

RESEARCH ARTICLE

Taxonomic Study of the Genus *Phlomis* L. (Lamiaceae) in Iraq Based on Trichome and Palynological Characters

Shaemaa Muhi Hasson*, Hanan Ahmed Hadi AL-Qaraawi, Rasha Hashim Hussein

Department of Biology, Faculty of Science, University of Babylon, Iraq.

*Corresponding Author: Shaemaa Muhi Hasson

Abstract

The current investigation included a comparative morphological study of the pollen grains of some *Phlomis* L species distributed in Iraq, as well as study of the Indumentum for species. The pollen grains and trichomes were examined under light and scanning electron microscope. The results showed that there is a limited variability in the characteristics of the pollen grains of the studied species, where the pollen grains were 3-zonocolpate, prolate-subprolate in the shape and the pattern of sculpture (ornamentation) grain surface was reticulate in all species under study, However, the present study showed variability in the dimensions of the pollen grains within the polar and equatorial view. There was also a contrast in indumentum characteristics, and each of these characteristics had a good taxonomic significance to distinguish between the species under study. The results of the current study were found to be taxonomic support to the morphological evidence.

Keywords: Pollen grain, *Phlomis*, Lamiaceae, Indumentums.

Introduction

The Lamiaceae family (or Mint family) is the seventh largest plant family in flowering plants, with about 7,000 species under 250 genera [1]. In Iraq, wild: the family consists of (32) genera and about (140) species, the largest number of species and distribution is genus *Salvia*, which includes (72) species [2].

The flowers in the Lamiaceae family were zygomorphic symmetrical, with (5) united petals and (5) united sepals. It is bisexual, and has a verticillate spiral arrangement [3]. Many of the Lamiaceae plants are aromatic herbs and have essential oils, some species are shrubs, trees and many members of the family are widely cultivated [4].

The pollen grains of Lamiaceae family were divided into two main groups based on Erdtman (1945) study of pollen grains. The division was based on their aperture number and number of nuclei in shed pollen [5]:

- The first group: It is characterized by tricolpate pollen grains, distinctive in subfamily (Lamiaceae).

- The second group: It is characterized by Hexacolpate, which was represented in the subfamily (Nepetoidea).

[6] Confirmed that pollen grains in Lamiaceae family members were Tricolpate, Tetracolpate and Hexacolpate.

The studies of pollen grains were adopted by several researchers to distinguish between Lamiaceae family members. The first study was conducted by [7], followed by several studies, including [8, 9, 10] and [11] of the genus *Salvia* L.

The genus (*Phlomis* L.) is a large genus of Lamiaceae, it is easily recognised [12]. Determined morphological differences within *phlomis* and he split the taxon into two separate genera: *Phlomis* and *Phlomoides* [13].

The genus *Phlomis* L. has more than 100 species, including herbs, shrubs and trees [14], distributed in the north of the warm regions, especially in Europe and Asia. In

Iraq there are about (6 -9) species of the genus, especially in the northern regions of the country. The current study was based on a phenotype study of the pollen grains and Trichomes for six species of genus *Phlomis* L. of the Lamiaceae family, distributed in different parts of Iraq.

Aim of this study is the production of an identification key based on pollen grains and trichomes morphology, for distinguishing between the species of *Phlomis* L.

Materials and Methods

- Pollen grains have been prepared according to the method of [15] with some modification.
- The slides were then examined under a Optica SN 281166 light microscope, and measurements were taken for more than 25 grain per species, and the length of the polar and equatorial axis was measured using the Ocular viewer (40X) and was photographed with the camera mounted on the compound microscope under 40x.
- The study of the pollen grains was also carried out under the scanning electron microscope (SEM). The materials used for the present study from plant specimens were deposited at National herbarium of Iraq (BAG) and Babylon University Herbarium (BUH). The terminology used is mainly that of [16].
- **Indumentums:** The surfaces of both vegetative and flowering leaves were examined by taking part of these leaves for each species and fixing them on a small glass slide.

The slides were coated with gold, then slides were transferred for imaging under the scanning electron microscope. The work was carried out at the University of Kufa / Faculty of Science.

The Results

Pollen Grains Study

Some of good taxonomic evidence of the pollen grains was used to separate the studied species, such as the pollen grain shape and grain size in polar and Equatorial view, as well as the study of the variety in the patterns of sculpture (ornamentation) of pollen grains using light and scanning electron microscopy.

Species

• *Phlomis armeniaca*. LM (Fig: 4): SEM (Fig: 1)

Pollen grains are 3-zonocolpate, (prolate) (P/E: 1.4), medium in size, polar axis length (37.5-50) 48 µm, Equatorial axis (27.5-35) 34 µm. Exine ornamentation (Reticulate).

• *Phlomis brugieri*. LM (Fig: 3)

Pollen grains are 3-zonocolpate, subprolate (P/E: 1.37), medium in size, polar axis length (40-50) 45.5 µm, Equatorial. axis (27.5- 37.5) 33.5 µm. with Exine ornamentation (Reticulate).

• *Phlomis herbventi*. LM (Fig 3): SEM (Fig: 1)

Pollen grains are 3-zonocolpate, prolate (P/E: 1.4), medium in size, polar axis length (45-47.5) 46 µm, Equatorial axis length (22.5-32.5) 32.5 µm., with Exine ornamentation (Reticulate).

• *Phlomis orientalis*. LM (Fig.3): SEM (Fig. 2)

Pollen grains are 3-zonocolpate, Spherical (Prolate), (P/E: 1.38), medium in size, polar axis length (35-47.5) 45 µm, Equatorial axis length (25-37.5) 32.5 µm. With Exine ornamentation (Reticulate).

• *Phlomis praetervis*. LM (Fig.4): SEM (Fig. 2)

Pollen grains are 3-zonocolpate, Subprolate, (P/E: 1.26), medium in size, polar axis length (40-45) 43 µm, Equatorial axis length (30-37.5) 34µm., with Exine ornamentation (Reticulate).

• *Phlomis rigida*. LM (Fig. 4): SEM (Fig, 2)

Pollen grains are 3-zonocolpate, prolate (P/E: 1.44), medium in size, polar axis length (30-37.5) 32.5 µm, Equatorial axis length (20-30) 22.5 µm., with Exine ornamentation (Reticulate).

Indumentums Study

The current investigation presents detailed morphological features of both leaves and corolla trichomes in *phlomis* L. the results of SEM analysis indicate that there are three types of (non-glandular hairs) distributed on

the leaves and gamopetalous leaves. It will be explained as follows:

- Uniserrate simple hairs (unbranched): distributed on both leaf epidermis surface of *Ph. praetervisa* and *Ph. brugieri*, Figure (5 and 6).
- Stellate hairs (branched): distributed two types on the upper surface of the leaves epidermis, and also on the upper surface of petals epidermis, they are as follows:

- With a long stalk stellate hairs: distributed densely on the surface of the upper epidermis of leaves and petals of the species *Ph. brugieri*, *Ph. orientalis* and *Ph. Pravetervisa* Fig. (5 and 6).
- Sessile or Semi-sessile Stellate hairs, also distributed densely on the surface of the upper epidermis of leaves and petals of other species, Fig. (5 and 6).

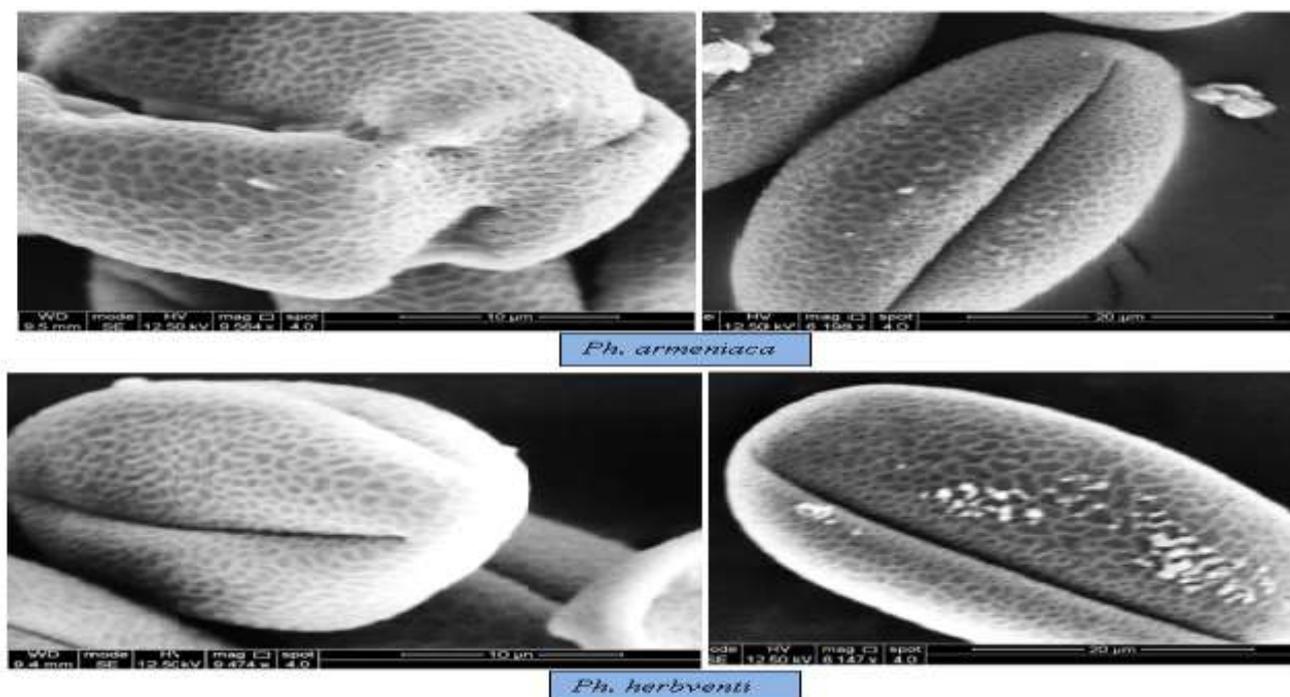


Fig. 1: Variations in pollen characteristics of some species of genus *Phlomis* under study”

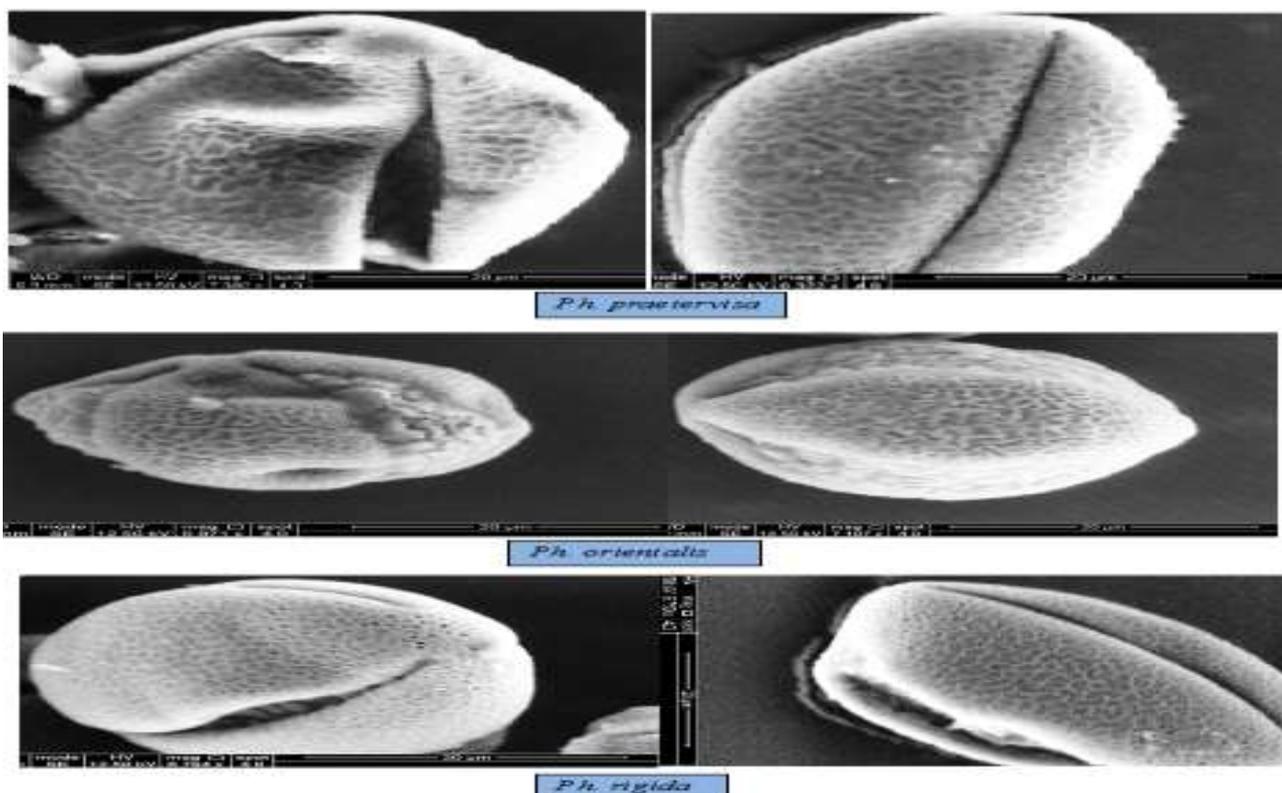


Fig. 2: Variations in pollen characteristics of some species of genus *Phlomis* under study”

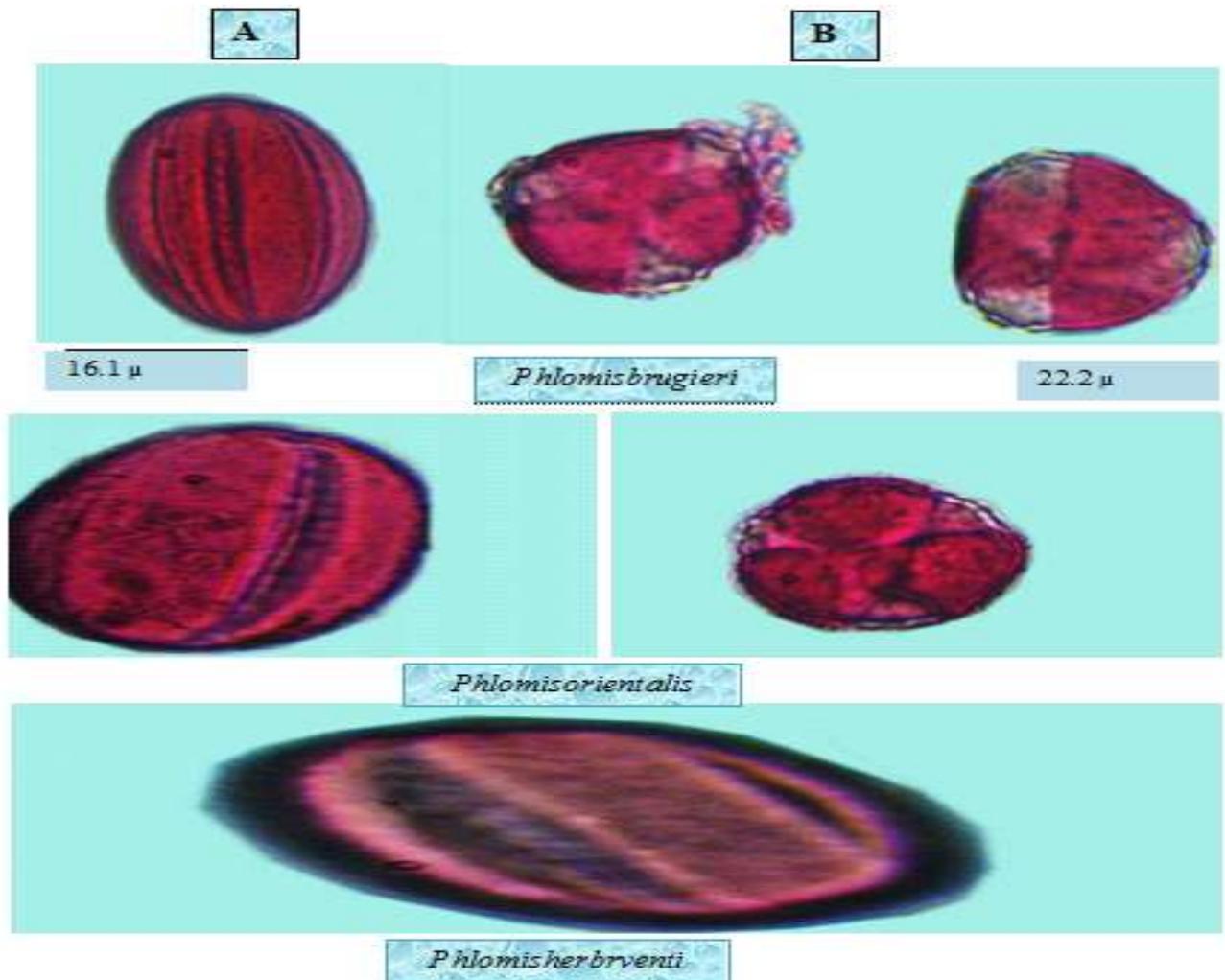


Fig. 3: Variations in pollen characteristics of some species of genus *Phlomis* studied (40x) A: Equatorial view B: Polar view

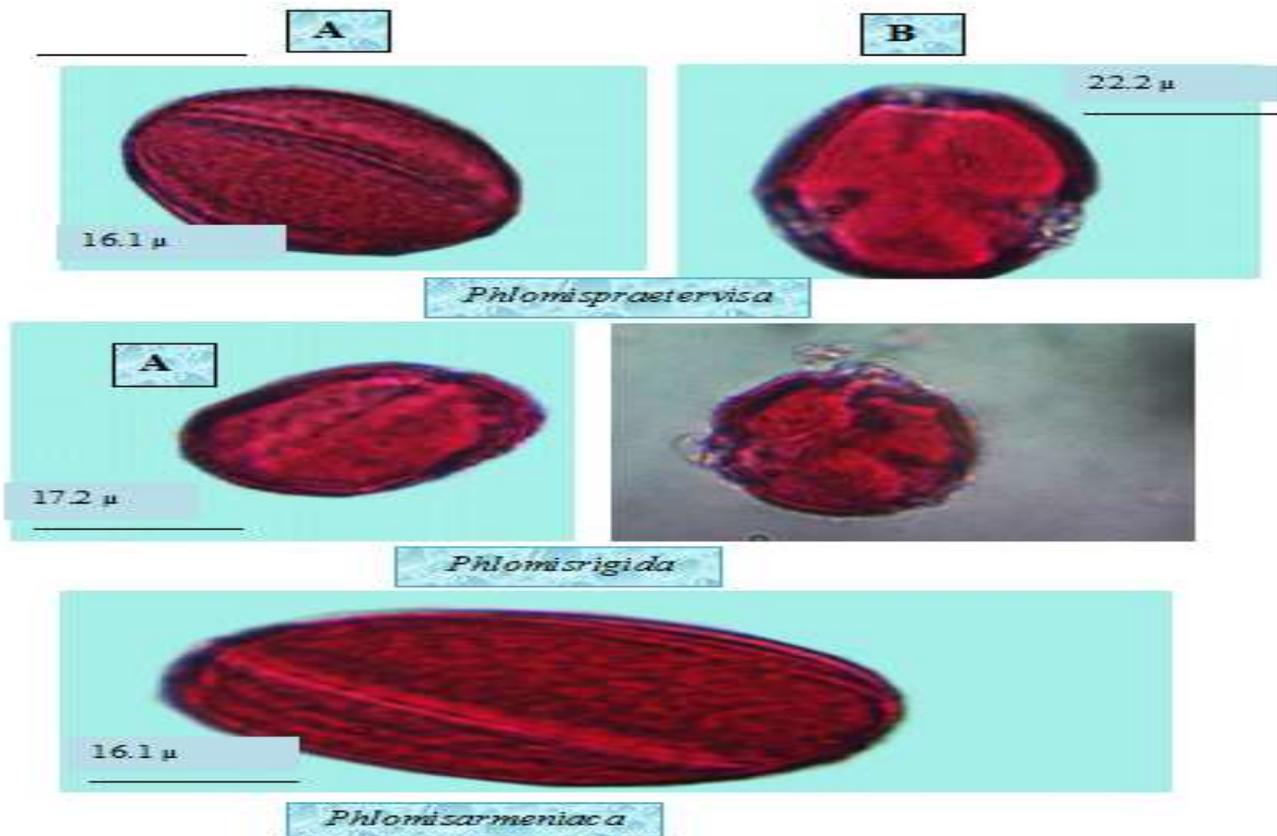


Fig. 4: Variations in pollen characteristics of some species of genus *Phlomis* studied (40x) A: Equatorial view B: Polar view

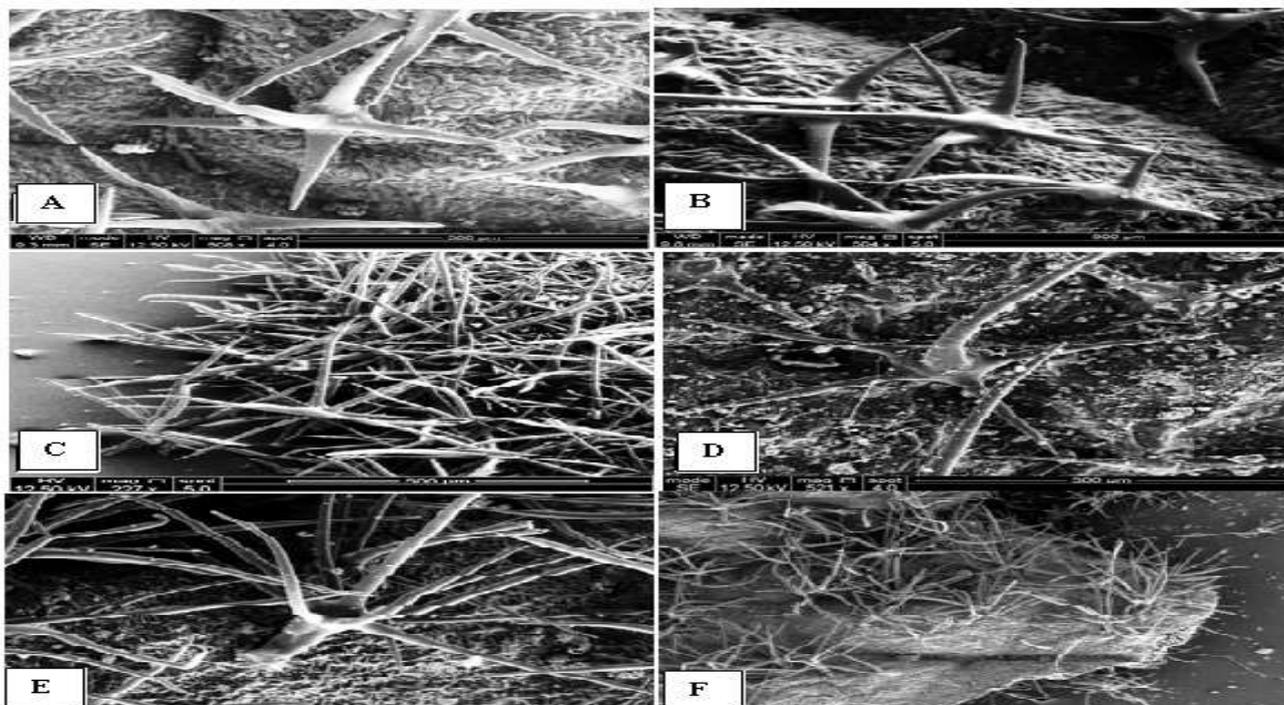


Fig. 5: Variations in trichomes characteristics of some species of genus *Phlomis* studied under SEM. A-D (*Ph. armenica*), E-F (*Ph. brugieri*.)

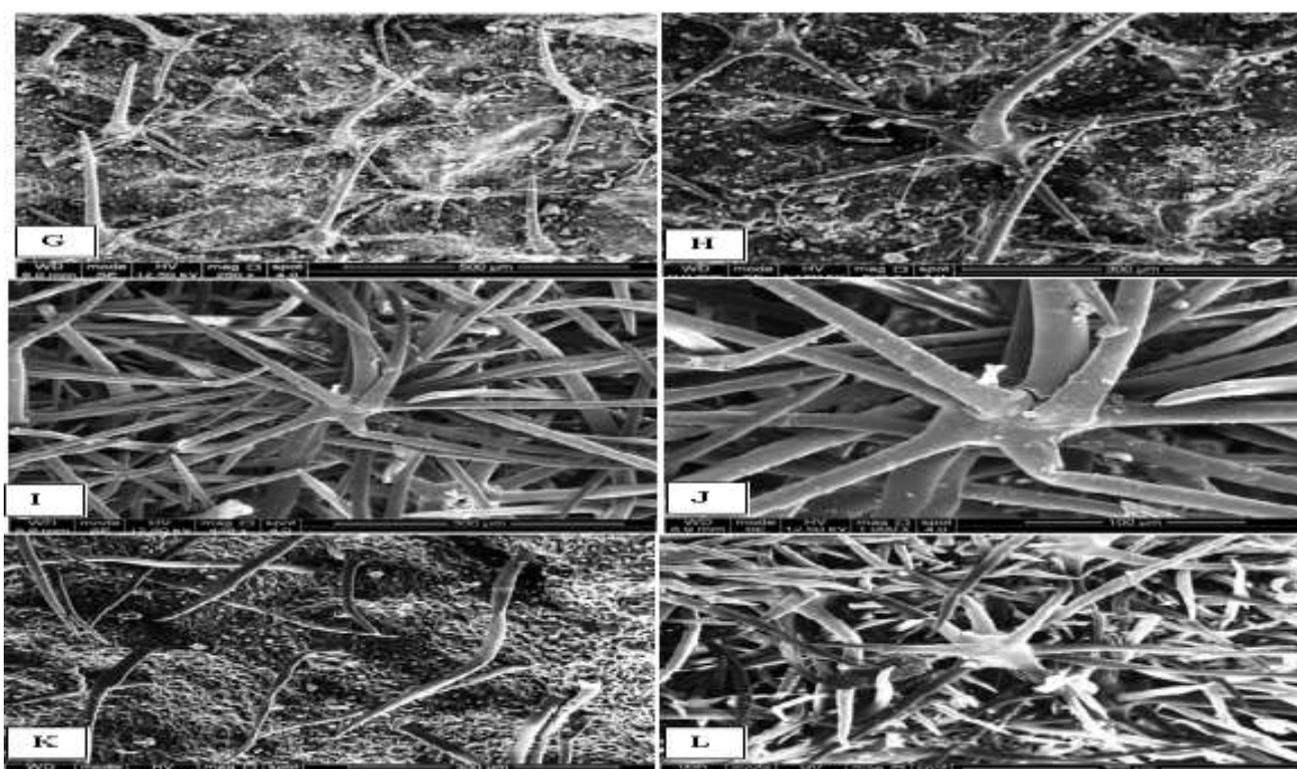


Fig. 6: Variations in trichomes characteristics of some species of genus *Phlomis* studied under SEM. G-H (*Ph. herbventi*), I-J (*Ph. orientalis*), K-L (*Ph. praetervisata*)

Discussion

In this investigation, pollen grains of six species belonging to genus *Phlomis* L. of the family: Lamiaceae have been studied. Palynology is considered a unique source of morphological information in that no other dataset can provide so great an amount of information from so little material in so short time [17]. The results of this study showed that the pollen grains are 3-zonocolpate in all

species of genus *Phlomis* L., This corresponds to the study of [18] which indicated that the apertures of *Phlomis* pollen grains are 3-colpate, [17] pointed out that tricoplate is the main and basic type found in eudicot, while other aperture types are regarded as derived among eudicot, such as 5-colpate, 6-colpate, porate, colporate. Depending on the measurement of the axis length measured in the pollen grain, the size is determined

within a specific range between the very small and giant as indicated by [19].

The results of the current study showed that the pollen grains of the studied species of (medium size) . The dimensions of the pollen grains, in the polar view, the maximum rate of the axis length (40) μm in *Ph. Praetervisa* and minimum rate was (27.5) μm. In *Ph.rigida*, but in Equatorial view the maximum rate of the polar axis length (48) μm in *Ph.armenica* and minimum rate was (32.5) μm. in *Ph.rigida*. While the length of the equatorial axis was (34) μm. at a maximum in *Ph.armenica* and (22.5)μm. At the minimum in *Ph.rigida*.

Exine ornamentation was characterized as a Reticulate in all the studied species; this corresponds to the study of [18] and [10]. Thus, the results of the present study confirmed the taxonomic importance of the pollen grains in addition to the morphological and anatomical studies. The study of the phenotypic characteristics of the pollen grains is an effective attempt to clarify the genetic evolutionary relationships between species [20], and more important after the use of electron microscope SEM in the separation of several taxonomic taxa.

Table 1: Variations in pollen characteristics of the species studied are measured by micrometer

Aperture type	Pollen grain shape P/E	Equatorial view			Polar view	Species	s
		P/E	E. axis length	P. axis length			
Colpate	(prolate)	1.4	34(35 - 27.5)	(50 - 37.5) 48	(40 - 27.5) 34.5	<i>Ph. armenica</i>	1
Colpate	prolate	1.37	33(37.5 - 27.5)	(50 - 40) 45.5	(35-30)32	<i>Ph. brugieri</i>	2
Colpate	prolate	1.4	32.5(32.5 - 22.5)	(47.5 - 45) 46	(40- 32.5)37	<i>Ph. herbventi</i>	3
Colpeate	Spherical Prolate)	1.38	32.5(37.5 - 25)	45(47.5 - 35)	32.5(35-27.5)	<i>Ph. orientalis</i>	4
Colpate	Sub-prolate	1.26	34 (37.5 - 30)	(45 - 40) 43	(47.5 - 35) 40	<i>Ph. praetervisa</i>	5
Colpate	Prolate	1.44	22.5(30 -20)	(37.5 - 30) 32.5	27.5 (32.5-25)	<i>Ph. rigida</i>	6

Pollen grain size	Medium	Medium	Medium	Medium	Medium	Medium
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Based on the results of the present study, the vegetative and petals leaves of the studied species were characterized by the predominance of non-glandular hairs are Uniserrate simple hairs and stellate hairs, This is consistent with several studies on *phlomis* L. species, including [21] and [22] study. The type of trichomes had an effective taxonomic role in separating the two species *Ph. praetervisa* and *Ph. Brugieri* from the others studied species. In addition, the densities of the distributed of stellatetrichomes had a good taxonomic role

in the separating *Ph. orientalis* and *Ph. Praetervisa*, and were more density on the petals leaves of *Ph. orientalis* of *Ph. praetervisa*.

Conclusion

The results of the study confirmed the taxonomic importance of the size of the pollen grain based on the polar and equatorial axis dimensions in species separation, as well as the effective role of the type and density of trichomes distributed on the leaves and petals. The values outside the brackets represent the average either inside the minimum and maximum.

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