

Morphology, Anatomy, Palynology and Seed Micromorphology of Libyan Endemic *Bellis sylvestris* var. *Cyrenaica* (Asteraceae)

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Abstract: *Bellis L.* Belonging to the family Asteraceae and has been included in subtribe Asterinae (tribe Astereae). In the present work, the morphological characters, stem, petiole and leaf anatomy, pollen micromorphology of Endemic *Bellis sylvestris* var. *cyrenaica* from Libya have been investigated on light and scanning electron microscope. *Bellis sylvestris* var. *cyrenaica* is an endemic species from Libya and limited information about the native taxon. All morphological, anatomical and palynological characteristics are firstly determined in this study. Palynology study showed that pollen characters were found medium size, oblate spheroidal, tricolporate and echinate ornamentation of the studied taxon. Seed micromorphological features were also given

Keywords: Endemic *Bellis sylvestris* var. *cyrenaica*., Asteraceae, Morphology, Anatomy, Palynology, Libya.

1. INTRODUCTION

Bellis L. popularly is known as Daisy flower belonging to the family Asteraceae and has been included in subtribe Asterinae -tribe Astereae (Bremer, 1994). This genus also known as a traditional wound herb (Ai- Douri and Al-Essa 2010) and it was used for the treatment of bruises, broken bones and wounds (Mitich 1997). It has also been used in traditional folk medicine for the treatment of sore throat (Uysalet *al.* 2010) headache, common cold, eczema, wound healing, diarrhea, peptic ulcer, gastritis, rheumatism, asthma, hemorrhoids and as a vulnerary expectorant and laxative (kavalcioglu *et al.* 2010, karakas *et al.* 2012, Melikoğlu 2015). *Bellis L.* is represented

15 species on Europe and Mediterranean region which is the largest center of diversity of the genus but is commonly found as invasive plant in North America (Webb 1976).

The previous studies reported that pollen and seed micromorphologies are efficient in the systematics of the Asteraceae family (Shabestari *et al.* 2013, Frangiote-Pallone and De Souza 2014). Akyalçinet *al.* (2011) studied the pollen morphology of six *Achillea L.* species and their results show that the pollen grains were oblate- spheroidal, prolate, sub-prolate and generally tricolporate, though at times tetracolporate or even pentacolporate. Joujehet *al.* (2019) reported that the pollen grains of *Centaurea* from Syria were similar in shape, size, aperture characters and symmetry, while they differ in color, exine ornamentation pattern and density of the spines. Karahan (2020) found that the pollen grains Some morphological and anatomical studies of the Asteraceae family have been carried out. Metcalfe and Chalk (1950) reported that due to the diversity of their habitats, Asteraceae species show various anatomical differentiations, especially in the structures of leaves. Fahn (1979) observed that secretory structures have an important diagnostic value in Asteraceae. These secretory structures such as ducts, cavities, idioblasts, laticifers, hydathodes, extrafloral nectaries and trichomes presented diagnosis value at genus level (Castro *et al.* 1997). It has been suggested that trichome micromorphology was useful in the systematics of Asteraceae (Ciccarelli *et al.* 2007). Napp-Zinn and Eble (1978) carried out a detailed ultrastructure study of the glandular and non-glandular hairs of twenty genera of the Anthemidae. Glandular hairs in Asteraceae may be widely distributed both on the vegetative and the floral parts (Ciccarelli *et al.* 2007).

Karahan (2020) studied the leaf anatomy of *Bellis* and showed that the upper and lower epidermis consist of uniseriate cells with different sizes and shapes. Also he found that the mesophyll of *B. annua* and *B. perennis* was unifacial, while the mesophyll of *B. sylvestris* was bifacial. In his study of the stem he found that the epidermis consists of 1-2 layered of oval cells.

In Libya, according to the Flora of Libya the genus *Bellis* is represented by three taxa with two species and one endemic variety (Jafri, 1983). *B. sylvestris* var. *cyrenaica* is an endemic variety collected from Cyrenaica which is located in the Northeast of Libya. This taxon is a very distinctive but there is limited information about it.

B. sylvestris var. *cyrenaica*, the object of the present study is an endemic plant in Libya that looks like *B. sylvestris* var. *sylvestris* from which it differs by its leaves and peduncle. The present study aims to give a first detailed account of the morphological, anatomical and palynological characteristics of *B. sylvestris* var. *cyrenaica* variety to see whether these data can be important from the taxonomic point of view (no study about the anatomy, palynology and micromorphology of *B. sylvestris* var. *cyrenaica* appear to be carried out until now).

2. MATERIALS AND METHODS

Five specimens were collected of *B. sylvestris* var. *cyrenaica* during their flowering and fruiting time, between 2017 and 2019. The research area was given in Fig. 1 according to Davis's Grid system (Davis 1965-1985). Specimens were dried according to standard herbarium techniques and are deposited in the Herbarium of Cyrenaica Herbarium (CH) at the Botany Department, Sciences faculty, Benghazi University). The plant samples were preserved in 70% ethanol for anatomical analysis. Transverse sections of stem, petiole and leaf samples were prepared. Slides were observed with a light microscope. For anatomical description, the terminology of Metcalfe and Chalk (1957) was used. Pollen grains were obtained from mature anthers of dried flowers and measured under a light microscope. The measurements of the polar axis (P), the equatorial diameter (E), the ornamentation, the number of apertures, the aperture type, the spine length, the spine width for 10 pollen grains were conducted under a light microscope. The terms used for describing the seed surface patterns have been adopted according to Koulet *et al.* (2000). All photographs were taken at the Central Laboratory of Alexandria University, Egypt.

3. RESULTS

3.1. Morphological Characters

B. sylvestris var. *cyrenaica* Béguinot in Atti Accad. Ven. Trent. Istr. 9:25.1916.

Type: Described from Libya: Cyrenaica

B. sylvestris var. *cyrenaica* is a perennial rosette herb, 4-18 cm long, with glaucous-colored leaves covered with dense hairs (Fig. 1). Stems are short rhizomes, covered by hairs. The leaves are spatulate, 4-10 cm in length, exstipulate, petiolate up to 3 cm petiole, sinuate-dentate with obtuse apex, covered with glandular and non-glandular trichomes. Stems bearing one peduncle. Peduncles are monocephalic, slender and 2-10 cm long. Involucre 22-27 arranged in two rows, lanceolate. Ray flowers ligules white with purplish tinge, 8-14 mm long. Disc corollas 2.5x1-1.5 mm long and yellow (Fig 1). Cypselas, brown, elongated oval shape, 1.8-2 mm long and 0.6-0.8 mm wide, quite strongly pubescent.



Fig1. General morphology of *Bellis sylvestris* var. *cyrenaica*

3.2. Pollen Morphology

Under light microscope, the pollen grains are isopolar, radially symmetrical and prolate-spheroidal. Their apertures are tricolporate; The ornamentation is echinate in the studied taxon. The spine base is determined as circular–elliptic to circular. The polar axis (P) ranged from 22-25 μm (23.5) μm and equatorial axis (E) ranged from 22-23 μm (22.5) μm . The spine lengths ranged from 1.7 μm to 2.3 μm and the spine widths ranged from 1.2 μm to 1.95 μm . Under scanning electron microscope, the distance between spines ranged from 1.17 μm to 2 μm (Fig. 2).

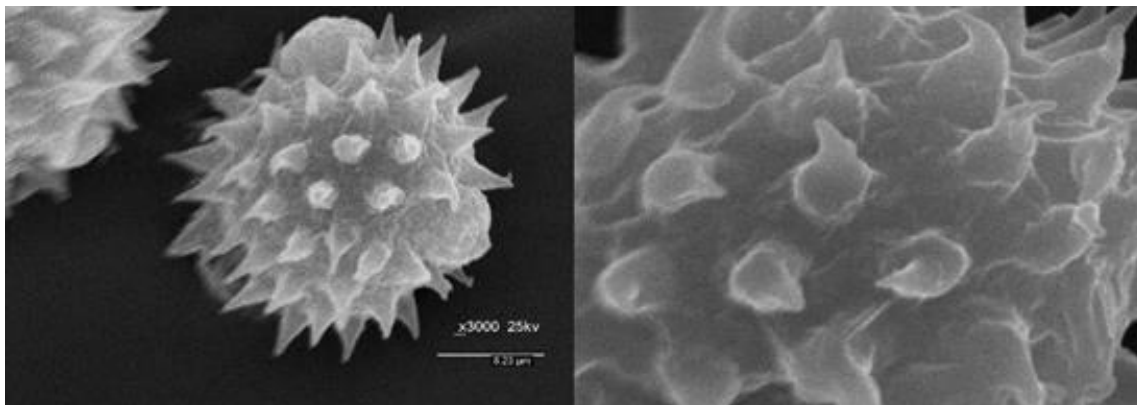


Fig.2. Scanning electron micrographs (SEM) of pollen grains of *B. sylvestris* var. *cyrenaica*.

3.3. Seed Morphology

Seeds have ovate shape with reddish brown color. The size of seeds was 0.5- 0.7 x 0.2-0.4 mm. The seed coat ornamentation is reticulate (Figure 3 A, B).

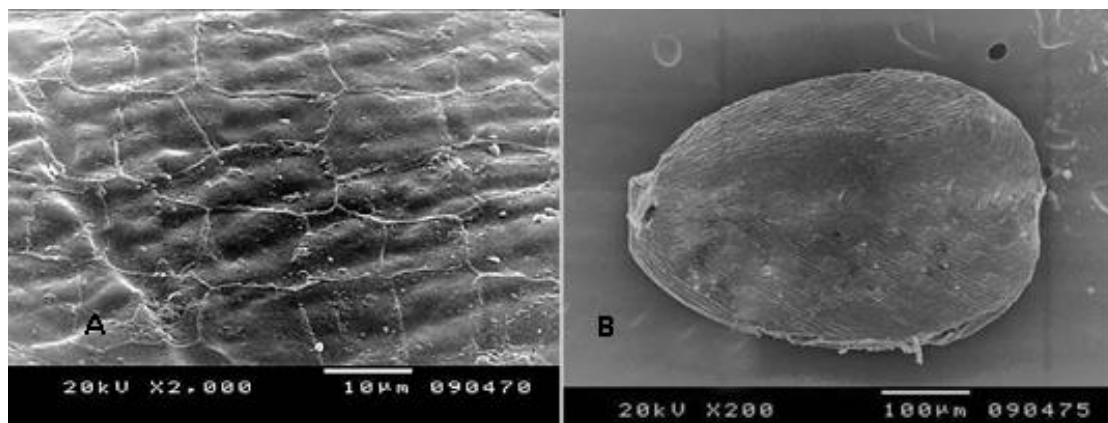


Fig.3. Scanning electron micrographs (SEM) of the seed. A-General appearance; B- Surface ornamentation of *B. sylvestris* var. *cyrenaica*.

3.4. Anatomical Characters

Stem: The transverse section of the stem, the epidermis consists of 2-5 layered lignified irregular cells. There were eglandular hairs on the epidermis. Under the epidermis, the cortex contains multi layers of different sized and shaped parenchyma cells. The cortex cells rich by starch and organic material and ends with endodermis layer. The vascular system was of open collateral type. The pith was wide and composed of polygonal or oval parenchymatous cells (Fig. 4A, B).

Leaves: The transverse section of the leaf of studied taxon showed that the upper and lower epidermis composed of uniseriate cells with different sizes and shapes covered by thin cuticle. There were non-glandular multicellular, uniseriate hairs on both sides and anomocytic stomata. Mesophyll tissue was differentiated as palisade and spongy parenchyma cells. Under the upper epidermis, the mesophyll contains 3-4 layers of palisade parenchyma and 3-5 layers of spongy parenchyma. The vascular bundle was located solely in the parenchymal tissue in the middle vessel region. The xylem arranged in straight line in a single layer and face towards the upper surface, while the phloem faces the lower epidermis. (Fig. 4B-C).

Peduncle: Peduncle cross-section was Elongated carusel in shape. The epidermis was composed of one layer of different sized cells, with thickened outer walls and were covered with non-glandular, multicellular uniseriate hairs. Epidermis was followed by collenchymatous hypodermis and parenchymatous cortex. In the central cylinder 12- 13 of collateral and open vascular bundles were arranged in a circle followed by hollow parenchymatous pith (Fig. 4 D).

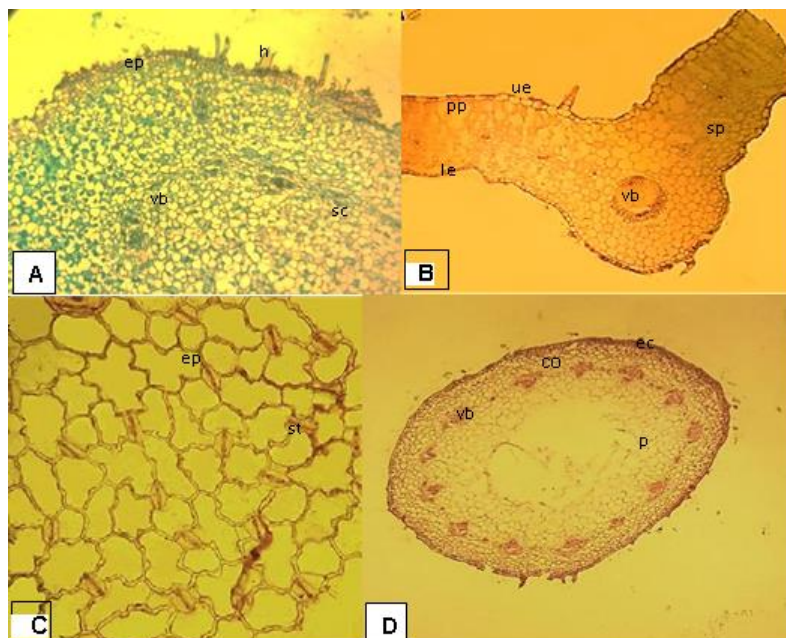


Fig.4. Light microphotographs of the transverse sections of *Bellis sylvestris* var. *cyrenaica*: A. Stem, B. Leaf, C. Stomata D. Peduncle; co-cortex, ep-epidermis, le-lower epidermis, p-pith, pp-palisade parenchyma, sc-storage cells, sp-spongy parenchyma, ue- upper epidermis, vb-vascular bundle.

4. DISCUSSION

From searching literature, it is clear that no study to date has examined endemic *Bellis sylvestris* var. *cyrenaica* except general taxonomic properties of the taxon. The morphological properties of investigated taxon were in agreement with the description of the plant in Flora of Libya. The anatomical characters of the studied plant are reported for the first time in the present study. The findings of *Bellis sylvestris* var. *cyrenaica* were compared with anatomical studies made on the genus *Bellis* in literatures. The results of palynological study showed that the palynological characters obtained were generally similar to the results of Akyalçin *et al.* (2011).

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Citation: Ghalia T. El Rabia, Seham H. Elbadry, (2020). "Morphology, Anatomy, Palynology and Seed Micromorphology of Libyan Endemic *Bellis Sylvestris* var. *Cyrenaica* (Asteraceae). *International Journal of Advanced Research in Botany (IJARB)*, 6(1), pp.25-29. DOI: <http://doi.org/10.20431/2455-4316.0601004>

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