



### MULTIVARIATE ANALYSIS OF MICROSCOPIC ELEMENTS OF HYOSCYAMUS MUTICUS L. SUBSP. FALEZLEZ (COSS.) MAIRE, POWDER FROM ALGERIA

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Hyoscyamus muticus L. Subsp. falezlez (Coss.) Maire is a toxic species, very well represented in the Algerian Sahara. It is one of the richest Solanaceae in tropanic alkaloids with pharmaceutical and economic interests. Microscopic test plays a leading role in species identification and was recently associated with statistical analyses. The work aims to study the microscopic characteristics of this plant from three harvesting stations in Saharan Algeria. Microscopic examination concerned all the organs' powder of the plant and was supplemented by a multivariate analysis study of elements. The distribution of microscopic elements was assessed using the analysis of main components (PCA). The associations between microscopic elements and the observations were made by dimension reduction analysis and the comparative study between the different stations and organs was performed by the Khi-deux test. The results of the botany essay revealed a high degree of structural variability, in addition to the elements usually found during microscopic observation, some elements identified during our analysis have been cited rarely in the literature, like sclereidal fibers and prisms that were numerous in our samples. Certain elements were not mentioned, like cluster prism crystal of calcium oxalate, glandular trichome with unicellular head and multicellular stalk, and the bulky single-cell trichome element with a rounded tip. The multivariate study allowed us to classify elements powder depending on the degree of correlation. It also facilitated the identification by developing a key of two groups and two subgroups of elements powder (depending on the presence or absence of some elements). No significant morphological or statistical differences were found between the three stations. However, their distribution between organs revealed differences. At the end of our study, the results contributed to enriching the plant botany data Keywords: Anatomy and histology, Algeria, Hyoscyamus, Multivariate analysis

### **INTRODUCTION**

*Hyoscyamus muticus* L. Subsp. *falezlez* (Coss.) Maire is a Saharian species, known under the vernacular name of Saharian

Henbane, in Arabic: El betima, in Tergui: Afléhlé. It is a perennial herbaceous plant found mainly in the Western and Central Sahara of Southern Algeria<sup>1-3</sup>. It belongs to the Solanaceae botanical family, characterized by

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the production of Atropine; an important compound belonging to tropane alkaloids<sup>4,5</sup>.

The Saharan Henbane is a perennial or annual herbaceous plant of the Saharan sandy pastures<sup>2</sup>. It is fleshy and very branched, up to 1.5m high. Its stems are numerous erect, thick, and succulent. The leaves are pale green to yellowish, petiolated, slightly fleshy, and arranged in a spiral<sup>6</sup>. The inflorescence is represented by a dense terminal and unilateral cyma and the flowers are bisexual pentamers with a hairy, tubular, and striated calyx. The corolla is a zygomorphic penta lobed with a black violet at the top of the tube and veined with white<sup>3</sup>, As shown in **Fig. 1**.

*Hyoscyamus muticus* L. Subsp. *falezlez* (Coss.) Maire has been extensively studied and exploited in countries neighboring Algeria: Egypt<sup>6,7</sup> and Sudan<sup>8,9</sup>. Microscopic analysis of the powder elements of the Saharan Henbane concerned only some Pharmacopoeia, the Egyptian and the American<sup>3,6</sup>.

In Algeria, since its description during the colonial period after the poisoning during the Flatters mission and that of Quezel and Ozanda<sup>2</sup>, very few studies have been undertaken on the botanical aspects of *Hyoscyamus* L. Subsp. *falezlez* (Coss.)

Maire in Algeria, they were mainly interested in the chemical profile of the plant. One study concerned the Ahaggar region, another the Adrar region<sup>10</sup>, and recently the Djanet region<sup>11</sup>. Other studies have only been addressed in ethnobotanical surveys<sup>12-16</sup>.

Botanical studies have considerably augmented in recent years, and the botanical test is the first diagnostic step in Pharmacognosy and helps guide the identification of a plant drug<sup>17,18</sup>. It is a standard method used and validated by several treaties<sup>17</sup>. It allows microscopic observation to identify the characteristic elements of a drug: it is an easy technique to implement and requires very few resources. Recently, the botanical trial was associated with multivariate statistical studies, a combination that made additional confirmation of the absence or presence of a botanical characteristic by the different correlations studied between the elements of a plant powder<sup>19,20</sup>. It leaves no room for doubt if the plant sample is from different organs.

The lack of studies on Saharian Henbane in Algeria, her wide distribution in the Algerian Sahara, and its importance in the pharmaceutical industry prompted us to explore the elements of its powder by microscopic botanical analysis and a multivariate study.



Fig. 1: Hyoscyamus muticus L. Subsp falezlez (Coss) Maire, harvested in Adrar, Algeria.

### MATERIALS AND METHODS

#### Sample processing

The aerial parts of Saharian Henbane that constitute the part used, as well as the root sample, were harvested in March 2018 in South-West of Saharian Algeria, in the following areas: Abadla: (GPS: 31.281, -2.439) 28.205. and Adrar (GPS: -0.172) and Tamanrasset (GPS: 26.913. 0.921). The identification was carried out by Pr. N. Benabadji in the laboratory of Ecology and Management of Natural Ecosystems, Faculty of Natural Sciences, by correlating the botanical characteristics of the plant with those described in the literature<sup>17,18</sup>. Specimens of plant samples were deposited in the drug user of the Pharmacognosy laboratory under reference number 61.

The organs (roots, leaves, stems, flowers, and fruits), were separated and spread on wooden boards lined with tulle for aeration and dried in the shade for several days before being finely crushed. Organoleptic characters were described, using Pharmacognosy and Botany treaties<sup>2,17,18</sup>.

The microscopic analysis concerned all harvested organs, but the multivariate analysis was conducted only on the aerial parts.

### **Microscopic observation**

A small amount of the powder from each organ was collected and filed between slide and slat in two drops of lactic acid and observed at x400 magnification, under an optical binocular microscope with reference DM 500, LED-type light source, from Germany<sup>21</sup>. The lignified elements take on a yellow coloration, however the secretion products turn orange-red<sup>21</sup>.Twenty-seven observations were concerned for all the samples, and nine observations for each organ (stem, leaf, flowers) of plants from the three harvesting stations.

### Multivariate statistical analysis and its purpose

The aim of associating statistical analysis with microscopic observation is to facilitate the diagnosis and identification of plants. The study of the correlation between the different elements of the powder and the microscopic observations allows the authentication of the species and the elimination of strange elements<sup>22</sup>. Data entry and analysis were performed by IBM SPSS Statistics 21.0. Data coding of the microscopic elements was carried out using the following variable codes (powder elements): Alb: Albumen, Ami: starch grains, Crista: Crystalline cells, Epi: Epidermis, EpiC: Epicarp, FibL: Simple fiber, FibS: Sclereidal fiber, Li: Phloem, Pap: Papilleous cells, Par: PoiSB: Bifurcate Parenchyma, glandular trichome with unicellular head and multicellular stalk. Glandular PoiSUP: with trichome unicellular head and multicellular stalk, PoiSUU: Glandular trichome with unicellular head and unicellular PoiTEA: Multicellular eglandular stalk. trichome with a round end, PoiTEP: Multicellular eglandular trichome with a pointed end, Pol: Pollen, Pris: Prism crystal of calcium oxalate, PrisC: Twin crystals of calcium oxalate, PrisM: cluster prism crystal of calcium oxalate, SecO: Orange secretions, Vai: Xylem vessels.

The distribution of microscopic elements according to the organs was conducted, using: comparing the means, and the Analysis of Principal Components (APC). The objective is to reduce the number of variables without losing information<sup>23,24</sup>.

Data is projected in a smaller dimension than the initial matrix, using MINITAB 19 software. The association between the microscopic elements and the observations was made, by a dimension reduction analysis. The position of those observations on both axes allowed us to make an identification key<sup>23,24</sup>.

The comparative analysis of the microscopic elements between the three stations (Abadla, Adrar, and Tamanrasset) and organs was conducted using the Khi-two test, to compare their frequencies in the samples.

### **RESULTS AND DISCUSSION**

### Results

### *Hyoscyamus muticus* L. *Subsp. falezlez* (Coss.) Maire root powder analysis

The root powder is yellowish, fibrous, and difficult to crush. Observed by an optical microscope, the various elements are shown in **Fig. 2** and represented by many parenchymal cells (Par) containing starch grains (Ami), isolated, yellowish, often fragmented fibers with a narrow lumen (FibL), many pitted xylem vessels (Vai), and calcium oxalate crystals with various forms (prism (Pris) or cluster prism (PrisM), and twin crystals (PrisC)). Crystal oxalate is either isolated or contained in crystalline cells (Crista).

### *Hyoscyamus muticus* L. Subsp. *falezlez* (Coss.) Maire leaf powder analysis

The leaf powder is greenish-brown, reminiscent of cumin color, with an unpleasant, weakly bitter flavor. Leaf microscopic elements powder are depicted in Fig. 3 and 4. It contained fragments of the epidermis (Epi) with polygonal cells, rectilinear walls, and smooth cuticle. Small anisocytic stomata were observed in the epidermis of lamina and midrib, both in surface view, as shown in Fig. 3. Numerous refractive calcium oxalate crystals were identified, in different sizes and shapes: cubic and twin prisms, and cluster prisms, either isolated or included in crystalline cells. A fragment of parenchyma with orange secretions, a rare fragment of collenchyma, simple fibers (FibL), and sclereidal fibers

(FibS) were found. Conducting tissues were represented by Xylem vessels; pitted instead of punctuated, isolated, or bound to fragments of Phloem (Li) and parenchyma (Par). Different glandular hairs were observed:

• Rare large-sized uniseriate multicellular eglandular trichomes often broken, smooth-walled, 2-3 articles, and are of two types; pointed end (PoiTEP), with a broad base, and rounded end (PoiTEA).

• Two types of glandular trichomes: with unicellular head and unicellular stalk (PoiSUU), few in number, and glandular trichome with bi or multicellular head and unicellular stalk, with a rounded or a pointed end, some showed strangulation (PoiSUP), as shown in **Fig. 4**.

• Large bifurcated glandular trichomes with double or triple bifurcation, and with strangulation (PoiSB).



**Fig. 2:** Elements of *Hyoscyamus falezlez* root powder, observed under an optical microscope, at 10x40 magnification. Calcium oxalate crystals in various forms: twin (a), cluster prism (b), crystalline cells (c), parenchymal cells containing starch grains (d), isolated simple fibers, yellowish often fragmented with narrow lumen, and smooth wall (e, f), xylem pitted vessels (g). Descriptions of the elements were in the article.



Fig. 3: Powder elements of *Hyoscyamus muticus* leaves observed under an optical microscope at 10x40 magnification.

Epidermis fragment with small anisocytic stomata; seen from two angles, face (a) and profile (b), refractive calcium oxalate crystals in different sizes and shapes: cubic and twin (d), prisms (e), cluster prisms (f), crystalline cells (c), xylem vessels of various types: spiral (most numerous) (g), pitted (rarely) (h), simple fibers (k), and sclereidal (j), collenchyma fragments (l).

### *Hyoscyamus muticus* L. Subsp. *falezlez* (Coss.) Maire stem powder analysis

The powder of the stem is yellowish, fibrous in appearance, difficult to grind also, and often remains crushed. It is marked by numerous spiral pitted xylem vessels, rarely reticulate, multiple calcium oxalate crystals in crystalline cells, and parenchyma fragments containing starch grains. Isolated orange secretions or secretions (SecO) contained in parenchymal cells may occur, and rare glandular trichomes were observed.



Fig. 4: The different glandular and eglandular trichomes of *Hyoscyamus muticus* leaves powder, observed by optical microscope; at magnification 10x40. Uniseriate multicellular eglandular trichomes of wide size, often broken, smooth-walled, of 2 - 3 articles, of two types; pointed end (m) and broad base, and rounded end (n), two kinds of uniseriate glandular trichomes: with single-celled base and one-celled secretory cell (r),(s), trichome with bi or multicellular base and rounded end (o), trichome with strangulation (q), simple, doubly or triply bifurcated glandular trichome (p).

# *Hyoscyamus muticus* L. *Subsp. falezlez* (Coss.) Maire flower and fruit powder analysis

The fruit and flower powder is buffcolored, fluffy to fibrous, and characterized by the presence of: papillose cells (Pap); isolated in clusters, giving the appearance of a flower,

epicarp fragments of fruits with thick, wavy-walled cells (EpiC), and albumen fragments with polygonal cells containing whitefly grains (Alb) as shown in **Fig. 5**. Bifurcate or glandular trichomes with unicellular head and multicellular stalk were observed, the same as those observed in leaves and stems. Sclereidal fibers were abundant, shorts, isolated or grouped in clusters, with wide punctuation lumen and canaliculated walls. Simple fibers were rare and isolated, long yellowish with a pointed end and a narrow lumen. Yellow pollen grains were identified with tri-aperturedexine, longitudinal rays from pores, (see (j) and (k) in **Fig. 5**). Contaminating pollens can be encountered.

Calcium oxalate crystals, orange secretions (isolated or contained in crystalline cells), parenchyma fragments (with starch grains) and spiral and pitted xylem vessels were noted.



Fig. 5: The different elements of the *Hyoscyamus muticus* flower and fruit powder, observed under an optical microscope at 10x40 magnification. Papilleous cells isolated in clusters (d), fragments of the epicarp of fruits with thick and wavy-walled cells (a), shards of albumen with polygonal cells containing whitefly grains (b), single-legged or pluricellular single-celled glandular trichome (i), sclereidal fibers (e), isolated simple fibers (f), tri-apertured pollen grains (j), Asteraceae pollen contaminants (k), (l), isolated orange secretions; contained in crystalline cells (h).

#### Results of multivariate statistical analysis

The Principal Component Analysis, extracts 83.32% of the information from the matrix, with a Cronbach alpha of 0.988 in both dimensions. The two axes (X) and (Y) explain respectively 27% and 25.9% of the variations.

The correlations table (see **Table 1**) shows the correlation coefficients between the

different elements after projection on the two dimensions.

The projection of the elements, allowed us to identify the microscopic items with close correlation coefficients and to eliminate the items not correlated with the rest of them; which are represented by (Multicellular eglandular trichome with a pointed end (PoiTEP), starch (Ami), crystalline cells (Crista), and sclereidal fibers (FS)), as shown in Fig. 6.

The projection of the observation items on the two dimensions, and the correlation coefficient allowed us to distinguish two main groups of observations, according to the presence or absence of Albumen (Alb), and two subgroups depending on the presence or absence of the cluster prism crystal of calcium oxalate (PrisM) as shown in **Fig. 7**.

Albumen element is positively correlated with the first factor (0.051, 0.288) and negatively correlated with the second factor (-0.1, -0.542). The cluster prism crystal of calcium oxalate element is positively correlated with both factors and highly correlated with the second (0.096, 0.744) (see **Table 1**).

The group of observations that did not contain the element albumen, named (AB) is more important than the group whose element albumen is present (AB+). The group of observations that contains albumen, concerned the observations<sup>7,8,16,17,18,25,27</sup>. Two subgroups were identified; from the group (AB-) based on the presence or absence of cluster prism crystal of calcium oxalate element, so the sub-group whose this element was absent is named (PM-), and concerned observations<sup>4,5,9,13,15,24,26</sup> and the sub-group whose cluster prism crystal of calcium oxalate element was present; is named (PM+) and concerned observations <sup>1,2,3,10,11,12</sup>.

Variables	Factor1	Factor 2
Epi	0.120	0.038
EpiC	0.088	-0.139
Par	0.119	-0.099
Vai	0.108	-0.045
PoiSB	0.119	-0.056
PoiSUU	0.105	0.116
PoiSUP	0.108	0.059
Li	0.065	0.060
PoiTEA	0.078	-0.114
PoiTEP	-0.104	0.043
PrisC	0.078	0.071
PrisM	0.096	0.137
Pris	0.069	0.145
Pol	0.067	-0.132
FibS	-0.007	-0.095
FibL	0.064	0.096
Alb	0.051	-0.100
Рар	0.084	-0.133
Ami	-0.071	0.002
Crista	-0.085	-0.027
SecO	0.142	0.063

Table 1: Scores correlation coefficient of Hyoscyamus muticus elements powder.

**Epi:** Epidermis, EpiC: Epicarp, Par: Parenchyma, Vai: Xylem vessels, PoiSB: Bifurcate glandular trichome, PoiSUU: Unicellular glandular trichome, PoiSUP: Multicellular glandular trichome, Li: Phloem, PoiTEA: Multicellular eglandular trichome with a round end, PoiTEP: Multicellular eglandular trichome with a pointed end, PrisC: Twin crystals of calcium oxalate, PrisM: cluster prism crystal of calcium oxalate, Pris: Prism crystal of calcium oxalate, Pol: Pollen, FibS: Sclereidal fiber, FibL: Simple fiber, Alb: Albumen, Pap: Papilleous cells, Ami: starch grains, Crista: Crystalline cells, SecO: Orange secretions.



Fig. 6: Correlation diagram between different powder elements of *Hyoscyamus falezlez*. The diagram represents the dispersion of the different elements recorded in the microscopic observation after projection on the two axes (F1, F2). The abbreviations of elements powder were defined as follows: Epi: Epidermis, EpiC: Epicarp, Par: Parenchyma, Vai: Xylem vessels, PoiSB: Bifurcate glandular trichome, PoiSUU: Unicellular glandular trichome, PoiSUP: Multicellular glandular trichome, Li: Phloem, PoiTEA: Multicellular eglandular trichome with a round end, PoiTEP: Multicellular eglandular trichome with a pointed end, PrisC: Twin crystals of calcium oxalate, PrisM: cluster prism crystal of calcium oxalate, Poi: Pollen, FibS: Sclereidal fiber, FibL: Simple fiber, Alb: Albumen, Pap: Papilleous cells, Ami: starch grains, Crista: Crystalline cells, SecO: Orange secretions.



Fig. 7: Correlation diagram between powder observations of *Hyoscyamus falezlez*. Alb- (Albumen absent, blue cercle), Alb+ (Albumen present, red cercle), PM- (cluster prism absent, green cercle), PM+ (cluster prism present, yellow cercle), A1: Axe 1, Axe 2.

### Results of comparative analysis of elements from different harvest stations

Microscopic observation of Hvoscvanus muticus L. subsp falezlez (Coss) Maire powder elements between the different sampling stations showed no significant morphological type difference. However, statistically, the comparison of the detection numbers of microscopic elements showed a significant difference only for two elements: the Glandular trichome with unicellular head and unicellular stalk (PoiSUU) which was more frequent in the samples of Abadla station (p=0.034) and Multicellular eglandular trichome with a pointed end (PoiTEP) (p=0.012), which was frequent in both stations (Abadla and Adrar) with a percentage (66%, 44% respectively), and absent in the Tamanrasset staion.

## Results of comparative analysis of elements from different organs

Statistical analysis showed no significant differences between organs (leaves, stems, flowers and fruits) for the elements: Epidermis, Parenchyma, Xylem vessels, Bifurcate glandular trichome with unicellular head and multicellular stalk, Glandular trichome with unicellular head and unicellular stalk, Phloem, Prism crystal of calcium oxalate, Twin crystals of calcium oxalate, Simple fiber, Orange secretions.

Some elements were significantly abundant in the fruit case of Pollen, Albumen, Papilleous cells, Epicarp and Glandular trichome with unicellular head and unicellular stalk. Cluster prism crystal of calcium oxalate were present in the leaves predominantly (75%, p<0.05) and Crystalline cells abundantly in the stem (50%, p=0.01). Finally, Sclereidal fiber were observed only in the stem and fruit while starch grains were present only in the stem and leaf.

### Discussion

The botanical test is an indispensable step in the identification of plants in Pharmacognosy; it highlights the characteristics of the family while identifying the characteristic elements of the plant.

The study found characteristic elements of Solanaceae, especially the numerous glandular trichomes of different forms, amplitude of calcium oxalate crystals, and the presence of cellulosic fibers<sup>6</sup>. Eglandular trichomes were rare or absent in all organs compared to other Solanaceae species.

Generally, the genus *Hyoscyamus* is characterized mainly by the presence of calcium oxalate prisms, but in our study; the presence of cluster prism crystal of calcium oxalate was noted in the Saharan Henbane, which is consistent with previous studies<sup>21,25,26</sup>, because it is possible to observe them in aged plant. The palisade parenchyma observed is confused with the loose lacuna parenchyma of the limb, which is rather equifacial and differs from the black Henbane, however, it is similar to other Henbanes (*H. pusillus, H. reticulatus*, and *H. aureus*)<sup>26</sup>.

By observing the powder of the different organs, we also noticed the abundance of bifurcate glandular trichomes with unicellular head and multicellular stalk, as well as the twin crystals of calcium oxalate that constitute characteristic elements of Saharian species that were cited by other studies<sup>25,26,27</sup>. Notable starch grains, as well as orange secretions, have been found in parenchymal cells and were cited<sup>17</sup>, however, the starch is absent in black Henbane<sup>21</sup>, another point that differentiates it from the latter. The presence of multicellular eglandular trichomes with a round end has also been highlighted on different organs and stations and is not cited in the literature. The Multicellular eglandular trichome with a pointed end and sclereids fibers were also identified in several organs. These last ones are not mentioned in the literature, though the eglandular trichomes with a pointed end can be found in some Solanaceae, particularly the Datura species<sup>18</sup>.

Contaminating pollen grains attributed to Asteraceae, encountered in the powder of flowers and fruits of Henbane; with the tripod pollen characteristic (corresponds to the description of Boukhalfa's thesis, 2017)<sup>28</sup>. It may be justified by the presence of Sagebrush in the vicinity of the Henbane at harvest.

The multivariate analysis of the elements of the Saharian Henbane powder concerned twenty-seven observations, including the three organs; was never realized in Algeria, but many species like Turmeric and Echinops were studied earlier<sup>19,20</sup>. The Principal Component Analysis (ACP) allowed us to group the correlated elements and ignore the weakly correlated items within the group. It permitted the achievement of an identification key based on the statistical data and provided additional confirmation of microscopic observations. The less correlated items were: the sclereids fibers and multicellular eglandular trichomes with a pointed end. Are they contaminating elements or specific to the Henbane? Only a microscopic study based on higher observation and station number can confirm this.

The multivariate analysis also allowed us to divide the observations into two groups, according to the presence or absence of albumen. It is logical since the albumen is a characteristic element of the fruit, so if the sample observed was from the aerial parts, it might or might not contain fruit. Two subgroups were identified according to the presence or absence of the cluster prism crystal of calcium oxalate; this can be explained by the fact that the sample can come from a young or aged plant because cluster prism crystals of calcium oxalate are found mainly in aged plants<sup>7,14,17</sup>.

The comparative analysis of the powder elements between the three harvest stations showed significant differences only for two elements, this reflects the homogeneity of the samples.

The comparative analysis of the elements between the different organs revealed the predominance of some in the stems and fruits (Sclereidal fiber for example), this can be explained by the nature of the lignified and fibrous tissue of these two organs<sup>17</sup>. Other elements such as oxalate crystals (Prisms and Clusters) were abundant in the leaves and stem; *Hyoscyamus muticus* L. Subsp *falelez* (Coss) Maire is a plant with fleshy stems and leaves that accumulate water, due to the desert climate in which it grows. It is known that calcium oxalate crystals testify to the richness of a plant in water<sup>17</sup>, which explains their abundance in growth organs.

### Conclusion

It is important to note that the study showed some items rarely cited in the literature (sclereids fibers, cluster prism crystals of calcium oxalate). The Multicellular eglandular trichome with a round end element has never been quoted in the literature, which constitutes a contribution to the botany data plant. The comparative study between the harvesting stations did not reveal any significant differences between the observed elements but the comparison between the organs showed the frequency of certain elements compared to others.

Multivariate analysis of powder elements is a topical method for developing a microscopic identification key, and this has never been done before for this plant in Algeria.

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التحليل متعدد المتغيرات للعناصر المجهرية لـ HYOSCYAMUS MUTICUS L. SUBSP. فالزليز (كوس.) مير، مسحوق من الجزائر

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فالزليز هي نبتة سامة، جد منتشرة في الصحراء الجزائرية. وهي واحدة من أغنى الباذنجانيات في

لقلويات الاستوائية ذات أهمية حد منتشرة في الصحراء الجرائرية. وهي واحدة من أعدى الباديجانيات في القلويات الاستوائية ذات أهمية صيدلانية وقتصادية.

يلعب الاختبار المجهري دورًا رائدًا في تحديد الأنواع وارتبط مــؤخرًا بــالتحليلات الإحصــائية، يهدف العمل إلى دراسة الخصائص المجهرية لهذا النبات من ثلاث محطات في الصحراء الجزائريــة وتم استكماله بدراسة تحليل متعددة المتغيراتة.

كان الفحص المجهري يتعلق بجميع مسحوق الأعضاء في النبات وتم استكماله بدر اسة تحليل متعددة المتغير ات للعناصر . تم تقييم توزيع العناصر المجهرية باستخدام تحليل المكونات الرئيسية . تم إجراء الارتباطات بين العناصر المجهرية والملاحظات عن طريق تحليل الحد من الأبعاد.

كشفت نتائج مقال علم النبات عن درجة عالية من التباين الهيكلي، بالإضافة إلى العناصر الموجودة عادةً أثناء المراقبة المجهرية، نادرًا ما يتم الاستشهاد ببعض العناصر التي تم تحديدها ، مثل ألياف التصلب ومنشورات الكالسيوم التي كانت عديدة في عيناتنا. لم يتم ذكر بعض العناصر، مثل بلورة المنشور العنقودي لأكسالات الكالسيوم، والتريكوم الغدي متعدد الخلايا، والتريكوم أحادية الخلية الضخم مع طرف مستدير. سمحت لنا الدراسة متعددة المتغيرات بتصنيف مسحوق العناصر اعتمادًا على درجة الارتباط. كما يسرت عملية التعرف من خلال تطوير مفتاح من مجموعتين ومجموعتين فرعيتين من

في نهاية در استنا، ساهمت النتائج في إثراء بيانات علم النبات النباتي.