Available online at http://bjas.bajas.edu.iq
https://doi.org/10.37077/25200860.2025.38.sp.11
College of Agriculture, University of Basrah

Basrah Journal of Agricultural Sciences

ISSN 1814 – 5868 Basrah J. Agric. Sci., 38(Special Issue), 130-140,2025 E-ISSN: 2520-0860

Estimate the Content of Aromatic Oils in Pollen for Five Male Cultivars of Data Palm *Phoenix dactylifera* L. Treated by Fanphthyl Acetamide

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Received 22nd October 2024; Accepted 22nd February 2025; Available online 24th August 2025

Abstract: The university of Basrah will estimate the pollen content from aromatic oils for five male cultivars of date palms and five male cultivars of date palms to find their content of aromatic oils. Male cultivares were selected (Ghanami Akhdar, Ghanamiahmar, Khakri, Wardi and Semismi). The study included pollen treatment with growth regulator fanphthyl acetamide with concentrations (0, 0.1, 0.3 g. L-1). The process of extracting aromatic oils from pollen was performed using the clavenger apparatus. The findings of the study indicated the preeminence of the Ghanami Akhdar. Male cultivar over the rest of the cultivars in all qualitative specifications of aromatic oils, the Wardi male cultivar had the minimal value of all such qualities. The study's results demonstrated the impact of the growth regulator fanphthylacetamidein on the qualitative attributes of volatile oils of pollen by improving all qualitative qualities of volatile oils of the percentage of essential oil (0.041. 0.039. 0.035. 0.030. 0.027) % respectively. The oil density (0.940. 0.915. 0.885. 0.860. 0.810) ml^{-1} sequentially and of essential oil (0.92,0.9,0.87,0.85, 0.82) espectively and PH essential oil (5.94,5.90,5.85,5.81,5.81) respectively number for crucial oil (178.76, 174.63, 171.87, 168.98, 165.34) respectively and all the male varieties under consideration.

Keywords: Aromatic oils, Essential oils, Ghanami, Growth regulator, Pollen grain.

Introduction

The date palm is a significant species within the Arecaceae family, with over 200 genera and 2,500 species. The most advantageous plant family for humans, after the Gramineae family, is also noted. The date palm is classified under the genus Phoenix and the species dactylifera (Saleh *et al*.,2023; Jain *et al*., 2011; EL-Hadrami & EL-Hadrami, 2009). The palm of date is dioecious and unisexual, indicating that male flowers are produced on one tree and female blooms on another, as pollen grains are crucial for fruit development (Abdul Qadir, 2021; Darhab, 2004). Interest in studying pollen has increased is a direct cause of date palm fruiting and because it contains good chemical compounds beneficial to humans thus, it is an integrated food substance. The chemical composition of pollen is a basic indicator for determining the

fertility of the male date palm in addition to the flowering date, the number of inflorescences, the mass of the inflorescence and the number of inflorescences present in one inflorescence (Alpresem *et al*.,2025; Aly, 2018; Kavand *et al*., 2014; Liu *et al*., 2013).

Pollen plays an important role in helping the ovaries to facilitate their development, and stimulate the ovaries to secrete hormones that initiate the process of auxin production from tryptophan, it is also a stimulating factor for the existing auxin (Haddad & Byerly, 2009). There is a significant effect of the male cultivar in increasing the yield of essential oil in pollen grains and the refractive index of essential oil, and pollen grains of different varieties of date palms differ in terms of the specific gravity of their essential oil, pH value and saponification number (Al-Tamimi, 2020).

Growth regulators are organic compounds other than nutrients, and they work to promote, inhibit, or modify physiological processes when used in small concentrations. They may be formed naturally in the plant or manufactured in the laboratory; they also play a significant role in the plant's reaction to external environmental stimuli. Auxins are the primary and most important category of plant growth regulators, since they irreversibly enhance development along the longitudinal axis when applied at low concentrations (Abdullah *et al*.,2025; Trivellini *et al*., 2015). Auxins govern numerous growth and maturation processes, promote cell division and elongation, induce rooting, enhance pollen germination and tube growth, improve fruit set and development, and facilitate parthenogenesis in plants by augmenting the transport of nutrients and essential substances (Sotomayor *et al*., 2012). Fanphthyl acetamide is a plant growth regulator that enhances the efficacy of other plant hormones and augments pollen viability, fertility, and chemical composition. It stabilizes flowers and increases their setting and fruit formation, producing prolific output. It further safeguards the plant against cold and elevated temperatures during pollination, fertilization, and fruit development. It is a useful plant growth regulator for fruit trees that struggle with fruit setting and development, it is a useful plant growth regulator (Al-Mahmoudi *et al*., 2023).

Materials & Methods

This research was performed in the Medicinal and Aromatic Plants Unit of the College of Agriculture/University of Basrah on pollen grains for five male varieties of date palms to determine their content of aromatic oils. The selected male cultivars: Ghanami Akhdar, Ghanami Ahmar, Khakri, Wardi, and Semismi.

Pollen extraction: The inflorescences were carefully harvested from the spathe of date palms using a sharp knife. Afterward, each cultivar was placed in separate, enclosed rooms to prevent cross-contamination, ensuring the samples remained distinct. Once the inflorescences had dried, the pollen grains were released into a paper bag by gently shaking them. The collected pollen was then sifted to remove any remaining fragments of the inflorescences. The study included treating the pollen grains with the growth regulator fanphthyl acetamide at concentrations (0, 0.1and 0.3 g L^{-1}).

Essential oil extraction: crucial oil extraction: The essential oil was extracted from the pollen grains using petroleum ether, following the method described by Al-Najjar *et al*., (2021).

Percentage of essential oil: The percentage of essential oil was determined using the equation outlined in Ansari *et al*., (2024) which is:

Percentage of essential oil (%) = (Output oil weight (mg)/ Sample Weight (mg)) × 10 **The specific gravity of crucial oil:** By placing 100 µs of essential oil in a precise volumetric pipette and weighing the volume using a sensitive Mettler balance with four decimal places after separation, the specific gravity of each oil sample from each treatment was calculated. By dividing the weight of that volume of oil by the Weight of the same volume of distilled water at the same temperature, the specific gravity values were determined using three measurements from each sample at a temperature of 20 °C. According to the following equation:

Specific gravity of essential oil = Weight of 100 µL of essential oil/ Weight of 100 µl of distilled water.

Density of essential oil (mg μ^{-1}) Density: By dividing the weight of 100 µs of oil by its volume (100 µs), the density of each oil sample was determined.

Refractive index of essential oil: The proportion of essential oil was determined using the formula outlined in Muhammad (2016), which is:

PH of crucial oil**:** The PH of essential oil samples was measured using a pH meter at 25°C.

Saponification number of essential oils: It was measured according to the method described by Al-Tamimi (2000) by weighing 0.025 g of essential oil, adding 3 ml of 0.5 N alcoholic potassium hydroxide solution, an air condenser was placed on the mouth of the flask, and put in a water bath at boiling point for half an hour. After cooling the flask, five drops of phenolphthalein indicator were added, the alkalinity of the solution was adjusted with hydrochloric acid (0.5 N), and the first reading (A) was recorded. Then the previous steps were carried out without oil and according to the number of centimeters of hydrochloric acid needed to neutralize the solution, and the second reading (B) was recorded according to the saponification number according to the following equation:

Saponification number of essential oil=((A-B) *28.05/Sample weight) *100

Statistical analysis: A Randomized Complete Block Design (RCBD) was used for factorial experiments, with two factors: the first factor being the male cultivars (five cultivars) and the second factor being the treatment with the growth regulator fanphthyl acetamide at concentrations of (0, 0.1, 0.3 g L^{-1}) with three replicates, thus the number of experimental units reaching 45 units. Statistical analysis was conducted using the GenStat statistical program to analyze variance. As for the comparison between means, the least significant difference (L.S.D.) was used at a probability level 0.05 based on Tabiah (2008).

Results & Discussion

Percentage of essential oil (%)

The results shown in Table (1) indicate the pollen content of the five male cultivars treated with the growth regulator fanphthyl acetamide in terms of the percentage of essential oil, as the male cultivar Ghanami Akhdar significantly outperformed the rest of the cultivars and recorded the highest rate of 0.037%, In contrast the male cultivar Wardi recorded the lowest value of 0.022%. As for the effect of the growth regulator, the level of

0.3 g L^{-1} recorded the highest percentage of essential oil at 0.034%, while the amount of 0 mg L^{-1} recorded the lowest value of 0.025%.
As for the interaction between the male cultivars and the growth regulator, the interaction between the Ghanami Akhdar cultivar and the growth regulator at a level of 0.3 g L^{-1} recorded the highest percentage of essential oil, reaching 0.041%, The interaction between the male cultivar Wardi and a level of 0 g L^{-1} of the growth regulator yielded the lowest value, at 0.017%. Applying a growth regulator at a level of 0.3 g L^{-1} to pollen grains from the male cultivar Wardi resulted in a 0.027% increase in essential oil content.

Table (1): The effect of male cultivar and growth regulator fanphthyl acetamide on the percentage of essential oil.

Male cultivars	fanphthyl acetamide			Mean of Male cultivars
	0 g L^{-1}	0.1 g L^{-1}	0.3 g L^{-1}	
GhanamiAkhdar	0.033	0.037	0.041	0.037
Ghanamiahmar	0.030	0.035	0.039	0.035
Khakri	0.025	0.030	0.035	0.030
Semismi	0.021	0.026	0.030	0.026
Wardi	0.017	0.023	0.027	0.022
Mean of fanphthyl acetamide	0.025	0.030	0.034	
L.S.D.	Male cultivars=0.003	fanphthyl acetamide = 0.002		Interaction= 0.005

The specific gravity of essential oil

The results in Table (2) indicate the specific gravity of the essential oil in pollen grains of the five male cultivars treated with the growth regulator fanphthyl acetamide, as the male cultivar Ghanami Akhdar significantly outperformed the rest of the cultivars and recorded the highest value of 0.880, In contrast the male cultivars Wardi recorded the lowest value of 0.743. The impact of the growth regulator at 0.3 g L^{-1} recorded the highest value for the specific gravity of 0.872, while "the 0 g L^{-1} recorded the lowest value of 0.750. The interaction between the male cultivar and the growth regulator showed that the Ghanami Akhdar cultivars, when treated with a growth regulator at a level of 0.3 g L^{-1}, achieved the highest specific gravity of the essential oil, was measured at 0.920, At the same time the interaction between the male cultivars Wardi and an amount of 0 g L^{-1} of the growth regulator recorded the lowest value, reaching 0.660. The treatment of the male cultivars Wardi pollen grains with the growth regulator at 0.3 g L^{-1} resulted in an increase in the specific gravity of the essential oil by a percentage of 0.820.

Density of essential oil (mg μ^{-1})

The data in Table (3) demonstrate the influence of the male cultivar and treatment with the growth regulator fanphthyl acetamide on the essential oil density in the pollen grains of the five cultivars. The Ghanami Akhdar cultivar significantly surpassed the others, achieving the greatest value of 0.903 mg μL^{-1}, whereas the Wardi cultivar exhibited the smallest value of 0.743 mg μ^{-1}.

Table (2): The effect of male cultivar and growth regulator fanphthyl acetamide on the percentage of Specific gravity of essential oil.

Male cultivars	fanphthyl acetamide			Mean of Male cultivars
	0 g L^{-1}	0.1 g L^{-1}	0.3 g L^{-1}	
GhanamiAkhdar	0.830	0.890	0.920	0.880
Ghanamiahmar	0.790	0.870	0.900	0.853
Khakri	0.750	0.840	0.870	0.820
Semismi	0.720	0.810	0.850	0.793
Wardi	0.660	0.750	0.820	0.743
Mean of fanphthyl acetamide	0.750	0.832	0.872	
L.S.D.	Male cultivars =0.053	fanphthyl acetamide=0.035		Interaction = 0.072

The application of the growth regulator at a level of 0.3 g L^{-1} yielded the maximum essential oil density at 0.882 mg μ^{-1}, whilst a level of 0 g L^{-1} resulted in the lowest density at 0.768 mg μ^{-1}. The interaction between the Ghanami Akhdar cultivar and a growth regulator at a level of 3 mg L^{-1} yielded the highest essential oil density, measuring 0.940 mg μ^{-1}. Conversely, the interaction involving the Wardi cultivar with 0 g L^{-1} of the growth regulator resulted in the lowest density, recorded at 0.670 mg μ^{-1}, demonstrating a significant difference from other interactions. Applying of a growth regulator at a level of 0.3 g L^{-1} to the pollen grains of the Wardi cultivar led to a 0.810 mg μ^{-1} increase in essential oil density.

Table (3): The effect of male cultivar and growth regulator fanphthyl acetamide on the percentage of Density of essential oil (mg μ^{-1}).

Male cultivars	fanphthyl acetamide			Mean of Male cultivars
	0 g L^{-1}	0.1 g L^{-1}	0.3 g L^{-1}	
GhanamiAkhdar	0.870	0.900	0.940	0.903
Ghanamiahmar	0.820	0.860	0.915	0.865
Khakri	0.770	0.820	0.885	0.825
Semismi	0.710	0.780	0.860	0.783
Wardi	0.670	0.750	0.810	0.743
Mean of fanphthyl acetamide	0.768	0.822	0.882	
L.S.D.	Male cultivars= 0.068	fanphthyl acetamide0.047 =		Interaction =0.087

Refractive index of essential oil

The results in Table (4) show the effect of the male cultivar and treatment with the growth regulator fanphthyl acetamide on the refractive index of essential oil. The Ghanami Akhdar cultivar significantly outperformed the rest of the cultivars and recorded the highest value of 1.566, while the Wardi cultivar recorded the lowest value of 1.447. The impact of the growth regulator at the level of 0.3 g L^{-1} recorded the highest refractive index of the essential oil, reached 1.536, while the level of 0 g L^{-1} recorded the lowest value of 1.474. As for the interaction between the male cultivar and the growth regulator, the interaction between the Ghanami Akhdar cultivar and the growth regulator at an amount of 3 m g L^{-1} recorded the highest refractive index of the essential oil, which reached 1.587, while the interaction between the Wardi cultivar and the 0 g L^{-1} and a level of the growth regulator recorded the lowest value of 1.410.

Table (4): The effect of male cultivar and growth regulator fanphthyl acetamide on the percentage of Refractive index of essential oil.

Male cultivars	fanphthyl acetamide			Mean of Male cultivars
	0 g L^{-1}	0.1 g L^{-1}	0.3 g L^{-1}	
GhanamiAkhdar	1.540	1.570	1.587	1.566
Ghanamiahmar	1.510	1.550	1.564	1.541
Khakri	1.470	1.510	1.535	1.505
Semismi	1.440	1.480	1.515	1.478
Wardi	1.410	1.450	1.480	1.447
Mean of fanphthyl acetamide	1.474	1.512	1.536	
L.S.D.	Male cultivars=0.023	fanphthyl acetamide=0.012		Interaction = 0.032

PH of essential oil

The results in Table 5 indicate the effect of the male cultivar and treatment with growth regulator fanphthyl acetamide on the PH of the essential oil of pollen grains of the five male varieties treated with the growth regulator fanphthyl acetamide, as the Ghanami Akhdar cultivar significantly outperformed the rest of the varieties and recorded the highest value of 5.883, while the Wardi cultivar recorded the lowest value of 5.637. As for the effect of the growth regulator, the level of 0.3 g L^{-1} recorded the highest value for the PH of the essential oil, which reached 5.862, In contrast the level of 0 g L^{-1} recorded the lowest value, which reached 5.626. As for the interaction between the male cultivar and the growth regulator, the interaction between the Ghanami Akhdar cultivar and the growth regulator at an amount of 0.3 g L^{-1} recorded the highest value of the essential oil PH, which reached 5.94, while the interaction between the male cultivar Wardi and amount of 0 g L^{-1} of the growth regulator recorded the lowest value, which reached 5.40, with a significant difference from the rest of the interactions. The treatment of pollen grains of the male cultivar Wardi with the growth regulator at 3 m g L^{-1} caused an increase in the essential oil PH by 5.81.

Saponification number of essential oils

Table (6) presents the impact of the male cultivar and treatment with the growth regulator fanphthyl acetamide on the saponification number of essential oils in the pollen of five male cultivars. The Ghanami Akhdar cultivar markedly surpassed all other cultivars, achieving the greatest score of 174.213, whilst the Wardi cultivar registered the lowest value of 158.560. The growth regulator at an A level of 0.3 g L^{-1} yielded the greatest saponification number of essential oils, measuring 171.916, a 0 g L^{-1} level yielded the lowest recorded value of 161.4.

Table (5): The effect of male cultivar and growth regulator fanphthyl acetamide on the PH of essential oil.

Male cultivars	fanphthyl acetamide			Mean of Male cultivars
	0 g L^{-1}	0.1 g L^{-1}	0.3 g L^{-1}	
GhanamiAkhdar	5.81	5.90	5.94	5.883
Ghanamiahmar	5.77	5.85	5.90	5.840
Khakri	5.65	5.81	5.85	5.770
Semismi	5.50	5.77	5.81	5.693
Wardi	5.40	5.70	5.81	5.63-7
Mean of fanphthyl acetamide	5.626	5.806	5.862	
L.S.D.	Male cultivars =0.045	fanphthyl acetamide =0.032		Interaction = 0.064

The interaction between the Ghanami Akhdar cultivar and a growth regulator concentration of 3 mg L-1 yielded the highest saponification number for the essential oil, measuring 178.76. Conversely, the interaction involving the Wardi cultivar and a growth regulator concentration of 0 g L^{-1} resulted in the lowest value, recorded at 152.45, exhibiting a significant difference from the other interactions. The treatment of pollen of the Wardi cultivar with the growth regulator at a concentration of 0.3 g L^{-1} caused an increase in the saponification number of the essential oil by a percentage of 165.34.

Table (6): The effect of male cultivar and growth regulator fanphthyl acetamide on the Saponification number of essential oils

Male cultivars	fanphthyl acetamide			Mean of Male cultivars
	0 g L^{-1}	0.1 g L^{-1}	0.3 g L^{-1}	
GhanamiAkhdar	169.98	173.90	178.76	174.213
Ghanamiahmar	165.67	168.78	174.63	169.693
Khakri	161.35	164.69	171.87	165.970
Semismi	157.65	161.73	168.98	162.787
Wardi	152.45	157.89	165.34	158.560
Mean of fanphthyl acetamide	161.42	165.398	171.916	
L.S.D.	Male cultivars=4.234	fanphthylacetamide =2.234		Interaction = 6.340

Discussion

The study's results demonstrated that the five male cultivars differed in the specifications of the essential oil of their pollen grains, which may be due to genetic differences between the different male cultivars (Ali-Dinar, 2021). The study also showed the effect of treatment with the growth regulator fanphthyl acetamide on the qualitative characteristics of the essential oil in the pollen grains of the different male varieties, which may be attributed to the growth regulators that affect the elasticity of the cell walls and thus expand them and increase the activity of the cytoplasm of the cells and activate the metabolic processes in them (Hopkins & Muner, 2008) and increase the levels of auxins and gibberellins in them (Abbas *et al*., 2000). Growth regulators are organic molecules distinct from nutrients., It promotes, hinders, or alters physiological processes. They may be found naturally in plants or synthesized in labs. They significantly enhance irreversible growth when introduced in minimal quantities (Trivellini *et al*., 2015).The reason for the increase in the refractive index in the oil may be due to the rise in the unsaturation of the essential oil bonds or the increase in the percentage of fatty acids with a high molecular weight, which led to an increase in the refractive index of the essential oil in Ghanami Akhdar pollen grains, the increase in the density of the essential oil may be due to genetic variation, which is reflected in the effect on the percentage of solid compounds (oxygen compounds) in the essential oil, which is why its density is raised (Pino *et al*., 2006). The growth regulator also causes an increase in the efficiency of the metabolic processes of pollen grains and an increase in their products, including the quality of the essential oil, which is positively reflected in the saponification value number of the essential oil (Thabet *et al*., 2014).

Conclusions

Treatment with growth regulator had a significant effect on improving the qualitative characteristics of volatile oils present in pollen grains.

Acknowledgments

We express our gratitude to the University of Basrah, College of Agriculture, for providing facilities that facilitated this research.

Contributions of Authors

W.F.F.: Specimen collection, laboratory methodologies, authored and edited the article.

M.A.H.A.: Propose the paper, compose and edit the document, and identify the plant.

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Ethical approval

This study implemented all ethical criteria about plant care published by national and international organizations.

Conflicts of interest

The authors declare that they have no conflict of interests.

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تقدير محتوى الزيوت العطرية لحبوب لقاح خمسة أصناف ذكرية من نخيل التمر *Phoenix dactylifera* L. المعاملة بمنظم النمو fanphthyl acetamide

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المستخلص: اجريت هذه الدراسة في وحدة النباتات الطبية والعطرية التابعة لكلية الزراعة /جامعة البصرة على حبوب لقاح خمسة أصناف ذكرية من نخيل التمر لمعرفة محتواها من الزيوت العطرية. تم اختيار الأصناف الذكرية الغنامي الأخضر، الغنامي الاحمر، الخكري الوردي والسميسمي. اخذت طلعات ناضجة قبل تشقق أغلفتها وشملت الدراسة معاملة حبوب اللقاح بمنظم النمو fanphthyl acetamide بالتراكيز (0 ،1 ،3 غم لتر$^{-1}$). تم إجراء عملية الاستخلاص الزيوت العطرية من حبوب اللقاح باستخدام جهاز الكلافنجر. بينت نتائج الدراسة تفوق الصنف الذكري الغنامي الاخضر معنويا على بقية الأصناف في جميع المواصفات النوعية للزيوت العطرية في حين سجل الصنف الذكري وردي اقل معدل لجميع تلك الصفات. كما بينت نتائج الدراسة تاثير منظم النمو fanphthyl acetamide في الصفات النوعية للزيوت الطيارة لحبوب اللقاح اذ حسنت جميع الصفات النوعية للزيوت الطيارة من حيث النسبة المئوية للزيت العطري (0.041، 0.039، 0.035، 0.030، 0.027) % بالتتابع. والكثافة (0.940، 0.915، 0.885، 0.860، 0.810) ملغم مايكروليتر$^{-1}$ بالتتابع ومعامل الانكسار (1.515،1.535،1.564،1.587، 1.48) بالتتابع والوزن النوعي للزيت العطري (0.92، ،0.82،0.85،0.87،0.9) بالتتابع وPH الزيت العطري (5.94، 5.85،5.9، 5.81،5.81) بالتتابع ورقم التصبن للزيت العطري (171.87،174.63،178.76، 168.98، 165.34) بالتتابع ولجميع الأصناف الذكرية قيد الدراسة.

الكلمات المفتاحية: الزيوت العطرية، الزيوت الثابتة، الغنامي، منظمات النمو، حبوب اللقاح.