

Surveying, identification and characterization for the potential honeybee (*Apis mellifera* L.) pollen sources in the arid region of Riyadh-Saudi Arabia

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Abstract

This study was conducted targeting identification and documentation of major honeybee plants as pollen sources and their phenology in the central arid region of Saudi Arabia (Riyadh). Pollen loads were collected using pollen traps and were classified according to their color then traced back to plant species level. Throughout the year, sixteen plant species belonging to 10 plant families has been recorded and investigated by Light Microscopy (LM) and Scanning Electron Microscopy (SEM). Asian mustard (*Brassica tournefortii* Gouan.) (15.91%), willow wattle (*Acacia salicina* Lindl.) (15.17%), mesquite (*Prosopis juliflora*) (Sw.) DC. (21.81%) and cat's head (*Tribulus terrestris* L. var. *terrestris*) (15.52%) were the dominant sources of pollen. The period from May to July was found to be a significant blooming period and the scarcity period was from December to March in the study area. Color, size and shape of the potential pollen sources were variable between different species. Beekeepers could trap pollen loads that were collected from these sources during February, April and July, respectively. In conclusion, wise use and rehabilitation of these potentially identified bee plant species shall be considered in attempting beekeeping development.

Introduction

Pollen grains play a vital key role for honey bee nutrition. Honey bees gather pollen grains from plants to get the protein they need to survive (Al-Ghamdi, 2007a; Arien et al., 2020), colony strength (Belay et al., 2015), and productivity (Shawer et al., 2021). The type and quantity of the accessible flora, which is considered the primary source of pollen, have a significant impact on the potential for various colony products and success in beekeeping development. (Lau et al., 2019). Natural vegetation botanical makeup changes according to topography, climate, and soil type (Asefa et al., 2020). Consequently, the diversity of pollen depends on plant habitat, environmental conditions, distributions and flowering season (Amro, 2021; Begum et al., 2021).

Many researchers focused on palynological studies to help beekeepers in the abundance, distribution and flower blooming calendars of the main pollen sources in a various study area (Abou-Shaara, 2015; Adgaba et al., 2017; Taha et al., 2019). In order to satisfy their nutritional needs, bees should be given an array of

different floral resources. This will promote healthier populations (Brodtschneider & Crailsheim, 2010). For optimal honey production, beekeepers need to know when the major and minor nectar- and pollen-producing plants in the area of their apiaries are in bloom. This will allow them to decide when to introduce various management methods to their colonies (Agashe, 2021). Moreover, highlighting the periods in which beekeepers can collect bee-pollen from the major pollen sources (Taha, 2015) and determine the suitable period to introduce pollen substitutes and supplements (Amro et al., 2020) in order to economize the cost of feeding.

Little is known about the indigenous melliferous flora of Riyadh region which described as the harshest environmental conditions of Saudi Arabia (Abou-Shaara et al., 2013). The density of blooming plants has decreased as a result of these circumstances, resulting in a shortage of pollen available and nectar sources (Al-Ghamdi, 2007a). Thus, a high percentage of imported honey bees and a significant number of local honey bee colonies are dying annually (Al-Ghamdi, 2009).

This study aims to survey, identify and provide an account of pollen morphological features of the plants in the central region of Saudi Arabia using light and scanning electron microscopy. As well as, further research on the ecology and floral preferences of honey bees in developed landscapes will be built on the quantity and suitability of each plant species as a pollen supply for honey bee nutritional demands.

Material and Methods

The experiments were carried out in the laboratory and apiary (24° 34' 27" N 46° 41' 18" E). Five colonies, equal in strength of the native honey bee race (*Apis mellifera jemenitica*) and imported Carniolan hybrid (*Apis mellifera carnica*) headed by sister queens, were located in Derab farm (56° 39' 36.21" E) in Riyadh during 2016 from January to December.

Sample Collection

To collect pollen samples from available pollen forage plants in the study region, five colonies from native honey bee race (*A. m. jemenitica*) and another from imported honey bee (*A. m. carnica*) were used. A pollen trap was fixed on the hive entrance of each colony. Every fifteen days, pollen traps were run for 3 days, giving the colonies 12 days of free entering pollen. Each sampling's pollen pellets were weighed and kept at -10°C until pollen classification and separation.

Description of Used Pollen Trap

The used pollen trap composed of wooden box with a slope roof and two vertical metal strips each of 32 cm length x 17 cm width. Each strip has 16 holes sq./inch (5 mm in diameter/hole). Pollen loads fallen through a horizontal wire gauze screen into a collecting tray which emptied as required (Abd El Salam et al., 2022). For increasing trap efficiency of native colonies, the board of trap entrance was changed with another one (3.75 mm in diameter/hole) to become suitable for the small size of native bee workers (*A. m. jemenitica*). 100 returning pollen foragers were observed as they entered the hive with pollen loads through an empty trap to gauge the effectiveness of the trap. The number of pellets that fell onto the tray was counted, and the efficiency was computed using the formula provided by Abd El Salam et al. (2022), as the following:

$$\text{Trap efficiency} = \frac{\text{Number of pollen pellets in the trap box}}{200} \times 100$$

Separation of Pollen Pellets

The fresh pollen pellets for every trap was cleaned and weighed. Pollen types were separated by color of pellets using small drawing brush. Every group contains the same color more than 70% of the total was considered as the major pollen source, while those with 50-70% and from 30-50% of the total were considered as moderate and minor pollen sources, respectively (Kirk, 1994).



Figure 1: Pollen pellets colors varied from brown in willow wattles (*Acacia salicina* Lindl.), lemon in asian mustard (*Brassica tournefortii* Gouan.), olive in mesquite (*Prosopis juliflora* (Sw.) DC.) and in cat's head (*Tribulus terrestris* L. var. *terrestris*) pollen pellets were appeared in dark beige coloration.

Identification of Pollen Sources

Once pellets were split into groups based on colour, using the structure of the pollen grains as a guide, groups were further separated into various pollen types. (Dimou & Thrasyvoulou, 2007). During the same flowering period, in order to prepare pollen grains reference slides, pollen grains were directly collected from the adjacent opening flowers of the congruent blooming plants around the apiary. The morphology of each species of pollen grains was then compared with that of pollen pellets collected from pollen traps.

Identification of collected plants was carried out at King Saud University Herbarium in College of Sciences, where they were deposited. Family names were according to angiosperm phylogeny website. Method of Collenette (1999), was used for nomenclature of genera and species, the latest taxonomic changes as well as author update.

Preparing Standard Pollen Grain Slide

The specimens' fully grown anthers were removed, and they were then processed using the standard acetolysis procedure (Erdtman, 1960). Only newly

opened flowers were used to prevent contamination from other pollen sources. Only newly opened flowers were used, so that contamination from other pollens is avoided. A few drops of isopropyl alcohol (IPA) were added and leaved for 10 minutes to detach the pollen grains from the anthers and remove the waxy covering from them. A drop of molten glycerin jelly was placed on a warm coverslip and slowly upturned slide lowered so that the pollen grains contact the jelly. The slide was left on the hotplate for 5 mins to allow the stain to penetrate the pollen grains. The slide was labeled with the name of the plant and the date of pollen collection.

Measurement of Pollen Grain Dimensions

The diameter of pollen grains was measured using an ocular micrometer (n=10) for each pollen type.

According to Erdtman (1954), the pollen grain's polar (P) and equatorial (E) axes' lengths and the P/E ratio (index shape) were measured.

Light Microscope Examinations

Under a light microscope (Olympus –CH20i BIMU) provided with Camera Axicocam 512 color, under E40, 0.65 and Oil immersion (E100, 1.25), using a 10x eye piece, measurements and morphological observations were conducted. This procedure was conducted to get a clearer vision for the structure of the tested pollen grains obtained from the main pollen sources in the study area.

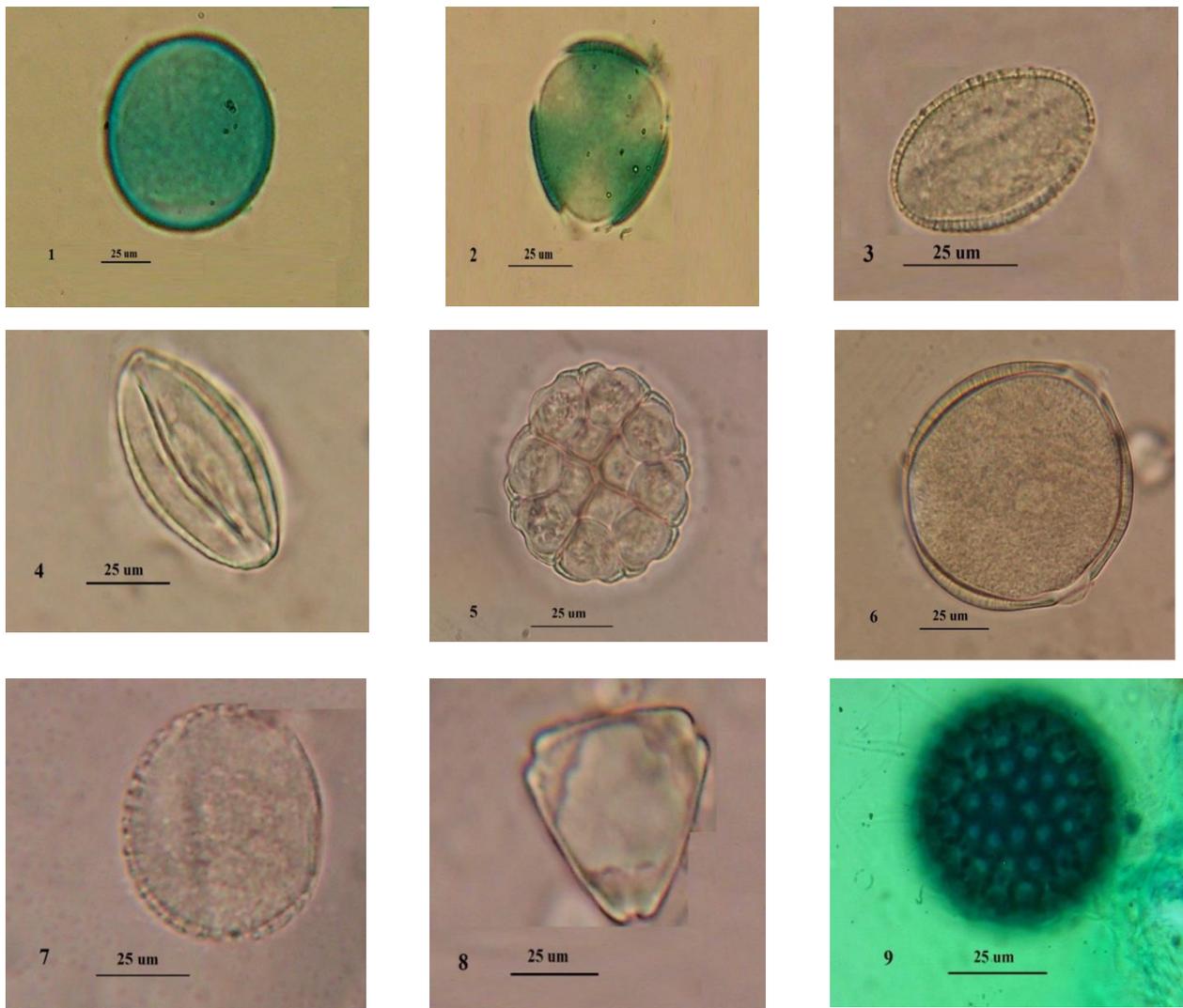


Figure 2: Light micrographs (LM) showing shaped and dimensions of pollen grain collected from Riyadh region.

1- *Phoenix dactylifera* L. 2- *Eruca sativa* (Miller) Thell. 3- *Brassica tournefortii* Gouan. 4- *Prosopis juliflora* (Sw.) DC. 5- *Acacia salicina* Lindl. 6- *Leuceana leucocephala* (Lam.) De Wit. 7- *Parkinsonia aculeate* L. 8- *Eucalyptus camaldulensis* Dehnh. 9- *Tribulus terrestris* L. var. *terrestris magnification* (1000 x).

Scanning Electron Microscopic Examinations

The scanning electron microscope was used for observing pollen wall surface. After dehydration on a silica gel drier, small dry quantities of pollen grain were mounted on scanning electron microscopy (SEM) stubs using double sided adhesive. The samples were then coated with gold in a JOEL JFC 1100E ion-sputtering device and examined in JOEL JSM 5400 LV scanning electron microscope, operated at accelerated voltage of 15 KV at the Scanning Electron Microscope Unit. The terminology of Boesewinkel and Bouman (1984), was used to describe the achene coat characteristics.

Statistical Analysis

The experiment was designed using a fully randomized design. For the aforementioned parameters, Analysis of variance (ANOVA) was used to statistically examine the given data. The Duncan's multiple range test (Duncan, 1955) was used to compare the means at a significance level of 5% with the SAS 9.1.3 programme (SAS Institute, 2004).

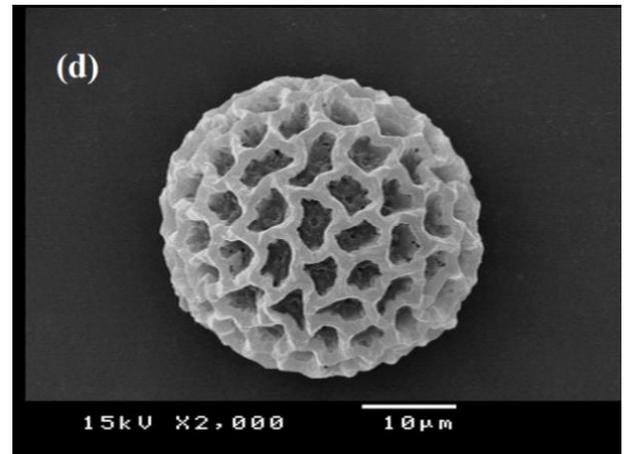
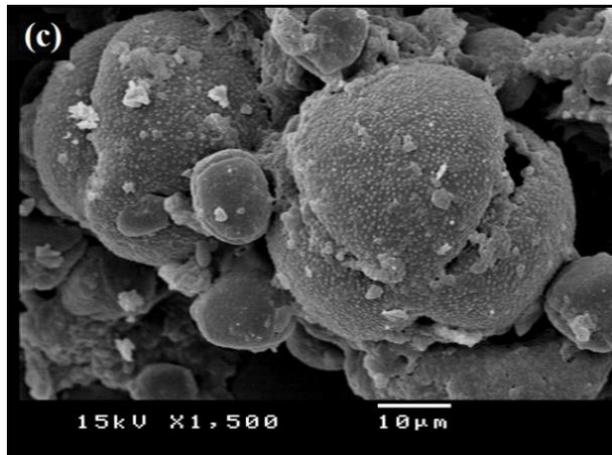
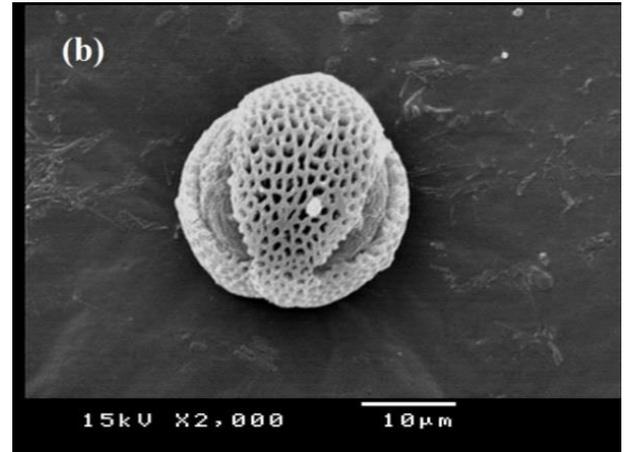
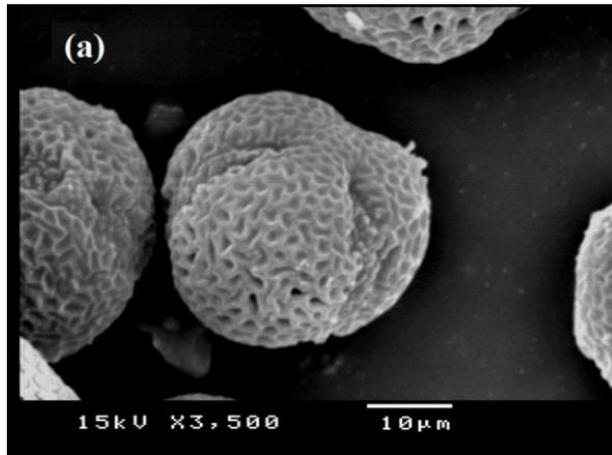


Figure 3. Scanning electron microscope (SEM) micrographs showing surface structure of selected pollen grains from Riyadh region.

a- *Prosopis juliflora* (Sw.) DC. b- *Brassica tournefortii* Gouan. c- *Acacia salicina* Lindl. d- *Tribulus terrestris* L. var. *terrestris* (Scale bars represent 10 µm for photomicrograph)

Results

Botanical Sources of Pollen and their Flowering Periods

The flowering periods of pollen sources in study area are shown in Table 1. Sixteen pollen sources belonging to ten plant families were identified. The most frequent pollen plant sources were from families Brassicaceae and Fabaceae. Honey bee plant species *Brassica tournefortii* Gouan, *Acacia salicina* Lindl., *Prosopis juliflora* (Sw.) DC., and *Tribulus terrestris* var. *terrestris* were ranked as the first four pollen plants and formed more than 70% in most of collected samples. These plants were considered the major pollen sources. On the other hand, *Phoenix dactylifera* L., *Eruca sativa* (Miller) Thell., *Leuceana leucocephala* (Lam.) De Wit., *Parkinsonia aculeata* L. and *Eucalyptus camaldulensis* Dehn which formed 50-70% of the samples considered

as moderate pollen sources. However, *Taraxacum officinale* F.H. Wigg, *Borago officinalis* L., *Citrullus colocynthis* (L.) Schrad., *Helianthus annuus* L., *Portulaca oleraceas* L. and *Nigella arvensis* var. *arabica* L., which formed 30-50% considered as minor pollen sources.

The asian mustard (*B. tournefortii*) was recorded during the periods from the first January till late March. However, flowering of willow wattle (*A. salicina*) was recorded three times, during mid-February till mid-April, mid-July till mid-August and from mid-October till mid-December. While, mesquite pollen was collected in the study area from mid-April till mid-August. The rest of pollen sources were recorded through 2016 season in differential periods. In general, results showed that the period from mid-April till late November recorded the highest presence of the pollen sources.

Table 1: Flowering periods of pollen botanical sources in Riyadh during 2016.

Common name	Family	Scientific name	Abundance	Months													
				Jan.	Feb.	Mar.	Apr.	May	Jun.	Jul.	Aug.	Sep.	Oct.	Nov.	Dec.		
Date palm	Arecaceae	<i>Phoenix dactylifera</i> L.	xx		■												
Dandelion	Asteraceae	<i>Taraxacum officinale</i> F.H. Wigg	x				■	■	■	■							
Sunflower	Asteraceae	<i>Helianthus annuus</i> L.	x			■					■						
Borage	Boraginaceae	<i>Borago officinalis</i> L.	x											■	■	■	
Salad rocket	Brassicaceae	<i>Eruca sativa</i> (Miller) Thell.	xx	■	■									■	■	■	
Asian mustard	Brassicaceae	<i>Brassica tournefortii</i> Gouan.	xxx	■	■	■											
Colocynth	Cucurbitaceae	<i>Citrullus colocynthis</i> (L.) Schrad.	x				■	■	■	■	■	■	■	■	■	■	
Willow wattle	Fabaceae	<i>Acacia salicina</i> Lindl.	xxx		■	■	■				■	■		■	■	■	
White lead tree	Fabaceae	<i>Leuceana leucocephala</i> (Lam.) De Wit	xx				■	■	■	■	■	■	■	■	■	■	
Parkinsonia	Fabaceae	<i>Parkinsonia leculeate</i> L.	xx							■	■	■	■	■	■	■	
Blue palo verde	Fabaceae	<i>Cercidium floridum</i> AZ.	x				■	■	■	■	■	■	■	■	■	■	
Mesquite	Fabaceae	<i>Prosopis juliflora</i> (Sw.) DC.	xxx				■	■	■	■	■	■	■	■	■	■	
Camphor	Myrtaceae	<i>Eucalyptus camaldulensis</i> Dehn.	xx					■	■	■	■	■	■	■	■	■	
Verdolaga	Portulacaceae	<i>Portulaca oleraceas</i> L.	x			■									■		
Nigella	Ranunculaceae	<i>Nigella arvensis</i> var. <i>arabica</i> L.	x					■					■				
Cat's head	Zygophyllaceae	<i>Tribulus terrestris</i> L. var. <i>terrestris</i>	xxx				■	■	■	■	■	■	■	■	■	■	

x = Minor Pollen source (30% – 50%), xx= Moderate Pollen source (50% - 70%) and xxx= Major Pollen source (more than 70%)

Abundance of the pollen sources

Availability and quantities of trapped pollen from the main pollen sources collected by Yemeni race and Carniolan hybrid honey bees was clarified in Table 2. The dominant plant families in the area were Fabaceae (22.64%) and Brassicaceae (15.53%), followed by Zygophyllaceae (14.58%). Pollen types varied in time, frequency, and species richness. The obtained data showed that the used plant sources were continuously presented in the area of study. In general, honey bee foragers collect most pollen amount from mesquite, asian mustard, cat's head, and willow wattle, respectively. Quantities of pollen loads collected by Yemeni race from all plant sources were higher than those collected by Carniolan hybrid. Highly significant variations ($P < 0.05$) were recorded between the quantity of pollen loads collected by Yemeni and Carniolan hybrid bees. The higher quantities of pollen

loads collected by Yemeni race can be arranged descending as follows: 2833.57 > 2067.50 > 2017.19 > 1970.84 gm/colony for mesquite, asian mustard, cat's head, and willow wattle, respectively. Asian mustard and mesquite plants were presented during 3 months each. Except December, the rest of plant sources appeared during two months each. Mesquite plants lasted in the study area for the longest period and their pollen were constantly present in the pollen traps and recorded in 11 samples during the entire study period. It followed by asian mustard, willow wattle and cat's head. However, salad rocket exhibited the lowest period of presence, where it occurred in 3 samples only. Although, the Yemeni race collected pollen loads more than Carniolan hybrid bees, the percentage from total collected pollen showed semi equal values between the two races.

Table 2. Presence and quantity of trapped pollen load (gm/colony/flowering period) in Yemeni race and Carniolan hybrid honeybee colonies from the main pollen botanical sources in Riyadh during 2016.

Common name	Family name	Scientific name	Presence in traps (Catch No.)	Quantity of pollen loads (gm/colony/flowering period)			
				Total (\pm S.D.**)		% from total collected pollen	
				Yemeni race	Carniolan hybrid	Yemeni race	Carniolan hybrid
Asian mustard	Brassicaceae	<i>Brassica tournefortii</i> Gouan.	Jan. 17 to Mar 6 (9)	2067.50 \pm 31.60 b*	1110.09 \pm 15.23 e	15.91 \pm 0.09 c	15.15 \pm 0.25 d
Willow wattle	Fabaceae	<i>Acacia salicina</i> Lindl.	Mar. 12 to Apr. 26 (9)	1970.84 \pm 56.72 c	933.12 \pm 30.04 g	15.17 \pm 0.24 d	12.74 \pm 0.49 f
Cat's head	Zygophyllaceae	<i>Tribulus terrestris</i> L. var. <i>terrestris</i>	May 2 to Jun. 19 (9)	2017.19 \pm 69.92 c	999.53 \pm 30.71 f	15.52 \pm 0.29 cd	13.64 \pm 0.29 e
Camphor	Myrtaceae	<i>Eucalyptus camaldulensis</i> Dehnh.	Jun. 25 to Jul. 13 (4)	1038.50 \pm 38.43 f	630.97 \pm 55.99 j	8.00 \pm 0.35 i	8.60 \pm 0.68 h
Mesquite	Fabaceae	<i>Prosopis juliflora</i> (Sw.) DC.	Jul. 19 to Sep. 19 (11)	2833.57 \pm 71.43 a	1719.61 \pm 31.89 d	21.81 \pm 0.32 b	23.47 \pm 0.45 a
White lead tree	Fabaceae	<i>Leuceana leucocephala</i> (Lam.) De Wit	Sep. 23 to Oct. 11 (4)	838.88 \pm 40.50 h	495.40 \pm 17.22 k	6.45 \pm 0.30 j	6.76 \pm 0.24 j
Parkinsonia	Fabaceae	<i>Parkinsonia aculeata</i> L.	Oct. 17 to Nov. 29 (8)	1128.31 \pm 26.94 e	742.79 \pm 13.51 i	8.68 \pm 0.17 h	10.13 \pm 0.12 c
Salad rocket	Brassicaceae	<i>Eruca sativa</i> (Miller) Thell.	Dec. 5 to Dec. 18 (3)	470.82 \pm 38.43 k	315.59 \pm 16.26 m	3.62 \pm 0.35 m	4.38 \pm 0.23 l
Date palm	Arecaceae	<i>Phoenix dactylifera</i> L.	Jan. 24 to Mar. 11 (4)	622.99 \pm 23.26 j	378.82 \pm 3.49 l	4.79 \pm 0.20 k	5.17 \pm 0.03 k

* Means followed by the same letter are not significantly different at 0.05 level of probability, by Duncan's multiple range test.

**S.D.= Standard deviation

Pollen Characterization

Pollen grain dimensions and coloration

Data presented in Table 3 show the shapes, colors and dimensions of the pollen grains collected from study region which represented the major and moderate pollen botanical sources. Pollen grains which contained the highest length and width belonged to 9 plant species and 5 families. It is clear that family Fabaceae was represented by 4 plant genera, followed by family Brassicaceae by 2 genera and families Arecaceae, Myrtaceae and Zygophyllaceae by one genus for each.

Pollen grains of the white lead tree (*L. leucocephala*) showed the highest length of polar and equatorial of the collected grains with an average of 50.4 μ m polar and 47.9 μ m equatorial lengths followed by cat's head (*T. terrestris*) grains by 47.4 polar and 47.6 μ m equatorial lengths. Pollen grains of willow wattle (*A. salicina*) ranked the third one by 42.7 polar and 36.4 μ m equatorial lengths. The remaining pollen grains exhibited the least values of polar and equatorial lengths, and camphor (*E. camaldulensis*) ranked the last with an average of 17.4 polar and 17.0 μ equatorial lengths. It is important to note that pollen grains length

Table 3. Pollen grain dimensions and pellets color of main pollen botanical sources collected by honeybee colonies from Riyadh during 2016.

Common name	Family	Scientific name	Length axis (μm)		Shape index P/E	Pollen pellets color
			Polar (P) \pm S.D.*	Equatorial (E) \pm S.D.		
Date palm	Arecaceae	<i>Phoenix dactylifera</i> L.	19.3 \pm 0.08	19.3 \pm 0.08	1.0	Creamy
Salad rocket	Brassicaceae	<i>Eruca sativa</i> (Miller) Thell.	19.9 \pm 0.11	20.1 \pm 0.08	1.0	Dark yellow
Asian mustard	Brassicaceae	<i>Brassica tournefortii</i> Gouan.	32.6 \pm 0.13	26.9 \pm 0.13	1.2	Lemon
Willow wattle	Fabaceae	<i>Acacia salicina</i> Lindl.	42.7 \pm 0.28	36.4 \pm 0.50	1.2	Brown
White lead tree	Fabaceae	<i>Leuceana leucocephala</i> (Lam.) De Wit	50.4 \pm 0.22	47.9 \pm 0.36	1.1	Light yellow
Parkinsonia	Fabaceae	<i>Parkinsonia leculeate</i> L.	22.4 \pm 0.31	21.9 \pm 0.21	1.0	Dark orange
Mesquite	Fabaceae	<i>Prosopis juliflora</i> (Sw.) DC.	28.1 \pm 0.23	27.3 \pm 0.11	1.0	Olive
Camphor	Myrtaceae	<i>Eucalyptus camaldulensis</i> Dehnh.	17.4 \pm 0.09	17.0 \pm 0.08	1.0	Bright brown
Cat's head	Zygophyllaceae	<i>Tribulus terrestris</i> L. var. <i>terrestris</i>	47.4 \pm 0.36	47.6 \pm 0.27	1.0	Dark beige

*S.D.= Standard deviation

of *L. leucocephala* is equal 2.89 and 2.82 fold of polar and equatorial lengths of *E. camaldulensis* pollen grain.

Morphological Characterization of Pollen Grains

By using the light microscope (LM), the shape and dimensions of pollen grains collected from botanical origin in the study area showed wide differences (Fig. 1). The pollen grains of *P. dactylifera*, *E. sativa*, *P. aculeate*, *P. juliflora*, *E. camaldulensis* and *T. terrestris*, were spheroid (shape index 1.0), whereas, *B. tournefortii* and *A. salicina* were sub-prolate (shape index 1.2). Only *L. leucocephala* pollen grains were prolate-spheroid (shape index 1.1).

Description of the main pollen sources using scanning electron microscope (SEM) is illustrated in Fig. 2. The results showed that pollen grains of *P. juliflora* (Fig. 2a) are largely homogeneous. The shape index is prolate-spheroidal. The mature pollen grains are tricellular and tricolporate with a regulate exine. On the other side, *B. tournefortii* pollen grains are tricoplate (Fig. 2b) whereas, they have a sub-prolate shape, and the exine is a finely perforated reticulate sculpture with lumina that are smaller than the muri or equal in size to them. However, *A. salicina* pollen grains are spherical ovoidal with openings at the proximal surface and colpus at the distal surface, scattered as an interconnected-polyad (Fig. 2c). In regards to *T. terrestris*, pollen grains were found to be spheroidal (Fig. 2d). The exine decoration is reticulate with straight to slightly expressed wavy boundaries (muri) and a straightforward columnar structure.

Discussion

Pollen Botanical Sources and Flowering Periods

The current results showed that rape, date palm and the Acacia trees are the main pollen sources in the harshest environmental region of Saudi Arabia (Riyadh). In the same area Alqarni (2020), recorded notable foraging behaviour for the *A. m. jemenitica* (indigenous) and *A. m. carnica* (exotic) on the Acacia trees (*Acacia gerrardii* Benth.). In eastern province of Saudi Arabia in Al-hasa region (semi-arid), Al-Jabr and Nour (2001), stated that rape, date palm and the Acacia trees considered main pollen sources for honey bees. In addition, Taha (2015), recorded family Brassicaceae among the most abundant pollen sources in the same region.

In the present study, acacia trees were notable along the study period, in addition the long presence period of *Acacia salicina* pollen flow in the field confirmed by Khan et al. (2019). He came to the conclusion that because Acacias have the majority of the characteristics needed to resist harsh climatic circumstances, they are the most effective "survivors" in desert environments. Also, Bilisik et al. (2008), stated that the honey bee foragers focused on a few plant species at a particular time despite the richness and diverse flora in the study area. According to their findings, the pollen varieties exhibiting high quantities were prevalent around the hives. They also, reported that the Brassicaceae (*Papaver* spp.), is the main pollen taxa collected by honey bees in the area. Among the

plant families which studied by Da luz et al. (2010), and Mayda et al. (2020), Asteraceae, Fabaceae, Arecaceae, and Myrtaceae were recorded as the major richness plant families of pollen sources types. In addition, the latter researcher confirmed that the elemental analysis showed that all samples were rich in essential minerals.

In reference to a particular plant, prosopis was recognized as a significant source of nectar for honey bees (Zaitoun et al., 2009). Also, Suryanarayana et al. (1992), found that numerous insects, notably honey bees, willfully visit Asteraceae inflorescences of the capitulum type. This information insures that the plant sources identifying during this study have special importance as pollen sources for honey bee in Riyadh region. In Saudi Arabia, Al-Ghamdi (2007b), confirmed our results by recorded *E. sativa*, *Eucalyptus rostrata*, *L. leucocephala*, *Brassica juncea*, *Acacia farnesiana* and *A. salicina* as the most promising bee plants during their flowering periods in relation to various honey bee activities. Also, he noted that most of the species were controllable and that weedy vegetation might be seen as a significant source of food for bees because it commonly appeared in the pollen spectrum even in well farmed areas.

Concerning the data obtained from Table1, it was observed that *H. annuus*, *E. sativa*, *A. salicina*, *P. oleraceas* and *N. arvensis* bloomed 2-3 times a year. The present results supported by the observation of Al-Ghamdi (2007b) and Amro et al. (2020), in which they stated that availability of secondary sources of honey bee forage plants is seen as a very critical issue and helps bees to live in times of scarcity in the Riyadh region. Date palm (*P. dactylifera* L.) (Arecaceae) was identified as another significant resource utilized by both Yemeni and Carniolan races in the study area, and in several samples, it was the dominant source of pollen. The importance of Arecaceae pollen as a resource for *A. mellifera* was described by Lau et al. (2019). In addition, Asteraceae ornamental species are thought to be particularly significant insect attractor plants (Denisow et al., 2014). Also, Importance of mesquite trees (*P. juliflora*) as a pollen source for honey bee colonies was confirmed (Zaitoun et al. 2009). They concluded that the mesquite is an important plant species which represents practically the only source of nectar and pollen to bees in April and May in several of the arid and semi-arid zones of Durango, Mexico. It is a crucial plant because it has a great capacity to fix nitrogen in arid environments and during droughts, and it offers shelter and food to a variety of species who feed its nectar, pollen, leaves, and fruits.

Abundance of the pollen sources

Our results showed highest collected pollen loads initiated by Yemeni race and Carniolan hybrid honey bee have been recorded during summer months when honey bees foraged on *L. Leucocephala*, *P. Juliflora* and *T. terrestris*, followed by spring season during the flowering periods of *P. dactylifera*, *B. tournefortii* and *A.*

salicina. In the same line, Alqarni (2020) recorded that *A. m. jeminica* recruited significantly ($P < 0.01$) more active pollen-gathering bees than the *A. m. carnica* especially on Acacia trees. Plants families Aracaceae, Brassicaceae, Fabaceae, Myrtaceae and Zygothyllaceae were the most flowering plants visited by honey bee colonies as a pollen sources in the present investigation. The importance of these families as a pollen sources for honey bees were confirmed (Alves & Dos Santos, 2016; Alqarni, 2020). They recorded that Aracaceae and Fabaceae were the most important families supplying honey bee colonies with high amount of pollen. In a comparative study from Turkey and Romania bee pollen, Mărgăoan et al. (2021), suggested that the fatty acids contents were closely correlated with the above-mentioned parameters especially with the botanical origin and antibacterial activity. Our findings suggest that BP is a rich source of unsaturated fatty acids and bioactive compounds, which can be considered a value-added product and concluded that geographical location is a determining factor for plant pollen viability.

Data presented in Table 2, highlighted that native bees were significantly ($P < 0.05$) more active foragers than the exotic one. In the same line, Al-Ghamdi et al. (2020), stated that the examined nursery stage under Acacia species flow performed well and within acceptable ranges, potentially qualifying them as suitable biological tools for introducing genotypes into apicultural landscape restoration projects. Also, Taha (2015), point out to families Arecaceae, Brassicaceae and Fabaceae as the most plentiful sources of pollen for honey bee colonies in Alhasa region, Saudi Arabia. Highly significant differences between quantities of pollen collected from different families, as well as, different plant species and honey races were recorded. Our field experiments indicated statistically significant differences among tested honey bee strains for all quantitative pollen yield parameters. The percentages of collected pollen loads were statistically significant higher ($P < 0.05$) in indigenous bees (*A. m. jeminica*) than the imported one especially for the plant species *A. salicina*, and *T. terrestris*. On the other hand, the statistical analyses showed the vigor of imported bees (*A. m. carnica*) to collect more pollen from *P. dactylifera*, *E. sativa*, *P. aculeate* and *P. juliflora*. Hence, the amount of pollen produced by different species can vary greatly, and the reward of a given species might vary depending on the area and growth season. In the same line Amro et al. (2020), conclude that it is indispensable to use both races (Carniolan and Yemeni bees) in extremely dry desert areas, because each of them has specifications and capabilities that allow them to forage and collect pollen in a way that distinguishes each one from the other.

The present results showed that mesquite (*P. juliflora*) pollen pellets achieve superior vs the other tested plants followed by asian mustard (*B. tournefortii*), cat's head (*T. terrestris*) and willow wattle (*A. salicina*) plants. These results were matched with Taha (2015),

who considered that *Prosopis* spp., *Brassica* spp., *Tribulus* spp. were the most important pollen sources for honey bee colonies. He observed that bee foragers focused only on four flowering plants to collect major quantities of pollen in Alhasa region. This phenomenon was previously illustrated by Klein et al. (2019). They verified that *A. mellifera* can concentrate foraging efforts on a single type of flowers. Therefore, the fidelity in the studied areas could be attributed to the scarcity of other flowering species, due to seasonal inundation of the floodplain. This fact could force honey bees to visit flowers of a limited number of plants. Also, floral fidelity (Goodwin, 2012) could be related to the distance of the hives to floral sources. In addition, color, shape and flowers odour, energy requirement and caloric rewards offered by flowers also determine whether honey bee can be a dependable flower visitor (Dobson & Bergström, 2000).

Continuous presence of *T. terrestris* in the present study reinforced by Yankova- Tsvetkova et al. (2011). They claimed that the high pollen and embryo viability established as well as sexual reproduction are important factors in determining if *T. terrestris* has a suitable amount of reproductive potential and whether its successful realization ensures the stability of its populations. Naghiloo and Siahkolaee (2019), also, referred to *T. terrestris* as being extensively dispersed due to its facile seed transmission in desert, warm, and moderately temperate parts of the globe. This species' ability to successfully use its reproductive potential is also a result of its trait as an entomophilous plant that generates a significant amount of pollen grains.

Pollen Characterization

The obtained results showed considerable variation in pollen morphology has been detected between the plant sources. Except of cat's head and asian mustard the largest polar length and equatorial length of the collected pollen grains in this work belonged to family Fabaceae. However, cat's head pollen shape was in agreement with Rouhakhsh et al. (2014), who documented that *T. terrestris* pollen grains are oblate-spheroidal, radially symmetric, pantoporate. Also, Semerdjieva et al. (2011), reported that exine ornamentation for *T. terrestris* is reticulate and the sizes of the pollen grains ranged between 35 and 47.5 μm with an average 43.5 μm .

In this investigation, pollen morphology of *E. sativa* and *B. tournefortii* showed similar phenomena with 39 species belonging to family Brassicaceae, studied in Saudi Arabia by Umber et al. (2021). The dominant feature is tricolpate for the pollen grains which ranged in form from prolate spheroidal to subprolate to prolate. The same author recorded that *Schimpera arabica* (Brassicaceae), which has the smallest grains in this family, has a polar axis of 16 μm and an equatorial diameter of 15 μm . However, *Malcolmia pygmaea*, has the largest polar axis of 37 μm and an equatorial diameter of 21.5 μm . Additionally, the finding of Gosling

et al. (2013) supported ours by classifying family Brassicaceae as a stenopalynous, whereas the pollen grains are usually reticulate and tricolpate.

Conclusion

The large amounts of pollen obtained from asian mustard and mesquite reflect its significance as important pollen forage plant for honey bees in this area. Plants families Aracaceae, Brassicaceae, Fabaceae, Myrtaceae and Zygophyllaceae were the most flowering plants visited by honey bee colonies as pollen sources in Riyadh region. The beekeepers interested in collecting pollen can be advised to hold their pollen traps from mid-January to the first of march and mid-July to mid-September as the most suitable periods for collecting pollen in the Riyadh region. Further studies on honey bee colonies operating in the harsh environmental conditions of central Arabia are needed.

Ethical Statement

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Conflict of Interest

The authors declare that they have no competing interests.

Author Contributions

All authors of this manuscript contributed equally to the design and/or execution of the experiments described in the manuscript. A.A prepared and edited the final version of this manuscript. All authors approved the final manuscript.

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References

- Abd El Salam, E. H., Ali, M. A., & Ghazala, N. A. (2022). Evaluation of the Efficiency Different Types of Bee Pollen Collection Traps in Honey Bee Colonies during Summer Season. *Arab Universities Journal of Agricultural Sciences*, 30(1), 141-146. <http://10.21608/AJS.2022.111909.1448>.
- Abou-Shaara, H. F. (2015). Potential honey bee plants of Egypt. *Cercetařri Agronomice în Moldova*, 48, 99-108. <https://repository.uaiaisi.ro/xmlui/handle/20.500.12811/1530>.
- Abou-Shaara, H. F., Ahmad A. Al-Ghamdi, & Abdelsalam, A. M. (2013). A suitability map for keeping honey bees under harsh environmental conditions using Geographical Information System. *World Applied Sciences Journal*, 22,

- 81099-1105. <https://doi.org/10.5829/idosi.wasj.2013.22.08.7384>.
- Adgaba, N., Alghamdi, A., Sammoud, R., Shenkute, A., Tadesse, Y., Ansari, M. J., Sharma, D., & Hepburn, C. (2017). Determining spatio-temporal distribution of bee forage species of Al-Baha region based on ground inventorying supported with GIS applications and Remote Sensed Satellite Image analysis. *Saudi journal of biological sciences*, 24, 1048–1055. <https://doi.org/10.1016/j.sjbs.2017.01.009>
- Agashe, S. N. (2021). Aerobiology for the Clinician: Basic and Applied Aspects, Pollen Sources, Pollen Calendars. In *Textbook of Allergy for the Clinician*, CRC Press, 49-68.
- Al-Ghamdi, A. A. (2007a). Evaluation of various honey bee foraging activities for identification of potential bee plants in Riyadh, Saudi Arabia. *Annals of Agricultural Science-Cairo*, 52(2), 487.
- Al-Ghamdi, A. A. (2007b). *Beekeeping and honey production in Saudi Arabia*. Fifth Conference of Arab Beekeepers Association. Tripoli, Libya.
- Al-Ghamdi, A. (2009). Comprehensive study for the current beekeeping industry of imported and native honey bee in Kingdom of Saudi Arabia. *King Abdulaziz City for Science and Technology*, Final report of project, AT-21-39.
- Al-Ghamdi, A. A., Tadesse, Y., & Adgaba, N. (2020). Evaluation of major Acacia species in the nursery towards apicultural landscape restoration around Southwestern Saudi Arabia. *Saudi Journal of Biological Sciences*, 27(12), 3385-3389. <https://doi.org/10.1016/j.sjbs.2020.09.002>.
- Al-Jabr, A. M., & Nour, M. E. (2001). Content of some Saudi honeys of pollen spectrum and sucrose. *Journal of Agriculture Science, Mansoura University*, 26: 4009-4014.
- Alqarni, A. S. (2020). Differential foraging of indigenous and exotic honey bee (*Apis mellifera* L.) races on nectar-rich flow in a subtropical ecosystem. *Insects*, 11(4), 254. <https://doi.org/10.3390/insects11040254>.
- Alves, R. De França, & Dos Santos De Assis R. F. (2016). Arecaceae potential for production of monofloral bee pollen. *Grana*, 56(4), 294-303. <https://doi.org/10.1080/00173134.2016.1239760>.
- Amro, A. M. (2021). Pollinators and pollination effects on three canola (*Brassica napus* L.) cultivars: A case study in Upper Egypt. *Journal of King Saud University-Science*, 33(1), 101240. <https://doi.org/10.1016/j.jksus.2020.101240>
- Amro, A. M., Omar, M. O., & Ahmed, A. G. (2020). Impact of using pollen substitutes on performance of honey bee (*Apis mellifera* L.) colonies under harsh environmental conditions. *Science and Animal Health*, 8(3), 236-256. <https://doi.org/10.15210/sah.v8i3.19992>.
- Arien, Y., Dag, A., Yona, S., Tietel, Z., Cohen, T. L., & Shafir, S. (2020). Effect of diet lipids and omega-6: 3 ratio on honey bee brood development, adult survival and body composition. *Journal of insect physiology*, 124, 104074. <https://doi.org/10.1016/j.jinsphys.2020.104074>.
- Asefa, M., Cao, M., He, Y., Mekonnen, E., Song, X., & Yang, J. (2020). Ethiopian vegetation types, climate and topography. *Plant Diversity*, 42(4), 302-311. <https://doi.org/10.1016/j.pld.2020.04.004>.
- Begum, H. A., Iqbal, J., & Aziz, A. (2021). Characterization of pollen profile of *Apis mellifera* L. in arid region of Pakistan. *Saudi Journal of Biological Sciences*, 28(5), 2964-2974. <https://doi.org/10.1016/j.sjbs.2021.02.035>.
- Belay, A., Solomon, W. K., Bultossa, G., Adgaba, N., & Melaku, S. (2015). Botanical origin, colour, granulation, and sensory properties of the Harena forest honey, Bale, Ethiopia. *Food chemistry*, 167, 213-219. <http://dx.doi.org/10.1016/j.foodchem.2014.06.080>
- Bilisik, A., Cakmak, I., Saatcioglu, G., Bicakci, A., & Malyer, H. (2008). Spectrum of pollen collected by honey bees in bursa lowland area in high season. *Uludag Bee Journal*, 8(4), 143-148. <https://dergipark.org.tr/en/pub/uluaricilik/issue/53267/162401>
- Boesewinkel, F. D., & Bouman, F. (1984). The Seed: Structure. In *Embryology of Angiosperms*, 1ed. Johri, Springer Berlin Heidelberg, B.,567-610.
- Brodtschneider, R., & Crailsheim, K. (2010). Nutrition and health in honey bees. *Apidologie*, 41(3), 278-294.
- Collenette, S. (1999). *Wild Flowers of Saudi Arabia*. National Commission for Wild Life Conservation and Development (VCWCD), Riyadh, Saudi Arabia, 799.
- Da Luz, C. F. P., Junior, G. L. B., Fonseca, R. L. S. E., & DE Sousa, P. R. (2010). Comparative pollen preferences by Africanized honey bees *Apis mellifera* L. of two colonies in Pará de Minas, Minas Gerais, Brazil. *Anais da Academia Brasileira de Ciências*, 82(2), 293-304.
- Denisow, B., Strzałkowska-Abamek, M., Bożek, M., & Jeżak, A. (2014). Ornamental Representatives of the Genus *Centaurea* L. as a Pollen Source for Bee Friendly Gardens. *Journal of Apicultural Science*, 58(2), 49-58. <http://doi:10.2478/jas-2014-0016>
- Dimou, M., & Thrasvoulou, A. (2007). A comparison of three methods for assessing the relative abundance of pollen resources collected by honey bee colonies. *Journal of Apicultural Research*, 46(3), 143-147.
- Dobson, H. E., & Bergström, G. (2000). The ecology and evolution of pollen odors. *Plant System Evolution*, 22(4), 63-87.
- Duncan, D. B. (1955). Multiple range and multiple F tests. *Biometrics*, 11(1), 1-42.
- Erdtman, G. (1954). *An introduction to pollen analysis*. Chronica Botanica Company, Waltham, Massachusetts, USA. 239.
- Erdtman, G. (1960). The acetolysis method – a revised description. *Svensk Botanisk Tidskrift*, 54, 561–564.
- Goodwin, M. (2012). Pollination of crops in Australia and New Zealand. *The Rural Industries Research and Development Corporation*. pp 121.
- Gosling, W. D., Miller, C. S., & Livingstone, D. A. (2013). Atlas of the tropical West African pollen flora. *Review of Palaeobotany and Palynology*, 199, 1-135. <https://doi.org/10.1016/j.revpalbo.2013.01.003>
- Khan, S. U., Zafar, M., Ahmad, M., Anjum, F., Sultana, S., Kilic, O., Ozdemir, F. A., Nazir, A., Yaseen, G., Aabidin, S. Z., & Abbas, Q. (2019). Pollen micromorphological analysis of tribe Acacieae (Mimosaceae) with LM and SEM techniques. *Microscopy Research and Technique*, 82(9), 1610-1620. <http://doi:10.1002/jemt.23327>
- Kirk, W. D. J. (1994). A colour guide to pollen loads of the honey bee Cardiff: *International Bee Research Association*, 54.
- Klein, S., Pasquaretta, C., He, X. J., Perry, C., Søvik, E., Devaud, J. M., & Lihoreau, M. (2019). Honey bees increase their foraging performance and frequency of pollen trips through experience. *Scientific Reports*. 9(1), 1–10. <https://doi.org/10.1038/s41598-019-42677-x>.
- Lau, P., Bryant, V., Ellis, J. D., Huang, Z. Y., Sullivan, J., Schmehl, D. R., Cabrera, A. R., & Rangel, J. (2019). Seasonal

- variation of pollen collected by honey bees (*Apis mellifera*) in developed areas across four regions in the United States. *PLoS One*, 14(6). e0217294. <https://doi.org/10.1371/journal.pone.0217294>.
- Mărgăoan, R., Özkök, A., Keskin, Ş., Mayda, N., Urcan, A.C., & Cornea-Cipcigan, M. (2021). Bee collected pollen as a value-added product rich in bioactive compounds and unsaturated fatty acids: A comparative study from Turkey and Romania. *LWT*, 149, p.111925.
- Mayda, N., Özkök, A., Ecem Bayram, N., Gerçek, Y.C., & Sorkun, K. (2020). Bee bread and bee pollen of different plant sources: Determination of phenolic content, antioxidant activity, fatty acid and element profiles. *Journal of Food Measurement and Characterization*, 14, pp.1795-1809.
- Naghiloo, S., & Nikzat Siahkolaee, S. (2019). Does breeding system affect pollen morphology? A case study in Zygophylloideae (Zygophyllaceae). *Plant Reproduction*, 32(4), 381-390. <https://doi.org/10.1007/s00497-019-00379-4>.
- Rouhakhsh, H., Davarynejad, G., Abedi, B., & Rahemi, M. (2014). Pollen grain morphological characteristics of ten Iranian jujube (*Ziziphus jujuba*) cultivars by using scanning electron microscopy. *Advances in Environmental Biology*, 1333-1339.
- SAS Institute, (2004). *The SAS System* (Version 9.1.3). SAS Institute, Cary, NC.
- Semerdjieva, I., Yankova-Tsvetkova, E., Baldjiev, G., & Yurukova-Grancharova, P. (2011). Pollen and seed morphology of *Tribulus terrestris* L. (Zygophyllaceae). *Biotechnology and Biotechnological Equipment*, 25(2), 2379-2382.
- Shawer, M. B., Taha, E. K. A., Mousa, K. M., Khan, K. A., Ibrahim, S., Hassan, S., & Elnabawy, E. S. M. (2021). Seasonal variations of colony activities linked to morphometric and glands characterizations of hybrid Carniolan honey bee (*Apis mellifera carnica* Pollmann) workers. *Journal of King Saud University-Science*, 33(6), 101543. <https://doi.org/10.1016/j.jksus.2021.101543>.
- Suryanarayana, M. C., Rao, G. M., & Singh, T. S. M. S. (1992). Studies on pollen sources for *Apis cerana* Fabr and *Apis mellifera* L. bees at Muzaffarpur, Bihar, India. *Apidologie*, 23(1), 33-46.
- Taha, E. K. A. (2015). A study on nectar and pollen sources for honey bee, *Apis mellifera* L. in Al- Ahsa Saudi Arabia. *Journal of Entomology and Zoology Studies*, 3(3), 272-277.
- Taha, E. K. A., Al-Kahtani, S. N., & Taha, R. (2019). Protein content and amino acids composition of bee-pollens from major floral sources in Al-Ahsa, eastern Saudi Arabia. *Saudi Journal of Biological Sciences*, 26 (2), 232–237. <https://doi.org/10.1016/j.sjbs.2017.06.003>
- Umber, F., Zafar, M., Ullah, R., Bari, A., Khan, M. Y., Ahmad, M., & Sultana, S. (2021). Implication of light and scanning electron microscopy for pollen morphology of selected taxa of family Asteraceae and Brassicaceae. *Microscopy Research and Technique*, 85(1), 373-384. <https://doi.org/10.1002/jemt.23912>.
- Yankova-Tsvetkova, E., Semerdjieva, I., Baldjiev, G., & Yurukova-Grancharova, P. (2011). On the reproductive biology of *Tribulus terrestris* L. (Zygophyllaceae): embryological features; pollen and seed viability. *Biotechnology & Biotechnological Equipment*, 25(2), 2383-2387. <https://doi.org/10.5504/BBEQ.2011.0030>
- Zaitoun, S., Al-Ghzawi, A. A. M., Samarah, N., & Mullen, R. (2009). Pod production of *Prosopis juliflora* (Sw.) DC. As affected by supplementary and honey bee pollination under arid conditions. *Acta Agriculturae Scandinavica Section B-Soil and Plant Science*, 59(4), 349-356. <https://doi.org/10.1080/09064710802227072>.