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Pollen morphology of the genus *Centaurea* L. (Asteraceae) in Middle and North of Iraq

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ABSTRACT

The current research was designed to study of pollen grains of the genus *Centaurea* L. of the Asteraceae. The study included 17 species (*C. cardunculus, C. scleroleis, C. cana, C. balsamita, C. solsititialis, C. sinaica, C. behen, C. thracica, C. laxa, C. aggragata, C. vergata, C. ibrica, C. bruguiriana, C. Phyllocephalla, C. rigida, C. hylolepis and C. hylolepis ssp. Hylolepis*). Samples utilized in the present study were collected from different areas in the middle and north of Iraq. A compound microscope was used to examine the slides of samples and records the required measurements. The properties of samples such as the length of the polar axis, the equatorial axis and the thickness of the grain wall were of each pollen grain were measured by using an ocular micrometer and photographed by the camera on the microscope. The results showed that the pollen grains were tricolporate, as it turned out that the size of pollen grains, the diameter of germination pore, the length of the colpus, the surface sculpturing, as well as the thickness of the wall (exine) of the pollen have a taxonomic value to isolate the species of the genus from each other.

Keywords: *Centaurea*, Asteraceae, Pollen grain, Exine and Colpus.

Introduction

The term palynology means the science of pollen and spores and this term was initially used by (Hyde and Willims, 1945). Palynology is an applied science, and the study of pollen is important to the ecologist, medical scientist and agriculturalist (Moore and Webb, 1978) and it is being the basic science for several other science branches including anatomy, morphology, cytology, genetics, etc... (Erditman, 1971). The characteristics of the pollen grains are essential in defining taxonomic categories in terms of morphological study and anatomy of pollen grains, the number of apertures and their position. Furthermore, the pollen grain is significant for classifying orders, families and tribes (Blackmore et. al, 1984). Asteraceae is the largest family in all vascular plants, and the number of its genera is estimated at around 950 genera, and the number of its species may reach 20000 species distributed in most parts of the world and all environments. Pollen morphology has been proved to be useful in the systematic of the Asteraceae family, as well as that of some of its genera and species (Moore et. al., 1991). Based on Exine structure and Sculpturing Wagentiz (1955) divided the genus *Centaurea* into eight subgenera called *Serratula, Centaurium, Scabiosa, Crupina, Jasea, Dealbata, Montana,* and *Cynus*. Al-Katib (1988) stated that 242 wild species and 52 cultivated species were found in Iraq. The genus *Centaurea* L. (Asteraceae) is represented by 500-600 taxa worldwide (Rendle, 1976; Heywood, 1979; Hickey & King, 1981). Numerous studies have attempted to study the genus *Centaurea* L. in Iraq. Zohary (1950) classified 23 species of the genus *Centaurea*. Al_Rawi (1964) summarized the previous literature on the genus *Centaurea* with a handout of 34 species. It is apparent from the literature review that numerous investigations have been conducted on the *Centaurea*. However, studies on the pollen morphology are still limited. Therefore, this morphological study aims to explore the structure of the pollen grains of the genus *Centaurea* L. For this purpose, a compound microscope and ocular micrometer were used to study different samples. The outcomes of the present study can be used in the classification of new groups.

Material and methods

The pollen grains of the genus *Centaurea* species were examined in the present study were taken from fresh samples collected during field trips from different regions located in the middle and north of Iraq. The samples collections period was one year between 2018 to 2019. The samples were preserved with 70% ethyl alcohol. Sample preparation technique adopted by AL-Mayah (1983) was followed in the current work with some modifications as follows: ripe and unfolded florets were selected for

several flowers heads and placed on a glass slide and put a few drops of water on it. The florets were opened with the use of tow dissection needles and some parts such as the corolla and pistil were removed and the stamina cylinder was extracted from them. Then, they transferred to another glass slide containing a drop of the dye safranin-glycerin. The anthers were opened to extract the pollen grains, after which the parts of the anther were removed, and the cover slide was placed. The slides were examined under a compound microscope and measurements were taken for (25-30) pollen grains. The length of the polar axis, the equatorial axis, the thickness of the grain wall, the diameter of germination aperture, the number of apertures, the surface sculpturing and the length of the groove (colpus) of each pollen grain were measured using an ocular micrometer and photographed by the camera on the microscope was under oil immersion lens.

Results

It was evident from the current study that the pollen in the studied species is of tricolporate type as it has three germination pores. As for the shape of the pollen grain, the ratio between the length of the polar axis and the length of the equatorial axis was extracted, it was clear that most of the pollen grain of the studied species had prolate-spheroidal shape, except the species *C. sinaica* and *C. ibrica* was spheroidal in shape and species *C. aggragata* pollen grain was prolate Fig. (3) & Fig. (4). As the surface sculpturing, the studied species can be divided into two groups. The first group includes the echinate type and includes three species which are *C. scleroleis*, *C. balsamita* and *C. thracica*. The second group includes the rest of the species Table (1), Fig. (3) & Fig. (4). The pollen grains of the studied species fall into small and medium category (Erdtman, 1971). Based on this the species *C. sinaica*, *C. vergata* and *C. phyllocephalla* fall into small categories. While the rest of the species fall into the medium category, Where the lowest length of the polar and equatorial axis is recorded in species *C. phyllocephalla* (22) & (21.5) micrometer respectively, while the highest length of the polar and equatorial axis is recorded in species *C. cardunculus* (52) & (50) respectively Table (1). Based on the ratio of the length of the polar axis and the length of the equatorial axis, the species could be isolated into two groups, the first group: in which the ratio less than one, including four species, which is: *C. solsititialis*, *C. behen*, *C. aggragata* and *C. bruguiriana*. While the second group includes the rest of the species Table (1) Fig (1) & Fig (2). As for the diameter germination pore, species *C. cana* was recorded the minimum average pore diameter (6) **µm**, While species *C. rigida* recorded the maximum average pore diameter (9**) µm**, while the rest of the species showed a clear interference with each other Table (1). From the observation the thickness of the outer layer (exine), it appears that it is different in the amount of thickness of the exine between the studied species, where the minimum average of exine thickness was recorded in the species *C. phyllocephalla* which reached to (2**) µm,** while the maximum average was recorded in the species *C. behen*, as it reached (5) **µm,** and the results of the rest of the species were interference with each other, Table (1). With regard to the average groove (colpus), by looking at the results of the studied species, we showed that species *C. vergata* had a minimum average of the colpus length (8) **µm**, while the maximum average of the colpus length had been recorded by species *C. beben* which reached to (15) **µm,** while the rest of the species intertwined with each other Table (1). The variation of polar axis length and equatorial axis length of studied species of the genus *Centaurea* were presented in Fig. (1) to Fig. (2).

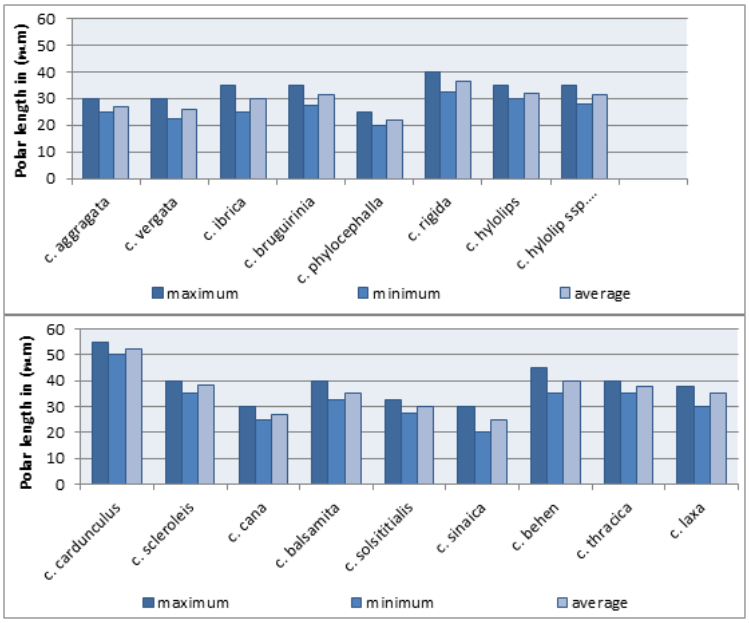

Figure (1) variation of the polar axis length of pollen grains in the studied species of the genus *Centaurea*

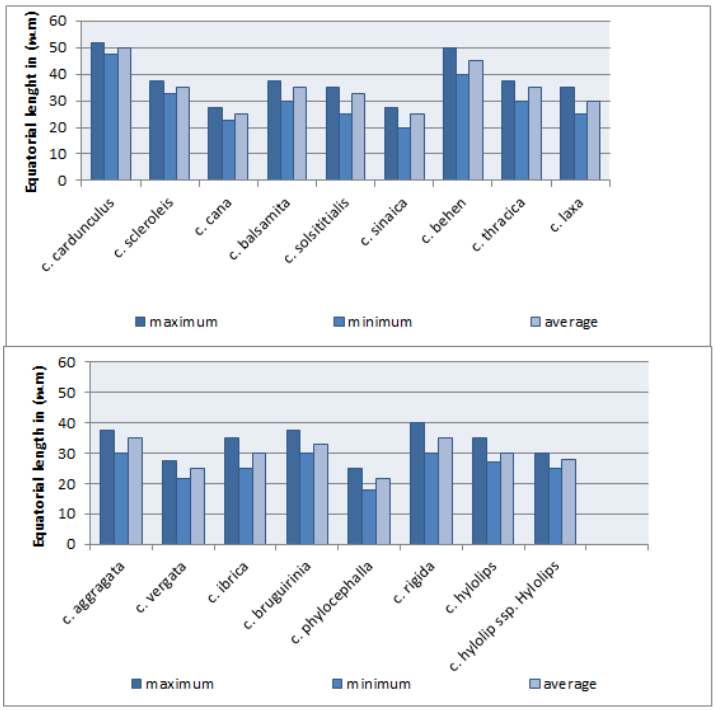

Figure (2) variation of the equatorial axis length of pollen grains in the studied species of the genus *Centaurea*

Table (1) pollen characters of the genus *Centaurea* specie measured in Micrometer

Species	P **(µm)**	E **(µm)**	P/E **(µm)**	Shape	Exine thickness **(µm)**	Surface sculpturing	Diameter of pore **(µm)**	Number of pore	Colpus **(µm)**
C. cardunculus	50-55 (52)	47.5-52 (50)	1.04	Prolate- spheroidal	5-7.5 (4)	psilate	5-9 (7)	3	12.5-17.5 (14.5)
C. scleroleis	35-40 (38)	32.5-37.5 (35)	1.09	Prolate- spheroidal	3-6 (4.5)	echinate	7.5-10 (8.5)	3	10-15 (12.5)
C. cana	25-30 (27)	22.5-27.5 (25)	1.08	Oblate- spheroidal	2.5-5 (3.5)	psilate	5-7.5 (6)	3	5-12.5 (10)
C. balsamita	32.5-40 (35)	30-37.5 (35)	1	Prolate- spheroidal	2.5-5 (3)	echinate	5-10 (7)	3	7-15 (10.5)
C. solsititialis	27.5-32.5 (30)	25-35 (32.5)	0.9	Prolate- spheroidal	2-5 (3.5)	psilate	3.5-7.5 (5)	3	5-15 (9)
C. sinaica	20-30 (25)	20- 27.5 (25)	1	spheroidal	3-6 (4.5)	psilate	2.5-7.5 (5)	3	7.5-15 (12)
C. behen	35-45 (40)	40-50 (45)	0.8	Prolate- spheroidal	2-7.5 (5)	psilate	5-12.5 (8.5)	3	7.5-20 (15)
C. thracica	35-40 (37.5)	30-37.5 (35)	1.07	Prolate- spheroidal	2.5-5 (3.5)	echinate	5-10 (7.5)	3	5-12.5 (9)
C. laxa	30-37.5 (35)	25-35 (30)	1.1	Prolate- spheroidal	2-5 (3)	psilate	5-9 (7.5)	3	7.5-15 (12)
C. aggragata	25-30 (27)	30-37.5 (35)	0.7	Prolate	2-3.5 (2.5)	psilate	5-12 (8)	3	7-13.5 (10)
C. vergata	22.5-30 (26)	21.5-27.5 (25)	1.04	Oblate- spheroidal	1-3.5 (2)	psilate	3.5-6.5 (4.5)	3	6-10.5 (8)
C. ibrica	25-35 (30)	25-35 (30)	1	spheroidal	2.5-5 (3.5)	psilate	5-10 (7.5)	3	7-15 (11.5)
C. bruguiriana	27.5-35 (31.5)	30-37.5 (33)	0.95	Prolate- spheroidal	3-5.5 (4)	psilate	5-11 (8.5)	3	7.5-15 (12)
C. Phyllocephalla	20-25 (22)	18-25 (21.5)	1.02	Prolate- spheroidal	1.5-3 (2)	psilate	5-10 (7.5)	3	5-13.5 (9)
C. rigida	32.5-40 (36.5)	30-40 (35)	1.04	Prolate- spheroidal	2.5-5 (3.5)	psilate	5-12 (9)	3	6.5-15 (11)
C. hylolepis	30-35 (32)	27-35 (30)	1.06	Prolate- spheroidal	2-4.5 (3)	psilate	5.5-12 (8.5)	3	7-13.5 (10)
C. hylolepis ssp. hylolepis	28-35 (31.5)	25-32 (29)	1.08	Prolate- spheroidal	2-5 (3.5)	psilate	5-10 (7.5)	3	6-12.5 (8.5)

P: Polar axis length
E: Equatorial axis length

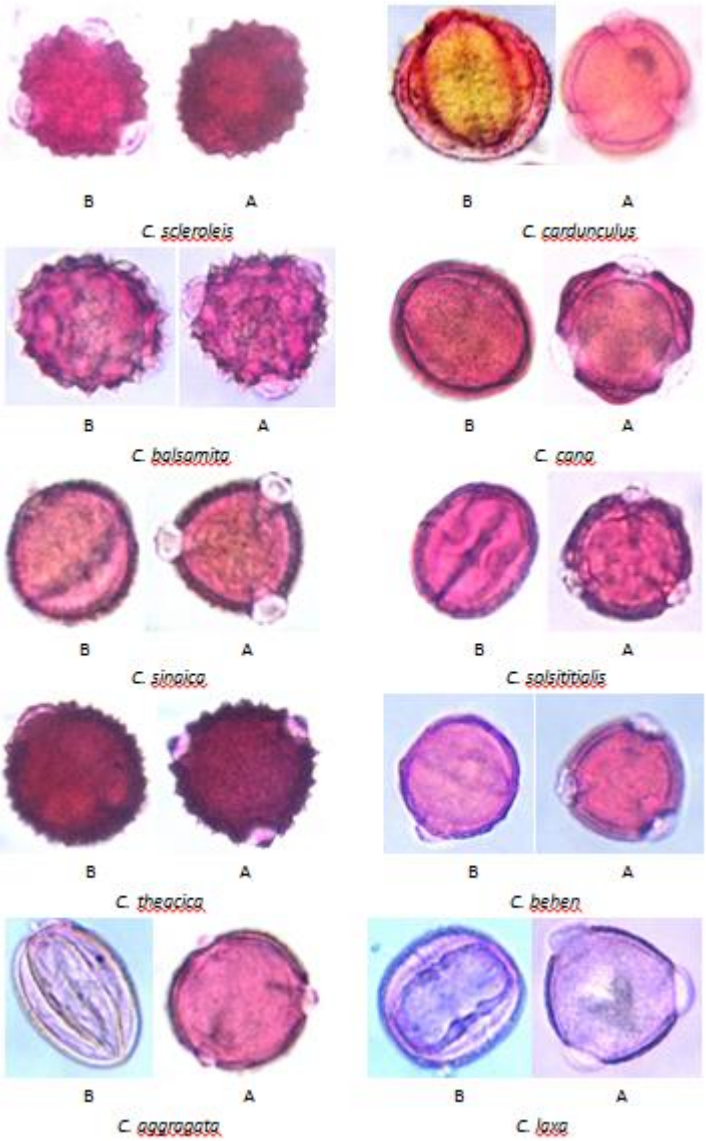

Fig (3) Pollen morphology of the genus *Centaurea* observed under light Microscope
A: Equatorial view B: Polar view

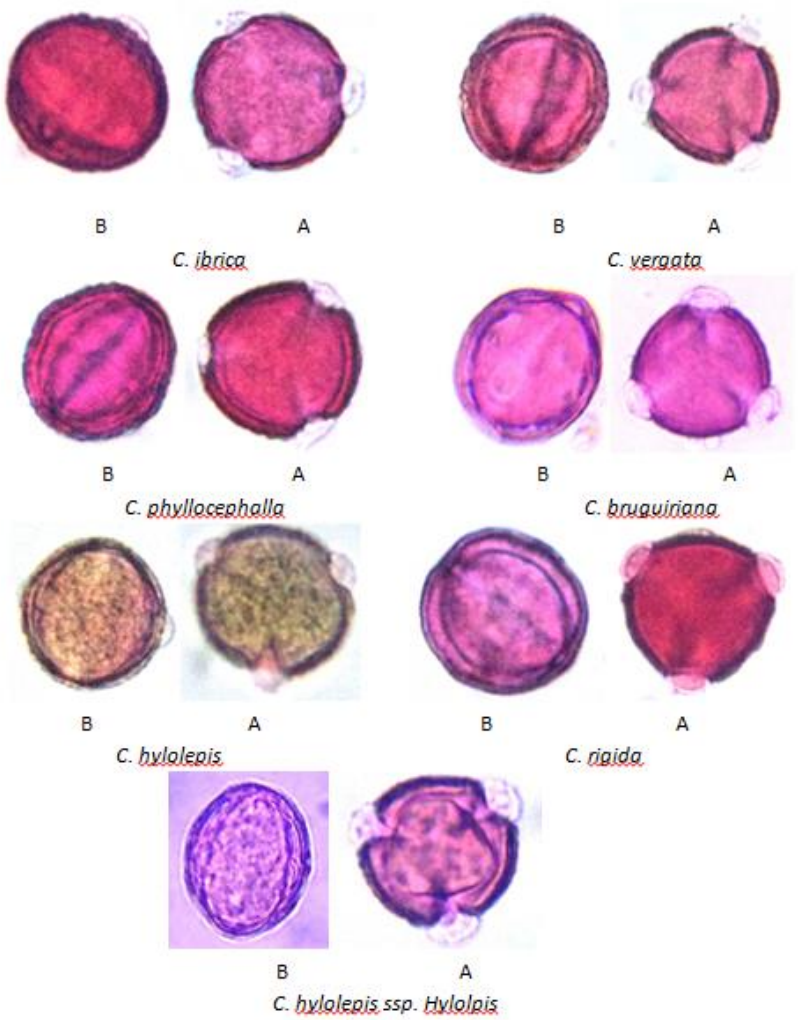

Fig (6) Pollen morphology of the genus *Centaurea* observed under light Microscope
A: Equatorial view B: Polar view

Disscusion

The current study revealed that the pollen grains of the *Centaurea* species studied in Iraq is Tricolporate, and this type is common in the Asteraceae (Wodehouse, 1935; Erdtman, 1971) and is consistent with what was proposed by (Abu-Seraj, 2008; Shabestari *et. al*, 2013; CitaK *et. al*, 2019). The shapes of pollen varied clearly between the studied species, so that the species divided into groups and each group includes a number of species which confirms the taxonomic importance of the pollen shapes, and this consistent with what was emphasized by (Shabestari *et. al*, 2013). The surface sculpturing of the pollen grain was characterized by being Psilate in most species except for species *C. scleroleis*, *C. balsamita* and *C. thracica* where it was Echinate, and on this basis it was possible to divide the studied species into two groups, as the common type of the tribe *Cynaraea* to which the *Centaurea* belongs is the echinate type in addition to the presence of the Psilate type (Skvarla, *et. al.*, 1977) and this is consistent with the current study, but it doesn't support previous divisions. From the foregoing, it is clear that the surface sculpturing has good taxonomic importance. Pollen sizes represent diagnostic characteristics in Asteraceae and have a relationship to dividing into lower taxonomic categories as indicated by (Qureshi *et. al.*, 2002). And for the species of the genus *Centaurea*, it showed a clear variation in term of pollen size, where the three species could be distinguished *C. sinaica*, *C. vergata* and *C. phyllocephalla* which fall into the small category from the rest of the studied species, which fall into the medium category, while (Torres, 2000) indicated that there is a positive linear relationship between pollen size and pistil length in Asteraceae, and that such a relationship is not clear in this study. The thickness of the pollen grain walls and the diameter of the germination pore showed a clear difference between the species, on the basis of which the studied species could be divided into two groups. The ratio of the average polar axis / equatorial axis also gave taxonomic significance on the basis of which the studied species were isolated into two groups as for the length of the Colpus, it was of taxonomic importance. (Wodehouse, 1935) indicate that the

length of Colpus has a relationship with the size of the pollen grain, since large pollen grains have long Colpus compared to small pollen grains. Such a relationship was not evident in this study.

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الصفات المظهرية لحبوب اللقاح لجنس *Centaurea* L. من العائلة (Asteracea) في وسط وشمال العراق

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الملخص

تناول البحث الحالي دراسة حبوب اللقاح لجنس *Centaurea* L. من العائلة Asteraceae. وشملت هذه الدراسة 17 نوعا من الجنس وهي (*C. carduncul* و *C. scleroleis* و *C. cana* و *C. balsamita*, و *C. solsititialis*, و *C. sinaica*, و *C. behen* و *C. thracica* و *C. laxa* و *C. aggragata* و *C. vergata* و *C. ibrica* و *C. bruguiriana* و *C. Phyllocephalla* و *C. rigida* و *C. hylolepis* و *C. hylolepis ssp. Hylolepis*). جمعت العينات قيد الدراسة من مناطق مختلفة من وسط وشمال العراق. وتم استخدام المجهر الضوئي المركب لفحص العينات وتسجيل القياسات المطلوبة. حيث تم قياس صفات حبوب اللقاح للعينات المدروسة مثل طول المحور القطبي والمحور الاستوائي وسمك جدار الحبة وطول الاخدود لكل حبة لقاح باستخدام المقياس الدقيق للعدسة العينية Ocular Micrometer وتصويرها بوساطة الكاميرا المثبتة على المجهر. وأظهرت النتائج أن حبوب اللقاح كانت من نوع ثلاثية الفتحات Tricolporate، حيث اتضح أن حجم حبوب اللقاح وقطر فتحة الإنبات Germination Pore وطول الاخدود Colpus والزخرفة السطحية Surface Sculpturing وكذلك سمك الجدار Exine لحبوب اللقاح لها قيمة تصنيفية لعزل أنواع الجنس عن بعضها البعض.