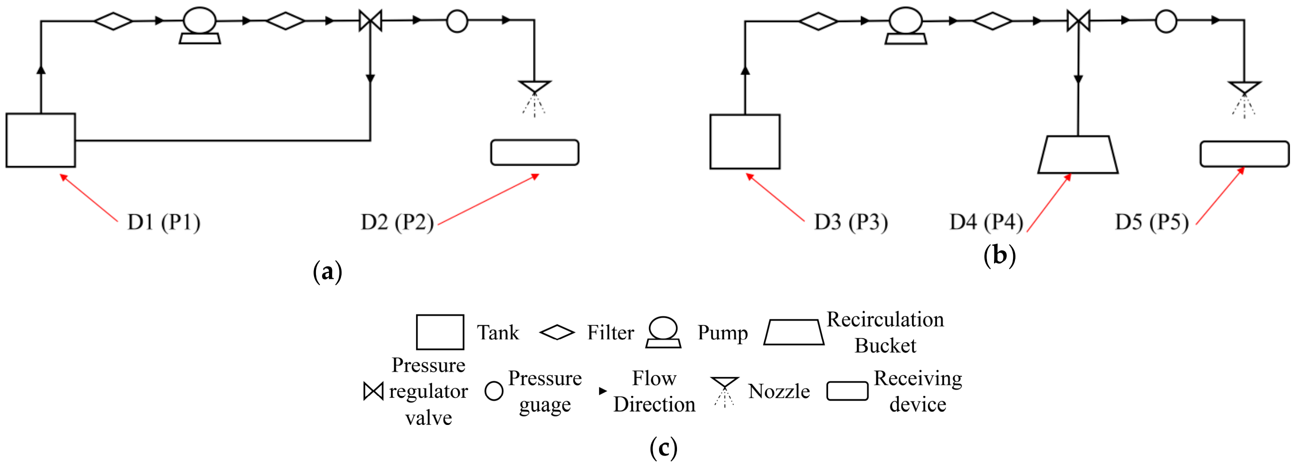


Figure 2. Tests on the influence of recirculation device on the pollen activity in pollen suspensions. (**a**) Pollen suspension recirculating into the tank. (**b**) Pollen suspension recirculating into the recirculation bucket. (**c**) The device represented by each symbol. D1 (P1) is used to indicate the sampling position, where D1 indicates that the system uses a diaphragm pump, and P1 uses a plunger pump.

To verify whether the recirculation of pollen suspension into the tank affects the pollen activity, two spraying methods were designed (Figure 2). In the case, one was that the pollen suspension was recirculated into the tank (D1(or P1)) through the recirculation device (Figure 2a), whereas the other was recirculated into the recirculation bucket ((D_4(or P_4)), Figure 2b). To avoid nozzle clogging, they were filtered twice by filters. A spraying pressure of 0.3 MPa (displayed on a pressure gauge (8001-A3, Ma'anshan Naite Instrument Technology Co., Ltd., Ma'anshan, China)) was maintained with the recirculation device. The pollen suspension that had been sprayed was collected in the receiving device. After the spraying was completed, the pollen suspension samples at the sampling positions were dripped into a concave slide using a pipette gun and then placed into an artificial climate box for three hours. The flow rates of both diaphragm and plunger pumps were adjusted to 5 L·min^{-1}, and the spraying system was run for 5 min. All pollen suspensions in the tank were theoretically passed through the pumps and the reflux devices. On 26 March 2021, this test was carried out inside at a temperature of 25 °C and a relative humidity of 41%. The air pollen suspension temperature was 25 °C, and these experiments were repeated three times.

2.6. Tests of Pump Types on Pollen Activity

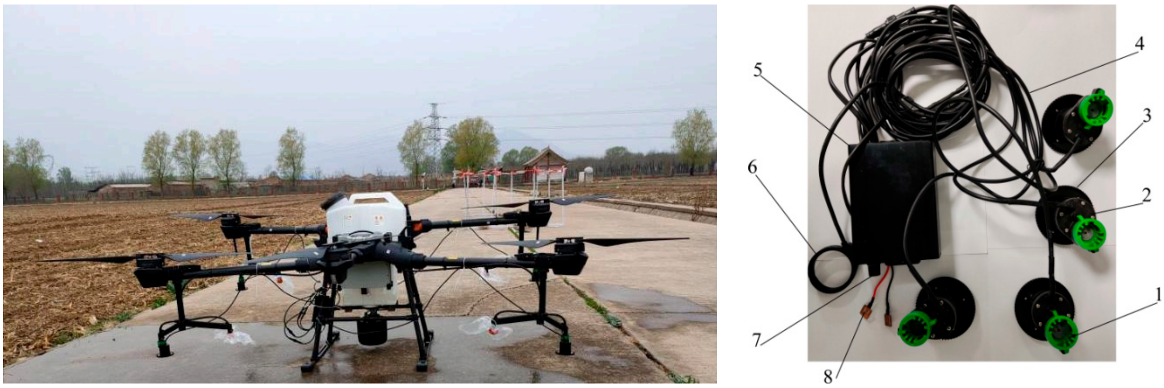
Four pump types were used in the study: air compression pumps, leather cup-type piston pumps, electric plunger pumps, and electric diaphragm pumps. Due to the tank of the air can sprayers can be used as both an air can and a liquid container. Therefore, these tanks need to be tightly sealed. The tank was cleaned three times using pure water. Next, the tank was filled with the prepared pollen suspension. Among the air can sprayers and knapsack hand sprayer must be compressed until they cannot be compressed. To maintain a constant spray pressure throughout the test, they must be continually compressed. Before officially applying pollen suspension, all sprayers must spray 5 to 10 s into the ground to remove pure water from the pipeline. It was necessary to complete each sprayer spraying test within 10 min. The pressure of all electric pumps was kept at 0.3 MPa during the test. No. 01 flat-fan nozzles (XR11001, TeeJet Co., Inc., Wheaton, IL, USA) were used. On 26 March 2021, this test was carried out inside at a temperature of 25 °C and a relative humidity of 41%. The pollen suspension temperature was 25 °C, and these experiments were repeated three times.

2.7. Tests of Nozzle Size and Spray Pressure on Pollen Activity

No. 01, 015 (XR110015), and 02 (XR11002) ordinary flat-fan nozzles and No. 01 (BYCO11001, Geqiang Nozzle Co., Inc., Foshan, China), 015 (BYCO110015), and 02 (BYCO11002) high-pressure flat-fan nozzles were used to study the effect of nozzle size on pollen activity. These flat-fan nozzles were used to spray pollen suspension by a homemade orchard sprayer (with diaphragm pump) at 0.3 MPa pressure, respectively. The No. 01 high-pressure flat-fan nozzle was used to apply the pollen suspension by homemade orchard sprayer (with plunger pump) at pressures of 0.3, 0.5, 1.0, 1.5, 2.0 and 2.5 MPa. No recirculation device was used for the whole test. On March 27, 2021, this test was carried out inside at a temperature of 25 °C and a relative humidity of 44%. The pollen suspension temperature was 25 °C, and all experiments were repeated three times.

2.8. Tests of Rotary Atomiser Revolution Speeds on Pollen Activity

The nozzle body of the rotary atomizer (RA) system was connected to the UAVS (Figure 3a). The RA system was designed for the DJI T20 UAVS by us in this study (Figure 3b). The pollen suspension coming from the UAVS pump flows through the nozzle body into the fluted disc, the edge of which was equally distributed with 180 triangular teeth. Additionally, the fluted disc was rotated at high speed by a brushless motor. The UAVS's battery provides electricity to the electrodes, and the induction ring is mounted to the pump of the UVAS. When the pump is operating, the electromagnetic characteristics of the pump change. By detecting electromagnetic changes, the induction ring controls the switching of the RA. Electronic speed control (ESC) is used to regulate the revolution speed of the brushless motor. A tachometer (DT2234C, Shenzhen Tony Electronics Co., Ltd., Shenzhen, China) was used to measure the revolution speed of the brushless motor. The maximum speed of the brushless motor is 14,000 rpm, and the minimum is 3000 rpm. On 29 March 2021, this test was carried out inside at a temperature of 25 °C and a relative humidity of 45%. The pollen suspension temperature was 25 °C, and all experiments were repeated three times.



(**a**) (**b**)

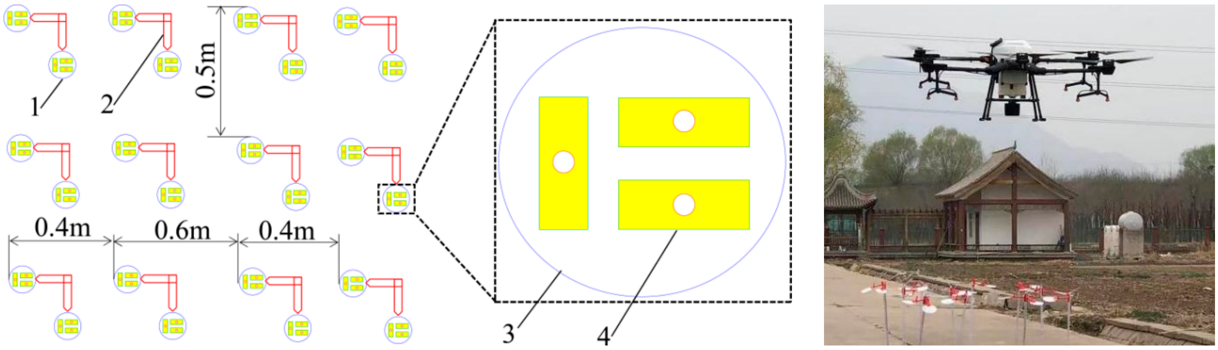
Figure 3. DJI T20 UAVS and electronically controlled RA system. (1) Nozzle body, (2) brushless motor, (3) fluted disc, (4) power line, (5) control box, (6) induction ring, (7) ESC and (8) electrodes. (**a**) DJI T20 UAVS carrying RA and (**b**) electronically controlled RA system.

A laser diffraction system (SprayTec, Malvern Panalytical Ltd., Worcestershire, UK) was used to measure the droplet size spectrum at different revolution speeds. The 10th percentile diameter (Dv10), volume median diameter (Dv50), 90th percentile diameter (Dv90), relative span (RS) and spray volume fractions generated with droplets finer than 75, 100, 150, 200, and 250 (V75μ, V100μ, V150μ, V200μ and V250μ) were recorded for further analysis [20]. Additionally, the revolution speeds were 3600 rpm and 12,400 rpm for pollen suspension droplet sizes Dv50 near 100 μm and 250 μm, respectively. Subsequently, pollen activity was measured at different rotational speeds, which were 3000, 3600, 4000, 5000, 6000, 7000, 8000, 9000, 10,000, 11,000, 12,000, 12,400, 13,000 and 14,000 rpm.

2.9. Tests of UAVS Downwash Airflow on Pollen Activity

Compared with other sprayers, the UAVSs have a strong downwash airflow field [18]. The droplets are easily blown to the target canopy to complete the spraying operation under the influence of the downwash air field. To verify whether there was an influence on pollen activity by the downwash airflow field of the UAVS. A receiving device was designed to receive the pollen suspension applied by the UAVS (Figure 4a) on 13 April 2021 at the Experimental Station of China Agricultural University (40.1462° N, 116.1898° E) in Shangzhuang, Haidian District, Beijing, China. It consists of petri dishes (diameter 24 cm), long pole double-ended type clamps, crossbars and uprights and was used to simulate a real fruit tree canopy. Three concave slides (size 24 mm × 50 mm) were placed in the petri dish. 50 μL of pollen solution was dripped into the concave slides and spread out by toothpicks. At 1.0 m above the ground, long pole double-ended type clamps were used to secure the petri dishes to the uprights.

The No. 01 ordinary flat-fan nozzle and the RA (8000 rpm) were selected for the test. A pipette gun was used to drip the pollen suspension (original pollen suspension) from the tank into a concave slide, which was then placed in an artificial climate box for incubation before the test. At the same time, the pollen suspension was sprayed from the nozzle into petri dishes when the pump was turned on by a remote control. It was used to compare the pollen activity in the pollen suspensions received by the downwash airflow field. In the test, the DJI UAVS's enhanced manual mode was utilized to control the UAVS's speed and keep it at 1.5 m/s (Figure 4b, 2.5 m above the deployment area). After each spraying was completed, concave slides with collected pollen suspension were placed in the artificial climate box for incubation. During the test, the air temperature was 14~16 °C, the wind speed was 0.8~1.2 m·s^{-1}, and the relative humidity was 16%. These experiments were repeated three times.



(a) (b)

Figure 4. Test of the effect of UAVS downwash airflow site on pollen activity. (1) Receiving device, (2) long pole double-ended-type clamp, (3) petri dish and (4) concave slides. (**a**) Pollen suspension receiving device. (**b**) Test site view.

2.10. Data Processing

In this study, the formula for calculating pollen activity is shown in Equation (1). A larger percentage indicates higher pollen activity. The percentage in all tests was counted to indicate pollen activity.

$$G_r = \left(\frac{T_g}{T_p}\right) \times 100\% \quad (1)$$

where G_r stands for pollen activity, T_g for the number of germinated pollen grains and T_p for the total number of pollen grains in the field of view.

The droplet RS can be obtained by Equation (2). A narrower droplet RS corresponds to a smaller value of the droplet spectrum, which denotes a more uniform droplet.

$$RS = \frac{DV90 - DV10}{DV50} \quad (2)$$

The Coefficient of Variation (CV) value was used to indicate the degree of dispersion between different measurement groups. In this study, it can be used to express the degree of dispersion of pollen activity in different groups. It can be obtained by Equation (3).

$$CV = \left(\frac{SD}{MN}\right) \times 100\% \quad (3)$$

where SD is the standard deviation of the group data, and MN is the mean of the group data.

MN and SD can be expressed by Equations (4) and (5), respectively.

$$MN = \frac{\sum_{i=1}^{n} P_i}{n} \quad (4)$$

where n denotes the sample count and P_i denotes the ith sample's data.

$$SD = \sqrt{\frac{\sum_{i=1}^{n}(P_i - MN)^2}{n-1}} \quad (5)$$

The test data were plotted using OriginPro Version 2020 (OriginLab Inc., Northampton, MA, USA). Before the data analysis, the experimental data were subjected to the normal distribution test using SPSS Statistics Version 22 (IBM Inc., Armonk, NY, USA). Additionally, the experimental data were subjected to one-way analysis of variance (ANOVA) and correlation analysis using SPSS. The Duncan test of SPSS one-way ANOVA was also used. In this study, $p = 0.05$ indicates significant difference. All experimental data conformed to a normal distribution.

3. Results

3.1. Influence of Storage Time on the Pollen Activity

The correlation analysis by SPSS showed that the storage time of the pollen suspension and the mean pollen activity were extremely significantly negatively correlated (r = −0.98). Additionally, the storage time of the pollen suspension and the CV value of pollen activity were positively correlated but not significant (r = 0.498).

Figure 5a shows that pollen activity gradually decreased when the storage time of the pollen suspension was increased. There was no significant difference in pollen activity within the first 15 min of the storage time of the pollen suspension being placed compared to the original pollen suspension (0 min in Figure 5a). When the storage time of the pollen suspension was 20 min, the pollen activity in the pollen suspension significantly changed from the pollen activity of original pollen suspension in terms of pollen activity. In comparison to the pollen activity of original pollen suspension, the mean pollen activity in the pollen suspension from 5 min to 30 min was reduced by 2.04%, 3.09%, 7.27%, 10.60%, 14.48% and 20.20%, respectively. However, the CV value trend of pollen activity was different. As the storage time of the pollen suspension increased, the CV value of pollen activity showed a tendency to decrease and then increase. When the storage time of the pollen suspension was 15 min, the CV value of pollen activity was at a minimum of 4.79%. Then, as the pollen suspension was left in for longer periods of time, the CV value of the storage time of pollen suspension gradually increased to 10.54%.

3.2. Influence of the Recirculation Device on the Pollen Activity

There was no significant difference between the original pollen suspension (or pollen suspension left 5 min) and D3 or P3 (Figure 5b), but there was a significant difference between the original pollen suspension (or pollen suspension left for 5 min) and D1 or P1 (Figure 5a). In addition, there was a significant difference between D3 (or P3) and D1 (or P1). This indicates that the recirculation device had a significant influence on the pollen activity in the tank. Compared to D3 and P3, pollen activity in D4 and P4 was reduced by 28.33% and 30.09%, respectively. There was no significant difference between pollen activity in D4 (or P4) and pollen activity in D1 (or P1). However, the pollen activity in the D4 and P4 was decreased by 16.12% and 16.92%, respectively, in comparison to the D1 and P1. With or without the recirculation device, there was a significant difference between the pollen activity from the spray nozzles and the pollen activity in the tank (D3 and D5, P3 and P5, D1 and D2, P1 and P2) but not with the pollen activity in the recirculation bucket (D4(or P4)).

3.3. Influence of Pump Types on Pollen Activity

Pollen activity was significantly reduced after the pollen suspension was sprayed out by different pumps (Figure 5c). This indicates that pump types have a large influence on the pollen activity. There was a higher reduction in pollen activity, but there was no significant difference between pollen activity in original pollen suspension and pollen activity in pollen suspensions sprayed with small air can sprayer and middle air can sprayer. Compared to the original pollen suspension, the pollen activity applied by the small air can sprayer and middle air can sprayer was decreased by 13.91% and 17.71%, respectively. As the pressure of air can sprayers was further increased, the pollen suspension that was sprayed had distinct changes in pollen activity from the original pollen suspension. Additionally, compared to that of the original pollen suspension, the pollen activity of the large air can sprayer was decreased by 25.53%. This indicates that air compression pumps at low pressure do not significantly reduce the pollen activity, but at high pressure do. Other sprayers and the original pollen suspension had a significant difference, with the exception of both a small air can sprayer and middle air can sprayer. There was a significant difference between the knapsack hand sprayer and the homemade orchard sprayer (plunger pump); however, there was no significant difference with the other three electric sprayers. In addition, the pollen suspension through the plunger pump showed the lowest mean pollen activity (40.7%) and highest CV value (56.25%) of pollen activity.

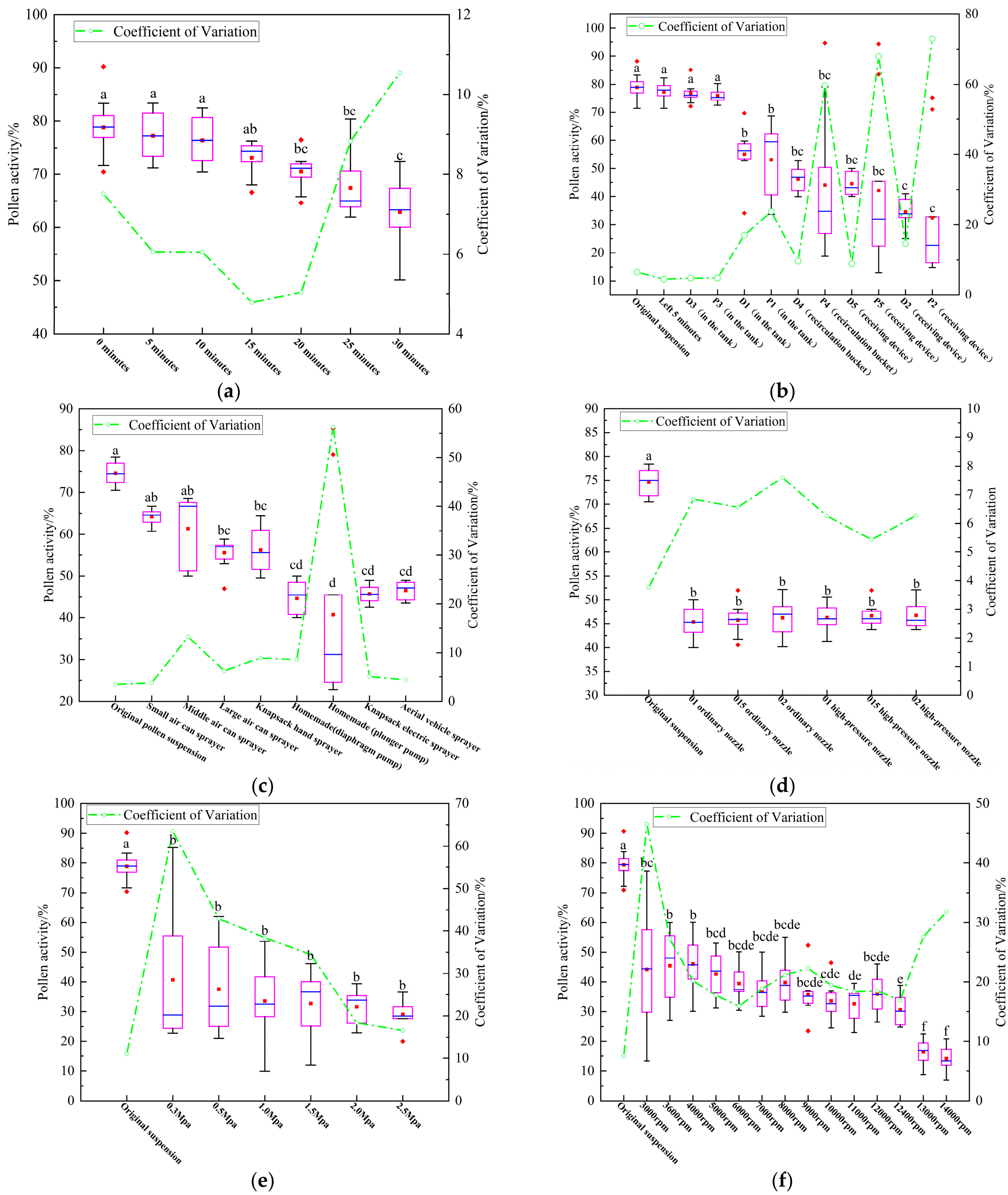


Figure 5. Results of the influence of pollen suspension storage time and sprayer parameters on pollen activity. (**a**) Pollen suspension storage time, (**b**) recirculation device, (**c**) different sprayers (pumps), (**d**) nozzle sizes, (**e**) spraying pressures and (**f**) rpm of RA. Different letters in (**b**) indicated different sampling positions (Figure 2). Different letters indicate significant differences (Duncan test, α = 0.05).

3.4. Effect of Nozzle Size on Pollen Activity

The pollen activity applied by different nozzles was significantly different from the pollen activity of the original pollen suspension (Figure 5d). However, there was no significant difference in pollen activity between different sized nozzles. With an increase in the nozzle size, the pollen activity of the same type of nozzles increased somewhat (lower than 1.28%), but the difference was not significant. Additionally, there was no significant increase in their CV value, which was lower than 7.64%.

3.5. Effect of Spray Pressure on Pollen Activity

Spray pressure was negatively and highly significantly correlated with the mean pollen activity (r = −0.951) and CV value of pollen activity (r = −0.936) by SPSS correlation analysis. The pollen activity gradually decreased from 40.7% to 29.02% as the spray pressure increased from 0.3 MPa to 2.5 MPa (Figure 5e). Additionally, the CV value of pollen activity decreased from 63.46% to 16.59%. There was a significant difference in the pollen activity sprayed from the nozzle compared to the original pollen suspension. However, there was no significant difference in the pollen activity at different spray pressures.

3.6. Influence of RA Revolution Speed on Droplet Parameters and Pollen Activity

The values of Dv10, Dv50 and Dv90 decreased with increasing revolutions speed of the RA and showed a significant difference (Table 3). Additionally, the percentage of small droplets gradually increases as the revolution speed increases. This shows that the overall droplet size of the RA decreases with increasing rotational speed. The droplets RS of the RA are uniform, whereas there are large differences at 3000 rpm, 3600 rpm, 12,400 rpm, 13,000 rpm and 14,000 rpm.

Table 3. Results of the influence of RA revolution speed on droplet parameters. The parameters of the pollen suspension droplets are listed in the table. The mean ± SE is used to tabulate the results.

rpm	Dv10/(μm)	Dv50/(μm)	Dv90/(μm)	RS	V75μ/(%)	V100μ/(%)	V150μ/(%)	V200μ/(%)	V250μ/(%)
3000	129.54 ± 10.46 a	294.03 ± 6.18 a	584.51 ± 28.12 a	1.55 ± 0.05 b	3.60 ± 0.62 hi	6.08 ± 1.05 g	13.72 ± 1.95 h	25.19 ± 2.22 h	38.52 ± 1.89 g
3600	125.62 ± 10.65 a	247.51 ± 7.21 b	436.35 ± 23.08 b	1.32 ± 0.11 abc	2.36 ± 1.02 i	4.49 ± 2.21 g	18.48 ± 3.41 gh	37.46 ± 2.84 g	54.58 ± 2.28 f
4000	121.55 ± 13.41 a	199.09 ± 7.30 c	337.24 ± 26.46 c	1.08 ± 0.21 cd	1.07 ± 1.50 i	3.77 ± 3.73 f	23.43 ± 5.50 g	50.24 ± 3.76 f	71.31 ± 3.24 e
5000	94.70 ± 11.28 b	161.32 ± 5.91 d	271.40 ± 9.69 d	1.10 ± 0.17 bcd	4.03 ± 2.41 ghi	12.70 ± 5.22 f	42.82 ± 4.85 f	69.41 ± 1.75 e	85.94 ± 1.56 d
6000	87.21 ± 7.34 bc	140.19 ± 7.61 e	225.03 ± 14.8 e	1.00 ± 0.09 d	4.94 ± 2.50 fghi	18.32 ± 4.96 f	53.44 ± 14.49 e	83.07 ± 4.05 d	94.20 ± 2.41 c
7000	79.18 ± 4.59 cde	130.40 ± 2.32 f	207.84 ± 9.13 def	0.99 ± 0.11 d	8.04 ± 2.13 efg	24.45 ± 2.96 e	64.50 ± 1.90 d	87.91 ± 2.46 c	96.92 ± 1.38 abc
8000	77.13 ± 4.44 de	124.18 ± 4.18 fgh	196.74 ± 8.69 ef	0.96 ± 0.03 d	8.84 ± 0.76 de	27.97 ± 2.43 ef	69.68 ± 3.73 cd	90.76 ± 2.16 abc	97.91 ± 0.79 a
9000	70.01 ± 2.71 e	117.04 ± 5.44 h	186.88 ± 27.88 f	1.04 ± 0.15 cd	13.23 ± 1.94 d	34.68 ± 3.44 c	74.35 ± 5.623 bc	92.13 ± 4.73 abc	97.50 ± 3.02 ab
10,000	72.85 ± 2.90 de	125.03 ± 5.64 fg	205.14 ± 21.60 ef	1.05 ± 0.15 d	11.13 ± 1.49 de	29.57 ± 2.28 cde	68.06 ± 6.29 cd	88.79 ± 5.23 bc	96.76 ± 2.76 abc
11,000	73.36 ± 4.21 de	120.77 ± 4.97 gh	188.96 ± 9.66 f	0.95 ± 0.08 d	11.08 ± 2.63 de	30.92 ± 4.01 cd	72.48 ± 3.87 c	92.84 ± 2.46 ab	98.84 ± 1.08 a
12,000	80.84 ± 10.77 cd	123.06 ± 3.58 fgh	182.24 ± 14.56 fg	0.83 ± 0.22 cd	7.62 ± 5.18 efgh	24.97 ± 8.05 e	74.57 ± 4.73 bc	94.96 ± 3.63 a	99.04 ± 1.27 a
12,400	80.84 ± 4.18 f	101.18 ± 5.05 i	187.27 ± 32.91 f	1.39 ± 0.32 bc	27.22 ± 4.18 c	46.42 ± 4.68 b	80.50 ± 3.86 ab	93.51 ± 3.04 a	96.58 ± 1.75 abc
13,000	35.04 ± 4.92 g	81.87 ± 8.20 j	189.19 ± 58.37 f	1.87 ± 0.53 a	44.17 ± 6.47 b	64.41 ± 6.56 a	86.48 ± 5.82 a	92.66 ± 4.59 ab	94.6 ± 3.19 c
14,000	26.99 ± 3.50 g	58.37 ± 6.18 k	149.31 ± 42.94 g	2.06 ± 0.49 a	50.28 ± 5.62 a	67.7 ± 5.76 a	87.68 ± 6.18 a	94.41 ± 3.19 a	95.03 ± 3.16 bc

Note: Significant differences between means are indicated by different letters at the level of 0.05.

The pollen activity gradually decreased from 44.25% to 14.14% with the increasing RA revolution speed (Figure 5f). The increase in revolution speed had a significant influence on the pollen activity. When the RA revolution speed was from 3000 rpm to 12,000 rpm, there was no significant difference in the pollen activity between different revolution speeds. At revolution rates greater than 12,400 rpm, pollen activity was significantly different from what it was at other lower revolution rates. The correlation between RS and the CV value of pollen activity was r = 0.645, indicating that RS was significantly and positively correlated with the CV value of pollen activity.

The revolution speed has no correlation with RS, and the other spray parameters are correlated (Table 4). Among them, the revolution speed has a negative correlation with Dv10, Dv50 and Dv90 of the droplets, indicating that the greater the revolution speed of the RA is, the smaller the droplet parameters. The revolution speed has a positive correlation with V75μ, V100μ, V150μ, V200μ and V250μ, indicating that the larger the revolution speed of the RA is, the larger the percentage of those droplet parameters.

And the correlations of different parameters with pollen activity were constructed by SPSS (Table 4). A significantly significant negative correlation was found between pollen activity and these parameters, which comprised rpm and the percentage of droplets smaller than 75, 100, 150 and 200 μm. Pollen activity was highly significantly positively correlated with these parameters, which included Dv10, Dv50, and Dv90. It was not significantly correlated with these parameters, which were RS and V250μ.

Table 4. Correlation analysis between RA revolution speed and spray parameters and pollen activity.

Types	rpm	Dv10	Dv50	Dv90	RS	V75μ	V100μ	V150μ	V200μ	V250μ
rpm and spray parameters	-	−0.827 **	−0.96 **	−0.908 **	0.059	0.889 **	0.925 **	0.978 **	0.943 **	0.6 *
spray parameters and pollen activity	−0.943 **	0.847 **	0.925 **	0.824 **	−0.16	−0.96 **	−0.952 **	−0.908 **	−0.837 **	−0.503

Note: * at the 0.05 level (two-tailed) with significant correlation, ** at the 0.01 level (two-tailed) with significant correlation.

The correlations of different droplet sizes and pollen activity were established by SPSS (Table 5). It can be shown that droplets smaller than 100 μm are the main cause of reduced pollen activity. Furthermore, the effects of droplets with sizes greater than 150 μm and less than 250 μm on pollen activity were significantly positively correlated. The effects of droplets with sizes greater than 100 μm and less than 150 μm on pollen activity were not significantly correlated but were positively correlated.

Table 5. Correlation analysis between pollen suspension droplet size and pollen activity. The different intervals of pollen suspension droplet size were divided to be used for correlation analysis with pollen activity.

Types (μm)	75–100	75–150	75–200	75–250	100–150	100–200	100–250	150–200	150–250	200–250
Correlation	−0.785 **	−0.42	−0.16	0.073	0.045	0.15	0.336	0.569 *	0.793 **	0.921 **

Note: * at the 0.05 level (two-tailed) with significant correlation, and ** at the 0.01 level (two-tailed) with significant correlation.

3.7. Influence of UAVS Downwash Airflow on the Pollen Activity

The UAVS downwash airflow had no significant influence on pollen activity but had a significant influence on the CV value of pollen activity (Figure 6). The pollen activity received by spraying in the downwash airflow with the No. 01 ordinary flat-fan nozzle and RA with 8000 rpm was 5.93% and 7.52% lower than the pollen activity from spraying on the ground, respectively. Compared to pollen that did not experience the following wash airflow field, the CV value of the pollen activity was raised by 128.72% and 40.67% for the No. 01 ordinary flat-fan nozzle and RA (8000 rpm), respectively.

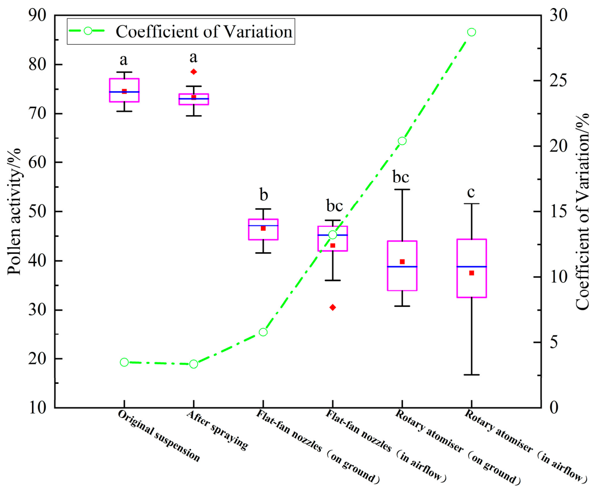


Figure 6. Results of the influence of the downwash airflow field of the UAVS on the pollen activity. The pollen suspension after the spraying was completed was used as the control group. Additionally, the pollen activity in the pollen suspension received when the UAVS was on the ground was compared with the pollen activity in the pollen suspension received in the UAVS downwash airflow field. Different letters indicate significant differences (Duncan test, α = 0.05).

4. Discussion

Pollination results are seriously affected by pollen application methods [23]. At present, LSP is an efficient and labor-saving pollination method [24]. The pollen activity of LSP is influenced by the pollen storage time [25–29]. Moreover, the pH value [13] and ions [30] in the pollen suspension have an impact on the pollen activity. Hence, the storage time of pollen suspension is likely to affect the pollen activity. In this study, the storage time of pollen suspensions had a significant influence on pollen activity (Figure 5a), and pollen activity decreased with increasing storage time of pollen suspension. In addition, there was a significant difference between the pollen activity after 20 min of storage and that after 0 min of storage. This explains why some literature specifies that the pollen suspension should be sprayed as soon as possible onto the flower stigma [13]. At the same time, the pollen activity in pollen suspensions remained stable for a period of time and then decreased rapidly with increasing storage time. The CV value of pollen activity decreased and then increased with increasing storage time. This may be because the short proper temperature makes the pollen grains in the pollen suspension more active, which gradually reduces the CV value of pollen activity. Furthermore, the osmotic pressure of the pollen grains in the pollen suspension is partly unbalanced with increasing storage time, resulting in an increase on CV values. . . To ensure pollen activity, the pollen suspension was sprayed as quickly as possible within 20 min of configuration.

There was a significant difference between the sprayer parameters (except nozzle size (Figure 5d) and pollen activity (Figure 5b–f). In this study, when the spraying system (Figure 2a) was operated for five minutes, the pollen suspension recirculated into the tank through the recirculation device, and its pollen activity was significantly reduced (Figure 5b). The recirculated pollen suspension passed through the pump and the recirculation device (pressure regulator valve and overflow pipeline), whereas the pump significantly reduced the pollen activity in the pollen suspension (Figure 5c). Therefore, the pollen activity of pollen suspensions applied by the pump and the pollen activity of pollen suspensions in the tank were significantly different, regardless of whether the sprayer had a recirculation device [31,32]. Our study showed that prolonged use of the recirculation device would cause a significant decrease in pollen activity. Some studies have shown that LSP is not effective, especially when pollination is carried out by large ground-based sprayers. It is reasonable to assume that they use a recirculation device to recirculate pollen suspension into the tank for regulating the pressure and stirring the pollen suspension [32]. Additionally, the large tanks require a lot of time (far over 20 min) for spraying. These two factors contribute to this phenomenon. However, the LSP tests did not experience the aforementioned phenomena using air can sprayers and knapsack hand sprayer (no recirculation device) [13]. Based on our research, we speculate that it is likely that the prolonged use of the recirculation device caused the above phenomenon.

Additionally, there was a significant difference in the pump type on the pollen activity. Although there was no significant difference between small air can sprayer or middle air can sprayer and original pollen suspension, it also significantly reduced pollen activity (Figure 5b). This was because their low pump pressure (Table 2) did not significantly disrupt the biochemical properties of the pollen grains. With the increase in air compression pump pressure, there was a significant difference between the large air can sprayer and the original pollen suspension. This indicated that the higher pressure of the air compression pump had a greater effect on the pollen activity. At the same time, knapsack hand sprayer with leather cup-type piston pumps was significantly different from homemade orchard sprayer (with plunger pump, Figure 5b). This was because the leather cup-type piston pumps worked similar to plunge pumps and air compression pumps, but their pressure range was much smaller than that of an electric plunge pump. Additionally, the plunger pump had a wider pressure range and a higher rpm and pressure (Table 2). The pumps of all three electric sprayers were electric diaphragm pumps, which had a limited range of pressure variation and lower rpm and pressure (Table 2). This may be the reason why they are not significantly different from the knapsack hand sprayer (Figure 5b). This also explains why, regardless of the suspension media utilized, the pollen activity that was sprayed showed a significant reduction [13,32]. However, at low pressure, the CV value of pollen activity was high for the plunger pump. This explains why diaphragm pumps or diaphragm-like pumps are used in commercial LSP sprayers. This is convincing evidence of our research, and every type of diaphragm pump we investigated is the most suitable pump for commercial LSP [32].

The size of the nozzle had no discernible influence on the pollen activity at 0.3 MPa. The droplets were only broken up by shear force when using the flat-fan nozzle. The small revolution and shearing forces produced had no impact on the biochemical properties of pollen. There was no significant effect of spray pressure (0.3 to 2.5 MPa) on pollen activity at spray pressure. LSP employing high-pressure sprayers (plunger pumps with spraying pressures between 2.0 and 3.0 MPa) did not significantly affect the fruit sitting rate in some countries of the Middle East [33]. Additionally, the CV value of pollen activity was even smaller for plunger pumps at greater than 2.0 MPa. As a result, it is feasible for high-pressure sprayers to be used for LSP. When enough pollen suspension droplets are made to fall on the flower stigma, this amount is sufficient to compensate for the absence of pollen activity. However, most of the sprayers in the Middle East are manually operated sprayers where people consciously spray on the flowers, which can compensate for the lack of pollen activity [16,33]. To reach a specific fruit setting rate, this strategy has been shown to consume more pollen than the typical spraying method [34]. According to our study, high-pressure spraying of pollen suspensions using plunger pumps is not recommended. A RA is a type of uniform spraying nozzle. It is frequently employed in UAVS operations for plant protection. The shear force on the produced droplets increases as RA's rotation speed increases, which results in smaller droplets size [35]. However, its use for LSP has rarely been reported. The size of the sprayed droplets decreases with increasing brushless motor speed in the RA. This is because the centrifugal force and shear force on the pollen suspension also increase when the speed of the brushless motor increases. The pollen grains are impacted by this. The centrifugal and shear pressures produced by brushless motor speeds greater than 12,000 rpm may be larger than what the majority of pollen grains can sustain, having an irreversible influence on pollen grains. Therefore, for LSP, we recommend that the RA speed be between 4000 rpm and 12,000 rpm.

UAVSs are frequently employed in orchards [20], and research on LSP has also been carried out [36]. Yet, different from the ordinary spraying, it has a strong downwash airflow [37]. There is evidence that aerodynamic shear has a certain impact on pollen germination [38]. There was no significant difference between the UAVS downwash airflow and pollen activity (Figure 6). However, it increased the CV value of pollen activity. This could be a result of the downwash airflow field's increased shear force on the pollen suspension droplets, which would change some biochemical characteristics of pollen grains. Additionally, the solution in the tank can be sprayed on the target canopy by UAVS within 15 min.

Therefore, it is recommended to design LSP sprayers with fast pollen suspension spraying, preferably using a diaphragm pump without a recirculation device. Thus far, the UAVSs meet the above conditions, and they are suitable for LSP of fruit trees. Nevertheless, the fruit setting mechanism by which UAVSs are used for LSP is unclear. Thus, the LSP of UAVS should be studied based on LSP parameters in the future, ensuring pollen activity in the commercial LSP of UAVS.

5. Conclusions and Future Arrangement

In this study, pollen activity is affected by different LSP parameters, excluding nozzle size and UAVS downwash airflow. The influences of different parameters on pollen activity are as follows:

(1) The storage time of the pollen suspension and the mean pollen activity were extremely significantly negatively correlated ($r = -0.98$). After 20 min of storage, the pollen

activity of the pollen suspension was significantly different from that of the original pollen suspension, and its activity decreased by 10.06%.

(2) Different nozzle sizes had no significant effect on pollen activity. There was no significant difference between the pollen activity in the tank (D3 and P3) without recirculation device and the original pollen suspension, whereas the pollen activity in the tank (D1 and P1) with recirculation device did. The different spraying pressure of the same pump was negatively and highly significantly correlated with the pollen activity and its CV value ($r = -0.951$ and $r = -0.936$). However, there was no significant difference in pollen activity between different spraying pressures. Different types of pumps had significant effects on pollen activity and decreased with the increase of pump pressure range. There was a significant correlation between the RA revolution speed and pollen activity (droplet parameters). The mean pollen activity decreased from 44.25% to 14.14% as the rpm of RA increased from 3000 rpm to 14,000 rpm.

(3) The UAVS downwash airflow had no significant influence on pollen activity but had a significant influence on the CV value of pollen activity. Compared with the pollen suspension sprayed on the ground, the CV value of pollen activity sprayed by UAV in the air increased by at least 40.67%.

Thus, the pollen suspension should be configured quickly and sprayed onto the stigma as quickly as possible, whereas the time is preferably within 20 min. The sprayers should use an electric diaphragm pump (with low pump pressure) and not carry a recirculation device. Since UAVSs satisfy the aforementioned requirements, they are recommended for LSP. Additionally, the rpm of the RA should preferably be between 4000 rpm and 12,000 rpm. This study provides a reliable theoretical basis for optimizing and advancing pear LSP technology. Based on the above research results, we will conduct orchard pollination operations to explore the impact of optimized LSP parameters on pollination fruit setting rate. At the same time, the effects of liquid shear force and aerodynamic shear force on pollen activity will be studied in view of the current theoretical deficiencies.

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References

1. Garibaldi, L.A.; Carvalheiro, L.G.; Leonhardt, S.D.; Aizen, M.A.; Blaauw, B.R.; Isaacs, R.; Kuhlmann, M.; Kleijn, D.; Klein, A.M.; Kremen, C.; et al. From research to action: Enhancing crop yield through wild pollinators. *Front. Ecol. Environ.* **2016**, *12*, 439–447. [CrossRef]
2. Castro, H.; Siopa, C.; Casais, V.; Castro, M.; Loureiro, J.; Gaspar, H.; Castro, S. Pollination as a key management tool in crop production: Kiwifruit orchards as a study case. *Sci. Hortic.* **2021**, *290*, 110533. [CrossRef]
3. Potts, S.G.; Imperatriz-fonseca, V.; Ngo, H.T.; Biesmeijer, J.C.; Breeze, T.D.; Dicks, L.V.; Garibaldi, L.A.; Hill, R.; Settele, J.; Vanbergen, A.J.; et al. Summary for Policymakers of the Thematic Assessment on Pollinators, Pollination and Food Production Intergovernmental Science-Policy Platform on Biodiversity and Ecosystem Key Messages Values of Pollinators and Pollination. *Biota Neotrop.* **2016**, *16*, e20160101.
4. Eyles, A.; Close, D.C.; Quarrell, S.R.; Allen, G.R.; Spurr, C.J.; Barry, K.M.; Whiting, M.D.; Gracie, A.J. Feasibility of Mechanical Pollination in Tree Fruit and Nut Crops: A Review. *Agronomy* **2022**, *12*, 1113. [CrossRef]
5. Barrera, W.B., Jr.; Trinidad, K.A.D.; Presas, J.A. Hand pollination and natural pollination by carpenter bees (*Xylocopa* spp.) in *Passiflora edulis* Sims. f. *flavicarpa* Deg. (yellow passion fruit). *J. Apic. Res.* **2020**, *60*, 845–852. [CrossRef]
6. Traynor, K.S.; Mondet, F.; de Miranda, J.R.; Techer, M.; Kowallik, V.; Oddie, M.A.; Chantawannakul, P.; McAfee, A. Varroa destructor: A Complex Parasite, Crippling Honey Bees Worldwide. *Trends Parasitol.* **2020**, *36*, 592–606. [CrossRef] [PubMed]
7. Ren, Z.; Zhao, Y.; Liang, H.; Tao, Z.; Tang, H.; Zhang, H.; Wang, H. Pollination ecology in China from 1977 to 2017. *Plant Divers* **2018**, *40*, 172–180. [CrossRef] [PubMed]
8. Ryalls, J.M.; Langford, B.; Mullinger, N.J.; Bromfield, L.M.; Nemitz, E.; Pfrang, C.; Girling, R.D. Anthropogenic air pollutants reduce insect-mediated pollination services. *Environ. Pollut.* **2022**, *297*, 118847. [CrossRef]
9. Yousefi, M.; Mohd Rafie, A.S.; Abd Aziz, S.; Azrad, S.; ABD Razak, A.b. Introduction of Current Pollination Techniques and Factors Affecting Pollination Effectiveness by Elaeidobius Kamerunicus in Oil Palm Plantations on Regional and Global Scale: A Review. *S. Afr. J. Bot.* **2020**, *132*, 171–179. [CrossRef]
10. Reitmayer, C.M.; Ryalls, J.M.W.; Farthing, E.; Jackson, C.W.; Girling, R.D.; Newman, T.A. Acute exposure to diesel exhaust induces central nervous system stress and altered learning and memory in honey bees. *Sci. Rep.* **2019**, *9*, 5793. [CrossRef]
11. Gianni, T.; Vania, M. Artificial Pollination in Kiwifruit and Olive Trees. In *Pollination in Plants*; IntechOpen: Cambridge, UK, 2018. [CrossRef]
12. Wurz, A.; Grass, I.; Tscharntke, T. Hand pollination of global crops—A systematic review. *Basic Appl. Ecol.* **2021**, *56*, 299–321. [CrossRef]
13. Sakamoto, D.; Hayama, H.; Ito, A.; Kashimura, Y.; Moriguchi, T.; Nakamura, Y. Spray pollination as a labor-saving pollination system in Japanese pear (*Pyrus pyrifolia* (Burm.f.) Nakai): Development of the suspension medium. *Sci. Hortic.* **2009**, *119*, 280–285. [CrossRef]
14. Hayati, A.; Marzban, A.; Rahnama, M. Occupational safety and health in traditional date palm works. *Int. Arch. Occup. Environ. Health.* **2021**, *94*, 1455–1473. [CrossRef] [PubMed]
15. Mokdad, M.; Bouhafs, M.; Lahcene, B.; Mokdad, I. Ergonomic practices in Africa: Date palm work in Algeria as an example. *Work* **2019**, *62*, 657–665. [CrossRef] [PubMed]
16. Dahab, M.H.; Osman, M.S.; Abdalla, O.A. Comparison of Locally Developed Date Palm Pollination Machine with Manual Pollination under Shambat Condition—Sudan. *Asian J. Agric. Hortic. Res.* **2020**, 16–25. [CrossRef]
17. Shi, F.; Jiang, Z.; Ma, C.; Zhu, Y.; Liu, Z. Controlled Parameters of Targeted Pollination Deposition by Air-Liquid Nozzle. *Trans. Chin. Soc. Agric. Mach.* **2019**, *50*, 115–124. [CrossRef]
18. Xiongkui, H.; Bonds, J.; Herbst, A.; Langenakens, J. Recent development of unmanned aerial vehicle for plant protection in East Asia. *Int. J. Agric. Biol. Eng.* **2017**, *10*, 18–30. [CrossRef]
19. He, X. Rapid Development of Unmanned Aerial Vehicles (UAV) for Plant Protection and Application Technology in China. *Outlooks Pest Manag.* **2018**, *29*, 162–167. [CrossRef]
20. Wang, C.; Liu, Y.; Zhang, Z.; Han, L.; Li, Y.; Zhang, H.; Wongsuk, S.; Li, Y.; Wu, X.; He, X. Spray performance evaluation of a six-rotor unmanned aerial vehicle sprayer for pesticide application using an orchard operation mode in apple orchards. *Pest Manag. Sci.* **2022**, *78*, 2449–2466. [CrossRef]
21. Montilla-Pacheco, A.D.J.; Pacheco-Gil, H.A.; Pastrán-Calles, F.R.; Rodríguez-Pincay, I.R. Pollination with drones: A successful response to the decline of entomophiles pollinators? *Sci. Agropecu.* **2021**, *12*, 509–516. [CrossRef]
22. Li, J.Y.; Lan, Y.B.; Wang, J.W.; Chen, S.; Huang, C.; Liu, Q.; Liang, Q. Distribution Law of Rice Pollen in the Wind Field of Small UAV. *Int. J. Agric. Biol. Eng.* **2017**, *10*, 32–40. [CrossRef]
23. Oh, E.U.; Kim, S.C.; Lee, M.H.; Song, K.J. Pollen Application Methods Affecting Fruit Quality and Seed Formation in Artificial Pollination of Yellow-Fleshed Kiwifruit. *Horticulturae* **2022**, *8*, 150. [CrossRef]
24. Naik, S.; Rana, V. Spray pollination: An efficient and labour saving method for kiwifruit (*Actinidia deliciosa* A. Chev.) production. *J. Appl. Hortic.* **2013**, *15*, 202–206. [CrossRef]
25. Wang, L.; Wu, J.; Chen, J.; Fu, D.; Zhang, C.; Cai, C.; Ou, L. A simple pollen collection, dehydration, and long-term storage method for litchi (*Litchi chinensis* Sonn.). *Sci. Hortic.* **2015**, *188*, 78–83. [CrossRef]
26. Wang, X.; Wu, Y.; Lombardini, L. In vitro viability and germination of Carya illinoinensis pollen under different storage conditions. *Sci. Hortic.* **2020**, *275*, 109662. [CrossRef]

27. Mauriello, G.; De Prisco, A.; Di Prisco, G.; La Storia, A.; Caprio, E. Microbial characterization of bee pollen from the Vesuvius area collected by using three different traps. *PLoS ONE* **2017**, *12*, e0183208. [CrossRef]
28. Filho, J.; Torres, A.; Nascimento, W.; Boiteux, L. Tomato hybrid seed production using stored pollen grains: Temperature and storage time interactions and their effects on pollen viability. *Acta Hortic.* **2019**, *1249*, 201–204. [CrossRef]
29. Kadri, K.; Elsafy, M.; Makhlouf, S.; Awad, M.A. Effect of pollination time, the hour of daytime, pollen storage temperature and duration on pollen viability, germinability, and fruit set of date palm (*Phoenix dactylifera* L.) cv "Deglet Nour". *Saudi J. Biol. Sci.* **2021**, *29*, 1085–1091. [CrossRef]
30. Dinis, A.M.; Mesquita, J.F. Uptake of heavy metal salt solutions by pollen grains of *Magnolia soulangeana*(Magnoliaceae). *Can. J. Bot.* **2004**, *82*, 1758–1767. [CrossRef]
31. Mostaan, A.; Marashi, S.S.; Ahmadizadeh, S. Development of a New Date Palm Pollinator. In Proceedings of the IV International Date Palm Conference, Abu Dhabi, United Arab Emirates, 15–17 March 2010; Volume 882.
32. Williams, H.; Nejati, M.; Hussein, S.; Penhall, N.; Lim, J.Y.; Jones, M.; Bell, J.; Ahn, H.S.; Bradley, S.; Schaare, P.; et al. Autonomous pollination of individual kiwifruit flowers: Toward a robotic kiwifruit pollinator. *J. Field Robot.* **2019**, *37*, 246–262. [CrossRef]
33. Munir, M. Influence of liquid pollination technique on fruit yield and physicochemical characteristics of date palm cultivars Khadrawy and Zahidi. *J. Appl. Hortic.* **2020**, *22*, 12–17. [CrossRef]
34. Cacioppo, O.; Michelotti, V.; Vittone, G.; Tacconi, G. Pollination of kiwifruit: 30 years of applied research leads to a model system for studying the interaction between pollination and flowering stage. *Acta Hortic.* **2018**, *1229*, 355–364. [CrossRef]
35. Craig, I.P.; Hewitt, A.; Terry, H. Rotary atomiser design requirements for optimum pesticide application efficiency. *Crop. Prot.* **2014**, *66*, 34–39. [CrossRef]
36. Yang, J.; Yang, H.; Xie, W.; Hao, H.; Feng, H.; Wang, L. Parameter Optimization and Economic Analysis of Unmanned Aerial Vehicle (UAV) Assisted Liquid Pollination for Kuerlexiangli Pear. *J. Fruit Sci.* **2021**, *38*, 2561. [CrossRef]
37. Yang, F.; Xue, X.; Cai, C.; Zhou, Q. Effect of down wash airflow in hover on droplet motion law for multi–rotor unmanned plant protection machine. *Trans. Chin. Soc. Agric. Eng.* **2018**, *34*, 64–73. [CrossRef]
38. Law, S.; Wetzstein, H.; Banerjee, S.; Eisikowitch, D. Electrostatic application of pollen sprays: Effects of charging field intensity and aerodynamic shear upon deposition and germinability. *IEEE Trans. Ind. Appl.* **2000**, *36*, 998–1009. [CrossRef]