

SEED MORPHOLOGY OF THE GENUS *HYOSCYAMUS* L. IN TURKEY AND ITS SYSTEMATIC SIGNIFICANCE

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ABSTRACT

The seed morphology of six species, representing genera of *Hyoscyamus* L. (Solanaceae) in Turkey was investigated using a binocular stereomicroscope and scanning electron microscopy (SEM), to determine the significance of seed coat features as taxonomic characters. Macro and micromorphological characters, including seed shape, size, hilum position and character, seed coat appearance, cell shape, sculpture characteristics of cell walls and wall ornamentation are presented. The results indicate that the seed coat sculpture is cerebelloid in *H. pussillus* and reticulate or reticulate–puzzle in all the remaining species. Two types of testal cells can be identified based on their general shape and linearity of the anticlinal walls. In one type, the cell shape is oblong to subpolygonal, with the anticlinal wall being sinuate to repandate; in the other type, the cell is triangular to subpolygonal, with the anticlinal wall straight or slightly curved. Fine ornamentation of the anticlinal walls of the testal cells varies from smooth to covered with, fibril, punctate or papillate projections. However, the size and shape of seeds and the position of hilum have partly systematic significance.

Keywords: *Hyoscyamus*, Morphology, Seed, Solanaceae, SEM

INTRODUCTION

Hyoscyamus L., belongs to the family Solanaceae. Almost 26 species of *Hyoscyamus* are believed to occur in all the world [1]. The genus *Hyoscyamus* is represented by six species in the Flora of Turkey [2-3]. *Hyoscyamus* species have medicinal importance because of their hyoscyamin and scopolamine content [4-6]. Therefore, it is widely used for their mydriatic, antispasmodic, anti-cholinergic, analgesic and sedative properties in folk medicine. [7]. In the east of Turkey, *Hyoscyamus niger*, traditionally named as ‘Henban’, is used for the treatment of asthma, diarrhea, abdominal pain and urinary incontinence [8].

Seed morphology has been shown to provide useful characters for the analysis of taxonomic relationships in a wide variety of plant families. Especially, morphology of seeds, the details of the sculpturing of the outer seed coat can be quite variable and of systematic importance [9-13].

Recent studies on Solanaceae have shown that seed morphology characters are of considerable systematic significance, both at the generic and specific levels. Characters of the seed coat can be used to distinguish the genera *Physalis* L., *Capsicum* L. and *Tubocapsicum* (Wettst.) Makino [14-15]. Several previous studies have examined seed morphology of various members of tribe Hyoscyameae [16-18].

The specific objective of this study was to assess the usefulness of the macro- and micromorphological characters of the seeds using stereomicroscope and scanning electron microscopy (SEM) between the species of *Hyoscyamus* under this investigation. The aim was also to obtain comparative information on the seeds, to improve the present knowledge of the species and to evaluate the usefulness of this feature for systematic purposes.

MATERIALS AND METHODS

Plant materials were collected from different localities in Turkey (Table 1). Voucher specimens were deposited in the herbarium of the Department of Biology in Harran University.

The seeds samples were taken for investigation and 15-20 dried seeds samples for each species were examined. Measurements and optical observations of seeds were carried out under a binocular stereomicroscope Wild M5. Axis parameters were measured with the aid of an ocular micrometer (Table

2). For scanning electron microscopy (SEM), mature nutlets were soaked in a 1:1 solution of chloroform and methanol for 48 h and dehydrated through an ethanol series (70, 90 and 100%) to detect. After air-drying, the seeds were mounted directly on stubs, using conductive double-side adhesive tape and coated with gold for 5 min. Coated seeds were examined and photographs were taken with EVO-50, at 15 KV and 15 mm working distance. The terms used for describing the seed surface patterns have been adopted according to Stearn [19] and Zhang *et. al.* [20].

Table 1. Collection data of studied *Hyoscyamus* L. species

| TAXA | COLLECTION DATA | COLLECTOR NUMBER |
|-----------------------------|---------------------------------------------------------------------------------------------------------------|------------------|
| <i>H. pusillus</i> L. | B7 Tunceli: 2 km north of Pertek port, crevices and fissures on rock faces.13.06.2004, 1200 m. | Aslan 1529 |
| | B8 Elazığ: Harput Castle, Stony or rocky places, waste places, roadsides, 15.05.2004, 1400 m. | Aslan 1539 |
| | C5 Niğde: 2 km north of Niğde, roadside verge, dumping ground, 25.05.2003, 1000 m. | Aslan 1451 |
| <i>H. niger</i> L. | B8 Batman: in downtown of Hasankeyf, on the castle wall, 19.05.2004, 800 m. | Aslan 1448 |
| | C6 Kahramanmaraş: 10 km south of Kahramanmaraş, the margins of arable fields, 14.05.2004, 550 m. | Aslan 1534 |
| | C8 Mardin: Waste places, roadsides, 19.05.2004, 1350 m. | Aslan 1536 |
| <i>H. reticulatus</i> L. | B8 Diyarbakır: Ergani, Şölen village, Steppe, Stony or rocky places, waste places, 14.05.2004, 1100 m. | Aslan 1521 |
| | C5 Niğde: North of Ulukışla, along forest margins, open habitats, 20.06.2003, 1100 m. | Aslan 1399 |
| | C7 Şanlıurfa: Küçüksergen village, stony steppe, 19.06.2003, 550 m. | Aslan 1411 |
| <i>H. albus</i> L. | C6 Gaziantep: Antep castle, rock crevices on the wall, 31.07.2003, 650 m. | Aslan 1352 |
| | C7 Şanlıurfa: Urfa Castle, Stony or rocky places, Foot of rocks and walls, 11.04.2004, 600 m. | Aslan 1501 |
| | C8 Mardin: Mardin castle, in fissures on limestone, 13.05.2004, 1300 m., | Aslan 1528 |
| <i>H. aureus</i> L. | C6 Gaziantep: Nizip, Rum castle, crevices and fissures on rock, 04.04.2003, 550 m. | Aslan 1254 |
| | C7 Şanlıurfa: Birecik castle, crevices and fissures on rock, 24.05.2003, 650 m. | Aslan 1356 |
| | C8 Mardin: Castle, Rock crevices, old walls, 14.05.2004, 1250 m. | Aslan 1525 |
| <i>H. leptocalyx</i> Stapf. | C 8 Mardin: Castle, Rock crevices, 13.05.2004, 1200 m | Aslan 1430 |
| | C8 Mardin: Mardin castle, 29.09.2003, 1250 m. | Aslan 1407 |

RESULTS

In species of the genus *Hyoscyamus*, seed shape is quite variable, ranging from oblong, subovoid-oblong,

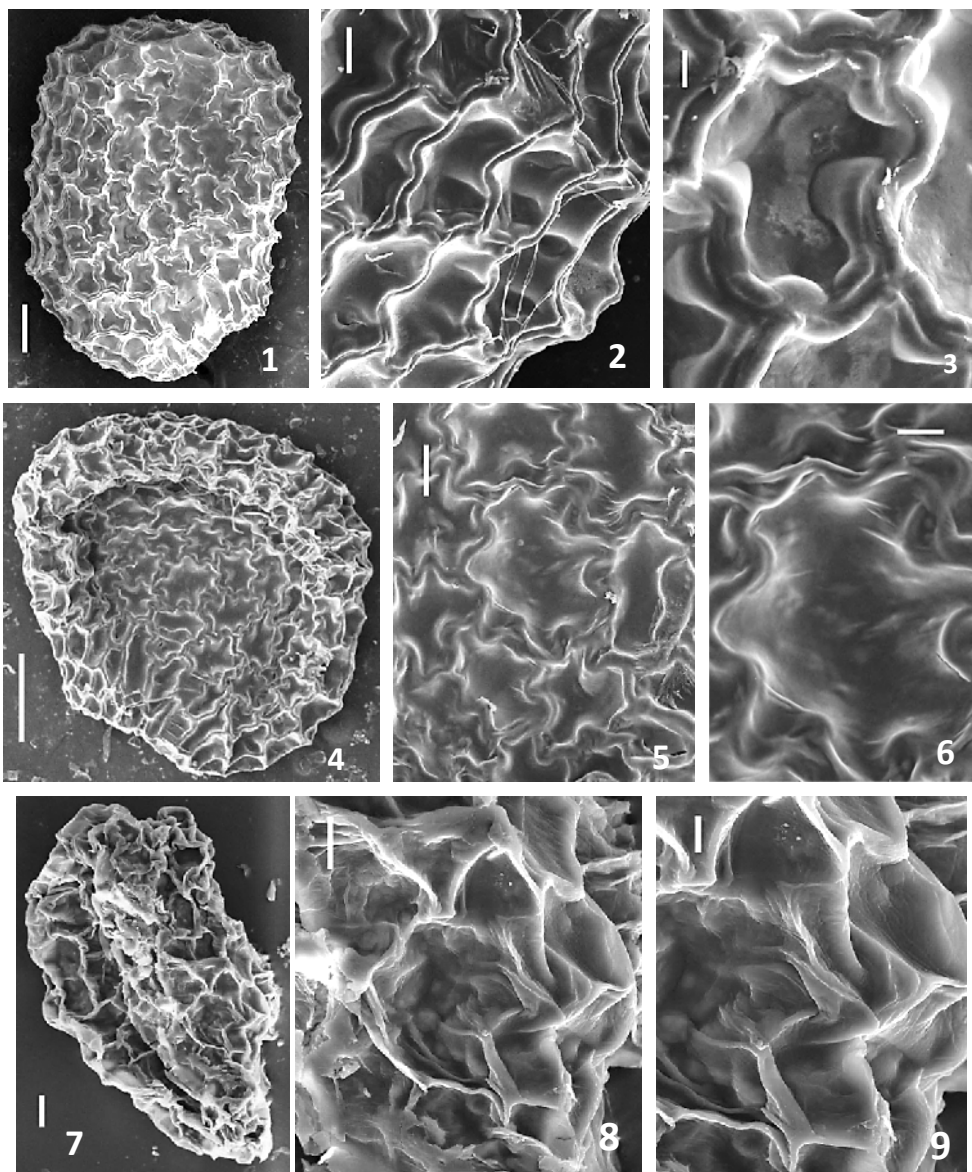
oblong to elliptic, broadly ovoid to suborbicular, triangular to reniform and compressed, subcompressed or not. The range of seed size in all species examined

are between 0.15 and 1.5 mm in length and 0.07-1.2 mm in wide (Table 2). The seed of *H. leptocalyx* was the smallest in the *Hyoscyamus* species.

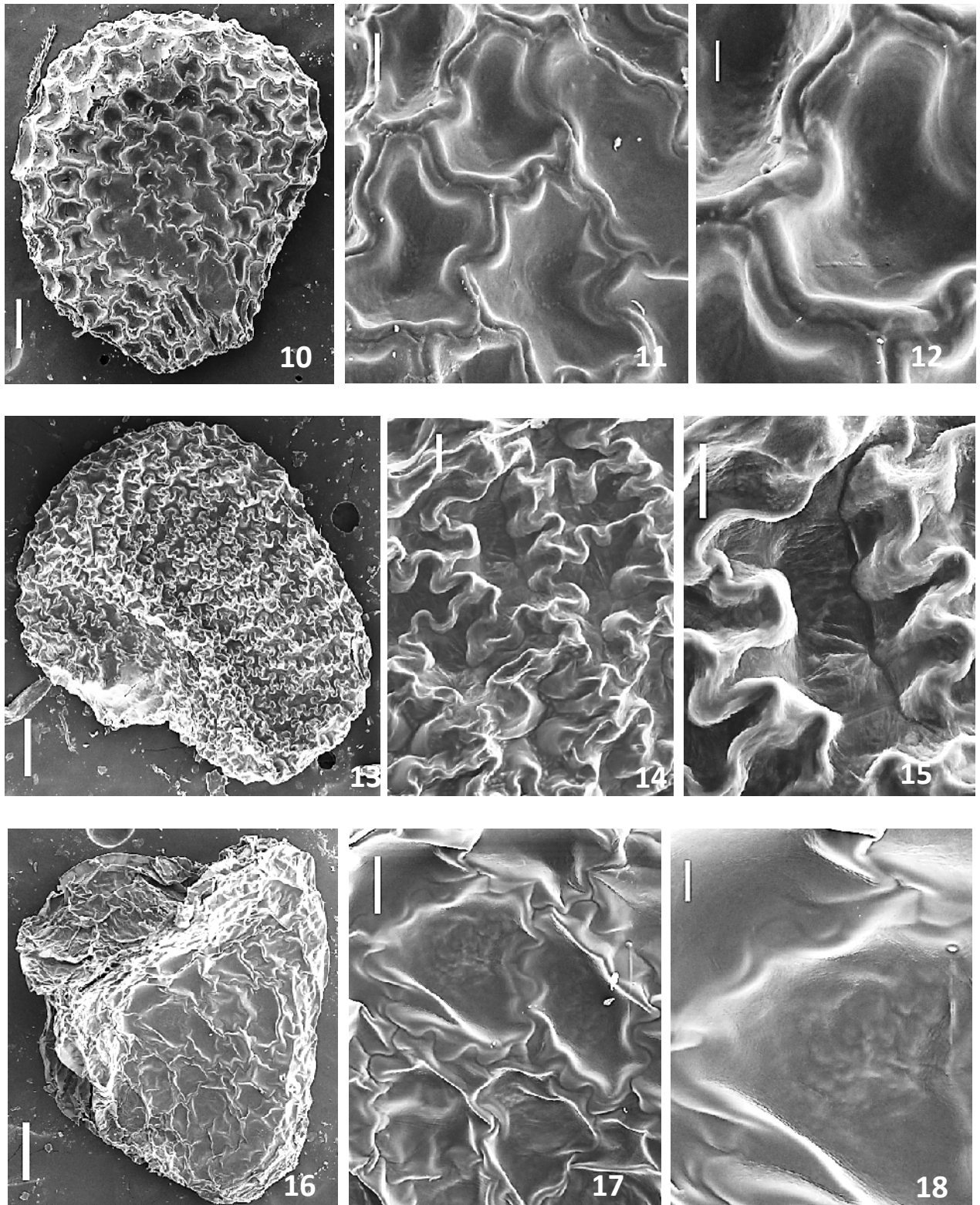
There were four major types of seed coat sculpture; reticulate, irregular-reticulate, reticulate-puzzle and cerebelloid which, only appeared in the seeds of *H. pusillus*. The shape of testal cells was irregular, oblong, triangular, triangular-ovoid to subpolygonal and deeply concave or not. The sculpture characteristics of cell walls were thin walls at the base of *H. reticulatus* or strongly or relatively thickened in the rest of the species. Additionally, the walls were sinuate-rependate

in *H. albus*, *H. aureus* and *H. niger*, as they were straight or slightly curved in *H. leptocalyx* and *H. reticulatus* and strongly sinuate or channelled in *H. pusillus*. The ornamentation of the anticlinal walls varied from papillate, fibril to occasionally punctate (Figs. 1-18).

The hilum position was basal or medial in *H. leptocalyx* and *H. pusillus* however, it was only basal in the remaining species. And, the basal hilum was generally small or large protruding in *Hyoscyamus* species or sometimes not protruding as *H. reticulatus*.



Figs. 1-9: *Hyoscyamus* seeds and their coat surfaces in SEM. 1-3 *H. albus*, 4-6 *H. aureus*, 7-9 *H. Leptocalyx*. Scale bars: 1, 4 = 200 μ m; 2, 5 = 40 μ m; 3, 6, 7 = 20 μ m; 8 = 10 μ m; 9 = 5 μ m.



Figs. 10-18: *Hyocyamus* seeds and their coat surfaces in SEM. 10–12 *H. niger*, 13–15 *H. pusillus*, 16–18 *H. reticulatus*.
 Scale bars: 10, 13, 16 =200 µm; 17 = 60 µm; 11 =40 µm; 14, 15 =30 µm; 12,18 =20 µm.

Table 2. Macro-micromorphological characters of the studied *Hyoscyamus* seeds

| Taxa | Seed shape and size | Seed coat appearance | Cell shape | Sculpture characteristics cell walls | Wall ornamentation | Hilum position and character |
|-----------------------------------------------|----------------------------------------------------------------------------------|----------------------|-----------------------------------------------------------------------------------|--------------------------------------------------------------------------------------|-----------------------------------|----------------------------------------|
| <i>H. albus</i> (Figs. 1-3) | oblong, subovoid-oblong, sometimes sub-compressed reniform 1,1-1,4 x 0,8-1 mm | Reticulate | relatively large longer than wide irregularly oblong subpolygonal deeply concave | base strongly thickened; walls sinuate sometimes repandate | papillate, occasionally punctate | basal small slightly protruding |
| <i>H. aureus</i> (Figs. 4-6) | broadly ovoid to suborbicular, compressed in the middle 0,7-1 x 0,5-0,8 mm | Reticulate-puzzle | relatively large irregular subpolygonal | base relatively thick, thinner in middle parts; walls sinuate-repandate | papillate | basal large slightly protruding |
| <i>H. leptocalyx</i> (Figs. 7-9) | oblong to subelliptic, compressed 0,15-0,25 x 0,07-0,13 mm | Irregular-reticulate | relatively small irregular triangular to subpolygonal | base thickened walls straight or slightly curved | obviously papillate | basal or medial small protruding |
| <i>H. niger</i> (Figs. 10-12) | broadly ovoid to suborbicular- reniform compressed 1,1-1,5 x 0,9-1,2 mm | Reticulate-puzzle | relatively large longer than wide subpolygonal | base relatively thick; walls sinuate- repandate | fine papillate | basal large slightly protruding |
| <i>H. pusillus</i> (Figs. 13-15) | subovoid to suborbicular reniform compressed 1,1-1,3 x 1-1,2 mm | Cerebelloid | relatively small longer than wide irregular oblong to subpolygonal deeply concave | base strongly thickened; walls strongly sinuate; channelled | fibril present and fine papillate | basal or medial large protruding |
| <i>H. reticulatus</i> (Figs. 16-18) | triangular to suborbicular compressed 0,4-1,2 x 0,4-1,2 mm | Irregular-reticulate | relatively large irregular triangular ovoid to subpolygonal | base shallow and thin walls almost straight, or slightly curved or sometimes sinuate | papillate | basal small or large protruding or not |

DISCUSSION

The seed morphology of six *Hyoscyamus* species, growing in Turkey, is investigated in this study. The characters of seed shape, size, seed coat appearance and shape of testal cells are highly variable within species. The seed coat sculpture is cerebelloid in *H. pusillus* and reticulate or reticulate-puzzle in all the remaining species. Two types of testal cells can be identified based on their general shape and linearity of the anticlinal walls. In one type, the cell shape is oblong to subpolygonal, with the anticlinal wall being sinuate to repandate; in the other type, the cell is triangular to subpolygonal, with the anticlinal wall straight or slightly curved. The seeds of *H. leptocalyx* (average size 0.15–0.25 × 0.07–0.13 mm) are obviously smaller than those of the other species (average size 0.4–1.5 × 0.4–1.2 mm). In addition, the seeds of *H. leptocalyx* are oblong and subelliptic shaped so they can be easily separated than the other *Hyoscyamus* species. *H. albus* has oblong or subovoid-oblong seeds and the other seeds shape are variable, ranging from triangular, broadly ovoid to suborbicular. Axelius [17] was published testa patterns in some species of *Physalis* and some other genera in the tribe Solaneae (Solanaceae). According to results of this study, the testa cells are angular, irregular and sometimes elongated; the anticlinal wall thickenings are thin to moderately thick and the anticlinal walls are more or less undulate. The anticlinal walls in seeds of *Physalis peruviana* and *Exodeconus miersii* are deeply undulated and seed coat sculpture cerebelloid as our *H. pusillus* samples. However, the anticlinal walls in seeds of *Jaborosa squarrosa* and *Oryctes nevadensis* are undulated and seed coat sculpture reticulate-puzzle as our *H. aureus* and *H. niger* samples.

Zhang et al. [20] investigated seed morphology of 22 species, representing seven genera of the tribe Hyoscyameae. In their study, seed morphology of our five *Hyoscyamus* species, with the exception of *H. leptocalyx* were examined using scanning electron microscopy (SEM) and they reported the seed coat sculpture as irregularly striate-reticulate, cerebelloid

and reticulate. According to findings of Zhang et al., seed shape in the genus *Hyoscyamus* was ranged from subovoid-oblong to triangular to irregularly rectangular and *Hyoscyamus* species have rather small seeds (0.7–1.6 × 0.9–1.2 mm) from the other six genera of the tribe Hyoscyameae. Some *Hyoscyamus* species have smooth lateral cell walls. Other species of *Hyoscyamus*, for example *H. albus*, are densely papillate over the entire inner surface of the cell. According to our findings, the cerebelloid type in seed coat of *H. pusillus* was observed however, this type was only recorded in the genus *Anisodus* of the tribe Hyoscyameae [20]. Wall ornamentation of our *Hyoscyamus* species is usually papillate. However, punctate and fibril types are also observed in *H. albus* and *H. pusillus*. Our findings are usually similar to Zhang et al. [20] findings. But wall ornamentation of *H. reticulatus* is reported as smooth type.

Gunn and Gaffney [21] were published seed characteristics of 42 economically important species of Solanaceae. In this study, the seed morphology of *H. niger* were also described as size, shape, seed coat pattern and hilum position and their results are similar to our results.

Archihyoscyamus is a monotypic genus and was segregated from *Hyoscyamus*. According to Lu, the seed coat morphology of this genus is similar to *Hyoscyamus*, but the ornamentation of the cell walls is smooth and the base of the lumen is densely papillate [22].

The seed morphology in *Hyoscyamus* species are extremely variable and perhaps quite ancient, group differentiating in dry habitats and Mediterranean climate [20]. These characters of seed morphology of the genus *Hyoscyamus* may be of value in the future taxonomic and phylogenetic study of the closely groups.

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