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Morphological and Anatomical Study of *Origanum cyrenaicum* Endemic to Libya

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Abstract:

The present study provides a detailed analysis of the morphological and anatomical characteristics of *Origanum cyrenaicum* Beg.et Vacc., an endemic species of the genus *Origanum* L. in Libya. This research represents the first documentation of these features for this particular species. The anatomical features of *O. cyrenaicum*, including the stem, petiole, and leaf, were thoroughly examined and presented in this paper. Cross-sections of stems and leaves were carefully analyzed and complemented with illustrations and photographs. Given the limited information available on this native taxon, this study contributes valuable insights into the unique characteristics of *O. cyrenaicum*.

Keywords:

Origanum cyrenaicum, endemic plant, morphology, anatomy, Libya.



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1. INTRODUCTION

The *Origanum* genus, a member of the Lamiaceae family, is commonly referred to as oregano and marjoram. With approximately 900 species worldwide, this genus is characterized by its richness in bitter substances and essential oils (Davis, 1982). *Origanum* species are either subshrubs or perennial herbs, featuring multiple stems, ascending or erect growth patterns, leaves that are either sub-sessile or petiolate, and flowers that form in verticillasters, clustered in dense or loose spikes, arranged in a paniculate or corymbiform inflorescence. Widely recognized as a popular spice under the vernacular name 'Oregano', *Origanum* plants are utilized globally.

Recent research has highlighted the traditional use of oregano due to its antimicrobial, cytotoxic, and antioxidant properties (Davis, 1982). Studies focusing on nutlet micromorphology within the Lamiaceae family have indicated that nutlet features, such as shape and surface sculpturing, can be valuable at various taxonomic levels. While the type of sculpturing is often deemed the most taxonomically significant nutlet characteristic (Gönüz A and Özürgücü, 1999), factors like color, size, and shape are generally considered unimportant due to their lack of variation or the presence of random or excessive variations (Navarro and El Qualidi, 2000).

The nutlets of Lamiaceae can be categorized based on their surface ornamentation, which includes irregularly reticulate, reticulate, undulate, and wrinkled-colliculate patterns. Apart from their morphological characteristics, the micromorphological traits of mericarps are valuable for taxonomic classification of the sections within the genus *Origanum* and differentiation of species within those sections (Ecevit-Genç, et al. 2020).

The anatomical features of vegetative organs are also crucial for characterizing Lamiaceae taxa (Kahraman et al., 2010). Glandular hairs and their distribution (Werker, 2006), stomatal distribution, and other anatomical attributes offer significant taxonomic information (Venkateshappa & Sreenath, 2013). Furthermore, these features play a vital role in understanding phylogenetic relationships across various taxa (Pandey & Misra, 2014). Fuad (2023) examined the leaf anatomical characteristics of 19 Lamiaceous species from different tribes using light and scanning electron microscopy to accurately identify the plants. The stem displays a quadrangular shape, with collenchyma tissue covering a significant area at the corners and one to two layers between them (Kahraman, et al. 2007 and Dinç et al. 2008). The vascular cambium is visible in two *Origanum* species, but not in the other Oran (1996). Two *Origanum* species that were studied have a pith cavity at the center of their stems. A previous study by Cantino (1990) reported the presence of starch in *O. onites* L. Additionally, it was noted that the cells of the palisade parenchyma were not elongated as in typical dicot leaves, but rather short, resulting in large intercellular spaces between them. The types and morphology of trichomes in the indumentum hold taxonomic significance in Lamiaceae (Bosabalitis and Kokkini 1997).

The leaves of the three taxa exhibit dorsiventral mesophyll. However, the number of palisade layers and spongy layers varies among the taxa. In some taxa, the upper epidermal cells are notably larger than the lower ones, whereas in *O. rotundifolium*, they are nearly equal in size. Additionally, it has been noted that the cells of the palisade parenchyma are not elongated as in typical dicot leaves; instead, they are short, resulting in large intercellular spaces between them. The types of trichomes and the morphology of the indumentum hold taxonomic significance in Lamiaceae (Kahraman, et al. 2007 and Metcalfe & Chalk 1972). In *O. vulgare* ssp. *viride*, the palisade tissue covers approximately half of the mesophyll. This plant is endemic to Libya, yet there have been no morphological or anatomical studies conducted on this species. The current research aims to identify the macro- and micro-morphological characteristics of this species.

2. MATERIALS AND METHODS

During the period of November to January 2016-2017, plant samples were collected from the Al-Jabal Al-Akdar (wadi Alkouf) area. The Botany department identified the collected samples of *O. cyrenaicum*, which were then dried using standard herbarium techniques. These dried samples were stored in the Cyrenaica Herbarium (CH) at the Botany Department, Sciences Faculty, Benghazi University. To observe the dried mature nutlets of *O. cyrenaicum* using a Scanning Electron Microscope (SEM), a SEM microscope model (Joel, JFC 1100) was utilized. The nutlets were directly mounted on aluminum stubs using double-sided adhesive and coated with a thin layer of gold. The coated nutlets were then examined, and SEM micrographs were captured at 200× and 500× magnification. The nutlet surface patterns were described according to the terms adopted from [12]. The photographs of the nutlets were taken at the Central Laboratory of Alexandria University in Alexandria, Egypt (Figure 2). Anatomical investigations were conducted on the cross-sections of the petiole, stems, and leaves. All sections were manually prepared and stained with safranin and fast green solution. The sections were then photographed using an Olympus BX-53 research microscope.

3. RESULTS

3. 1. MORPHOLOGICAL CHARACTERS (Fig.1.)

The shrublet is perennial, growing in rocks, with branches 20-40 cm tall, quadrangular, ascending, minutely puberulent. The leaves are small with short petioles, ovate, obovate, or elliptic, with a rotund or often subcordate acute base, entire and cartilaginous margined, ciliate. The larger leaves are ovate, orbicular, or oblong with an obtuse apex, rounded base, entire and cartilaginous margined, mostly greenish and scabrid, ciliate along the veins and margin. The verticils are oblong, few flowered, short pedunculate, slightly pendulous on one side, some white purplish, ovate-elliptic, concave, closely imbricate, longer than the calyx, hairy, not punctuate. The calyx is glandular-pellucid and adpressed minutely glandular hairy, bilabiate, with the upper lip obtuse, ciliate and slightly repando-tridentate, and the lower lip small, bilobed, with long weak hairs in the throat. The corolla is pink, twice as long as the calyx, glabrous, with a thin (slender) tube; lobes are subequal, entire, obtuse, with the upper broadly 3-lobed and the lower smaller and shortly 2-lobed, stamens and styles exerted.



Fig. 1 . General view of *Origanum cyrenicum*

3.1.1. NUTLET CHARACTERS (Fig. 2.):

The nutlet characters are as follows (Fig. 2): Nutlets are 4-5 mm in length, 2-3 mm in width, broad ovate, and dark brown in color. The hilum is small, located almost in the center. The surface ornamentation is irregularly reticulate and glabrous. The anticlinal walls are raised and smooth with some globules, while the periclinal walls are depressed with some wrinkles.

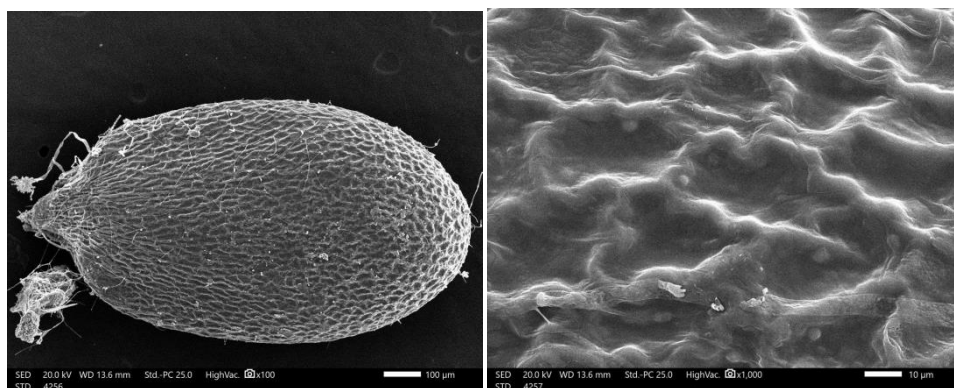


Fig. 2. Scanning electron micrographs (SEM) of Nutlets in *Origanum cyrenaicum*

3.2. ANATOMICAL CHARACTERS

3.2.1. Stem Anatomy (Figure 3 A&B):

The stem's cross-section is distinctly quadrangular in shape. The outermost layer, known as the epidermis, is made up of either oval or irregular cells arranged in a single layer and is covered by a thin layer called the cuticle. The epidermis also contains either single-celled or multicellular hairs, which can be either non-glandular or glandular in nature. Just below the epidermis lies the collenchyma tissue, which consists of 6-8 layers of oval-shaped cells. The cortex tissue, located beneath the collenchyma, is composed of both oval and quadrangular cells with thin walls. Adjacent to the cortex parenchyma, there are small clusters of phloem sclerenchyma cells positioned above the phloem, and the vascular cambium cannot be distinguished. The xylem elements have thick walls. Lastly, the pith is made up of both large and small cylindrical, polygonal, or oval-shaped parenchymatous cells.

3.2.2. Petiole Anatomy (Fig. 3 C):

The petiole exhibits a somewhat reniform shape in cross-section, with a slightly curved or concave adaxial surface. Both the adaxial and abaxial epidermis consist of single-layered cells, with numerous glandular and non-glandular hairs present on the ovoidal-rectangular shaped epidermal cells. The parenchymatous cortex is composed of 4 to 8 layers of cells. A large crescent-shaped vascular bundle is located at the center of the petiole, surrounded by parenchymatous bundle sheets. The vascular bundle type observed is collateral.

3.2.3. The anatomy of leaves (Fig. 3 D&E)

reveals that in cross-sections, the upper and lower epidermises consist of uniseriate, quadrangular, and oval cells. Notably, the lower epidermal cells are larger than those in the upper epidermis. Both epidermises are adorned with non-glandular and glandular hairs (Fig. 2 D,E). Additionally, 2-3 layers of collenchyma cells can be found in the midrib, situated between the upper and lower epidermis. The leaves exhibit a dorsiventral structure. The palisade tissue is made up of single-layered cylindrical cells abundant in chloroplasts. Meanwhile, the spongy parenchyma, characterized by large intercellular spaces, is irregular in shape.

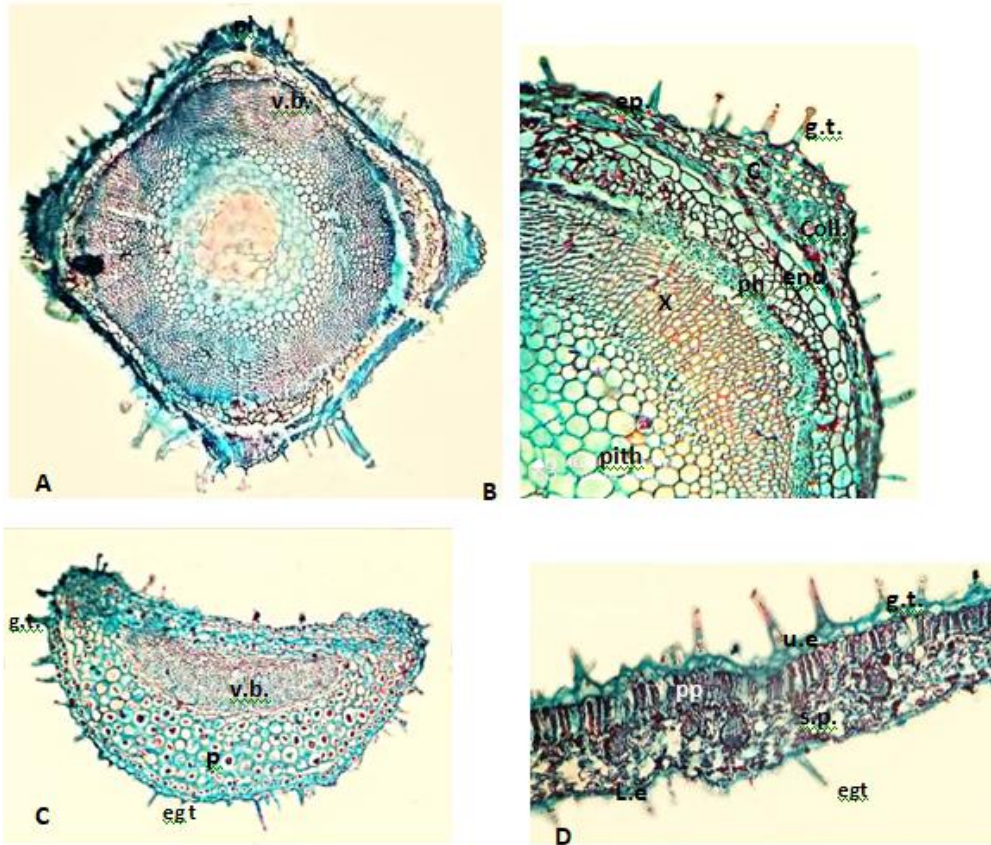


Fig. 3. Stem, petiole and leaf anatomy of *O. cyrenaicum*. A,B-Cross-section of the stem; C-Petiole and (D,E) the leaf. (C-cortex, Co-collenchyma, ep:-epidermis, egt-eglandular trichome, gt-glandular trichome, Le-lower epidermis, P-parenchyma, Ph-phloem, Pp-palisade parenchyma, Sp-spongy parenchyma, Ue-upper epidermis, X-xylem).

4. DISCUSSION

As far as we are aware, the morphological and anatomical traits were newly recognized for the endemic species (*Origanum cyrenaicum* Beg. et Vacc.) apart from the general taxonomic characteristics of the taxon. The morphological features of the studied taxon corresponded with (Jafri, S. M, and A. El Gadi. 1985). the plant description in the Flora of Libya. Our results were juxtaposed with anatomical research conducted on the genus *Origanum* in various publications.

5- CONCLUSION

In conclusion, the purpose of this study was to provide a detailed analysis of the morphology of the endemic plant *Origanum cyrenaicum*. The morphological characteristics of this particular taxon were found to be inadequately described in the Flora of Libya by Jafri and El Gadi (1985).

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