

THE STRUCTURE OF THE FLOWER OF *HYOSCYAMUS MUTICUS* LINN.

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INTRODUCTION

THE earliest written diagnosis of the plant *Hyoscyamus muticus* is that given by Linnaeus¹ in the "Mantissa Plantarum," 1767, and it is to this book that reference is made in the Index Kewensis as the authority for the name. This description was transferred to the fourth edition of the "Species Plantarum." Other early descriptions are given by Forskal², 1775, Lamarck³, 1789, Dunal⁴, 1852, Boissier⁵, 1875 and Hooker⁶, 1885. Mansfield⁷, 1908, describes certain histological characters of *H. muticus* in comparison with those of *H. niger* and *Atropa belladonna*; his work is illustrated by very inadequate drawings and he gives a list of diagnostic characters of *H. muticus*, the only satisfactory ones being the "compound multicellular branched hairs" and the "yellow sinuous-walled testa cells." Sterling⁸ (1908) also gives a partial description of the histology of *H. muticus* which includes some information about the calyx, fruit and seeds illustrated by a few drawings; he directs attention to the characteristic trichomes as distinguishing *H. muticus* from *H. niger*. His descriptions and drawings of the epidermis, of the calyx and of the crystals are unsatisfactory and do not clearly differentiate between the two species. Further work on the drug has been done by Fahmy (1931-2), who gives a more detailed account of the gross morphology of the flower with a rather sketchy description of its anatomy. Fluck, Schlumpf and Siegfried¹⁰, in 1935, deal briefly, with the macroscopical characters of both the flower and the fruit. The drug appears in the Pharmacopœa Helvetica, V Edition, 1933, which gives a few details of the gross morphology of the flower. The first official British monograph appears in the British Pharmaceutical Codex, 1949, where the flower is very briefly dealt with. The Indian Pharmacopœial List, 1946, gives a monograph but the description does not check with that of *H. muticus* Linn.

In view of the scanty information existing about the histology of the flower, it was necessary to make a detailed study which would provide data for the correct identification of the flower of *H. muticus* and would serve as a basis for comparison with the flowers of other closely related species of plants used as drugs. The following description is comparative with that of the flower of *H. niger*, of which a description is given by Rohatgi and Fairbairn⁹. Where no comment is made, it is to be inferred that the structure of the flowers of the two species is similar.

When recording measurements, the letters L, T and R are used to indicate measurements in a longitudinal, tangential and radial direction respectively, the directions having reference to the axis of the relevant plant member.

MATERIAL

Four different specimens were examined for the purpose of this work. The following is a list of the samples used, the first two being representative of the commercial drug available in this country :

1. Flowers from the collection of the School of Pharmacy, University of London.
2. Flowers from the Museum of the Pharmaceutical Society of Great Britain, presented by S. K. Crews, September, 1942.
3. Flowers sent from Cairo by Dr. Fahmy, July, 1949.
4. Fresh and dried flowers collected from the garden of the Institute of Pharmacy, Kasr-el-Aini, Cairo, by courtesy of Dr. A. H. Sabir, September 1949.

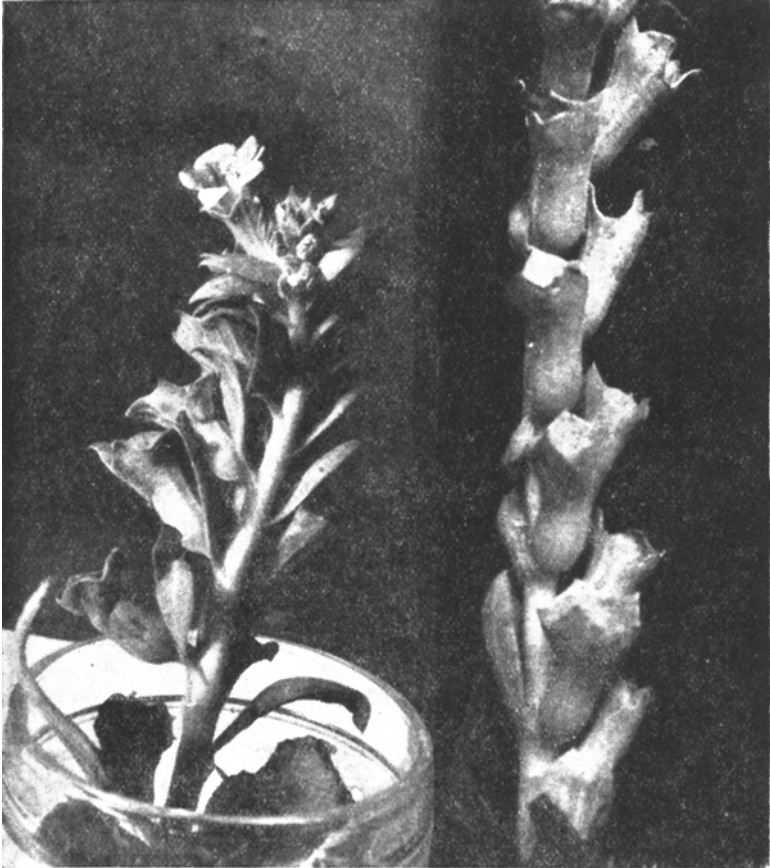


FIG. 1. *Hyoscyamus muticus* Linn. On the left a photograph of a flowering top, showing the curled unilateral inflorescence, the axis being bare on one side, and on the other side bearing flowers arranged in two vertical rows, the oldest flowers being at the base. On the right some fruits showing the calyx which is sub-cylindrical at the base and devoid of spines on the lobes. $\frac{2}{3}$ natural size.

THE FLOWER OF *HYOSCYAMUS MUTICUS* LINN.

GROSS MORPHOLOGY

The inflorescence closely resembles that of *H. niger*, which has been variously described as a unilateral raceme and as a cincinnus or a scorpioid cyme, the flower is hermaphrodite and slightly irregular (Figs. 1 and 2).

Calyx : (Fig. 2). The calyx of *H. muticus* differs slightly from that of *H. niger* in being slightly longer (12 to 36 mm. long), campanulate, ribbed and fluted; the lobes are spreading and short, being about 1/7th to 1/5th of the length of the calyx (in *H. niger* they are about 1/3rd of the length if the calyx); they are broad and acute, but do not run out into a spine, hence the specific epithet *muticus* meaning "pointless, blunt, awnless."

Corolla : (Figs. 1 and 2). The main noticeable difference lies in the colour of the petals; one anterior and two lateral petals are deep purple

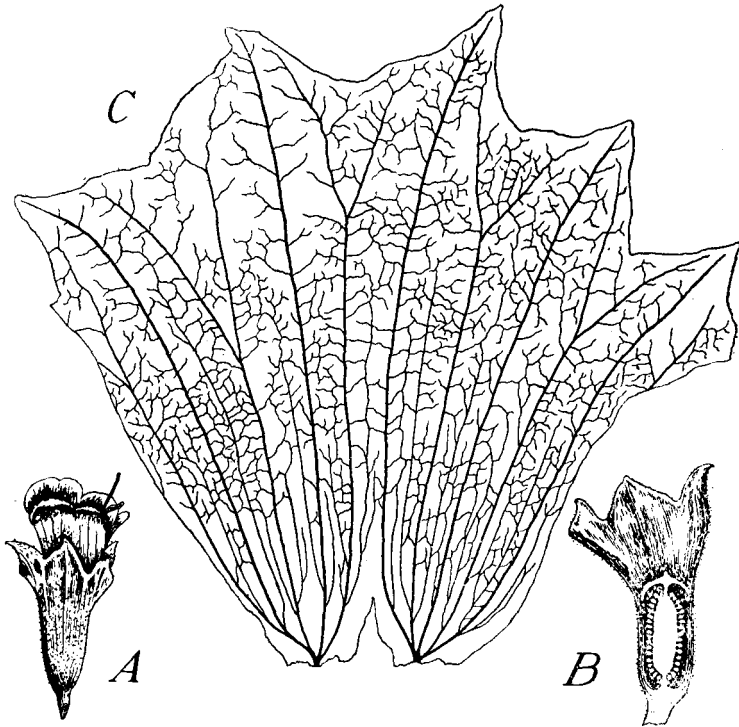


FIG. 2. *H. muticus* Linn. A, Flower $\times 1$. B, an immature fruit cut longitudinally $\times 1$. C, Calyx, spread out, showing the venation and the five rather blunt lobes $\times 3$.

inside and greenish-white outside; the remaining two small petals are greenish-white inside and out.

Androecium : (Fig. 2, A). The free part of the *filament* measures about 7 to 14 mm. in length, the adherent part being about 2 to 5 mm. long; the filaments are white in the lower part and purple at the tips; for the

rest, the stamens are similar to those of *H. niger*. The *anthers*, however, are straight and not curved as in *H. niger*.

Gynæcium: (Fig. 6, A). The gynæcium differs in having a longer style (about 2 to 3 cm. long); the stigma is sometimes slightly capitate but much less so than that of *H. niger*, often however, it has the same diameter as that of the style; the *ovary* measures about 1 to 4 mm. in length and 1 to 3 mm. in diameter at the widest part.

Fruit: (Figs. 1 and 2, B). The pyxis is more elongated and cylindrical than that of *H. niger*. The persistent calyx tapers slightly towards the base; the lobes of the calyx are weaker, have no spine at the tips or cusps at the sinuses and when dry, have a tendency to crumple and break. The seeds are lighter in colour than those of *H. niger*, being greenish-brown to yellowish-brown.

HISTOLOGY OF THE CALYX

Outer (abaxial) epidermis: The cells in the different regions of the sepals do not show variations in shape similar to those found in *H. niger*. All the cells are tabular, subrectangular to polygonal, with straight or slightly curved anticlinal walls (Fig. 3, Ao and Co), the dimensions being approximately L and $T = 12$ to **60** to 150μ and $R = 24$ to **54** to 105μ . The cuticle is markedly striate, as also is the cuticle of the trichomes, which has transverse circumferential striations. Cruciferous (anisocytic) *stomata* occur frequently on all parts of the calyx and are usually slightly depressed below the general epidermal surface (Fig. 3 F), whereas in *H. niger*, they are raised. The epidermis bears numerous glandular *trichomes* which are more abundant towards the base of the calyx, where some much enlarged trichomes are present in addition to the smaller ones. Certain cells of both the epidermises contain one or more *crystals* of various shapes, double pyramids or prisms being more common.

Inner (adaxial) epidermis: The anticlinal walls show a gradual transition from slightly wavy on the lobes to straight at the base of the calyx, and this change is accompanied by a gradual increase in the size of the cells (Fig. 3, Ai, Bi and Ci). The dimensions of the cells are approximately; on the lobes, L and $T = 15$ to **45 to 90** to 135μ and $R = 24$ to **45** to 60μ ; at the middle of the sepal, L and $T = 30$ to **90 to 120** to 300μ and $R = 24$ to **60** to 75μ and at the base, L and $T = 30$ to **90 to 150** to 390μ and $R = 15$ to **45** to 75μ . The cuticle, including that of the trichomes is markedly striate. Cruciferous *stomata*, which are slightly recessed, occur quite frequently, but are rare at the base. Long glandular *trichomes*, similar to those on the outer epidermis, are quite frequent on the lobes but less so at the base where very short trichomes occur. A small number of *crystals* occurs either as double pyramids, rectangular prisms or small groups or clusters of incomplete prisms.

Trichomes: (Fig. 3). All the trichomes of the calyx are glandular, each having a 1 to 6-celled uniseriate stalk and a small unicellular globular head with yellowish-brown granular contents which stain yellowish-orange with Sudan III reagent; small crystals of calcium oxalate (about

THE FLOWER OF *HYOSCYAMUS MUTICUS* LINN.

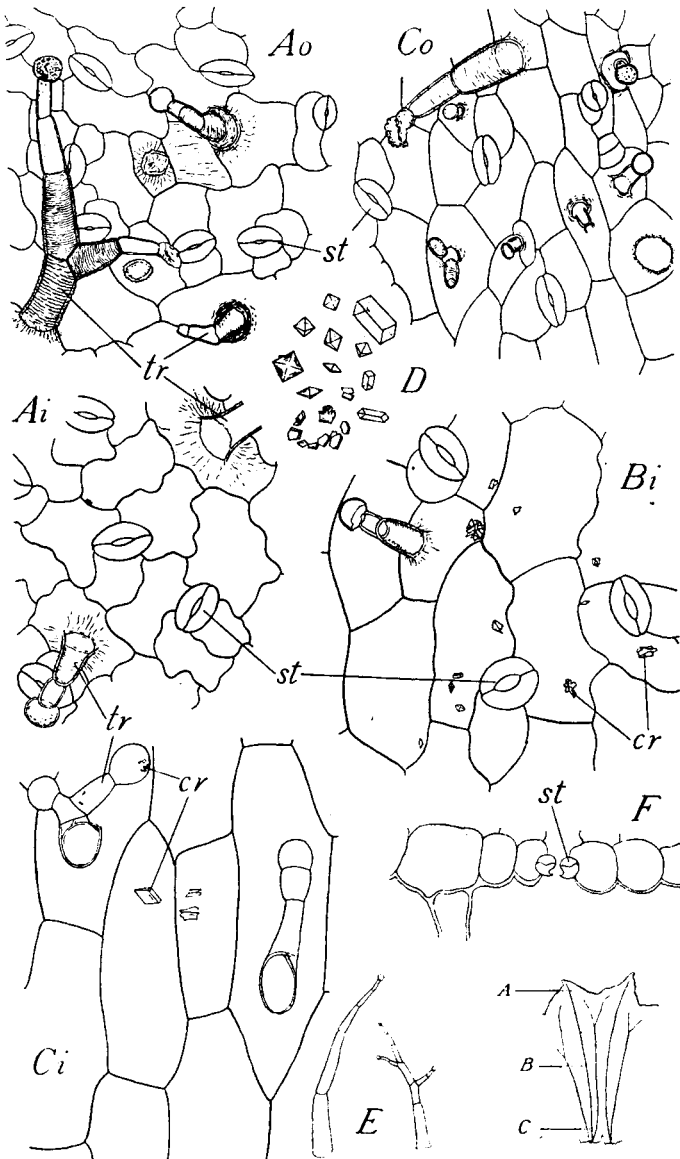


FIG. 3. *H. muticus* Linn. Calyx. A, outer epidermis at A (as marked on the diagram at bottom right-hand corner), the cuticular striations are shown on the trichomes and a few only of the cells; A i, inner epidermis at A; B i, inner epidermis at B; Co, outer epidermis at C; Ci, inner epidermis at C; D, crystals from the mesophyll of the calyx; E, large simple and branched trichomes from the base of the outer epidermis of the calyx, $\times 30$; F, transverse section through the outer epidermis showing details of a stoma. cr, crystals of calcium oxalate; st, stoma; tr, trichome. All $\times 160$, except E.

3 to 12 μ) occur in the trichomes. Many of the trichomes are branched either once or twice (Fig. 3, Ao,Ci,tr and E). They are covered with a fairly thick cuticle, the striations of which appear on the stalk, especially on the cells towards the base, as transverse circumferential ridges and on the epidermal cell, from which each arises, as lines radiating from the base of the limb. The trichomes measure approximately, 45 to 105 to 980 μ in length, 30 to 45 to 90 μ in diameter at the base, the diameter of the head being 21 to 30 μ .

The *mesophyll* consists of 5 to 10 layers of thin-walled rounded parenchyma irregularly arranged; palisade cells are present in the lobes where they are rather short and occur under the adaxial epidermis only. At the mouth of the tube, a short celled palisade is present under the adaxial epidermis and a less definite and interrupted palisade is present under the abaxial epidermis. In the lower two-thirds of the tube, palisade cells are absent. *Crystals* of calcium oxalate are abundant in the cells of the mesophyll in all parts of the calyx, being present in greater numbers towards the base; the crystals are usually double pyramids (about 18 μ), right-angled prisms (about 12 to 16 μ) or angular crystals of indeterminate form, either single or in groups of a few components; also in many cells adjacent to the veins there are numerous scattered small crystals of different forms (about 3 to 6 μ), but idioblasts filled with sandy crystals such as occur in the calyx of *H. niger*, are absent.

Venation : (Fig. 2, C). The chief differences from *H. niger* in venation are: (1) the absence of a strong and pointed apical spine to each lobe, (2) the absence of marginal veins in the lobes and (3) the prominent lateral vein between the midribs bifurcates before reaching the sinus, whereas in *H. niger*, the corresponding vein travels right up to the sinus where it meets the marginal vein.

A transverse section of a midrib is similar to that of *H. niger* except that there is a more pronounced development of perimedullary phloem groups and the bundle of xylem fibres diminishes towards the tip in the lobes, whereas in *H. niger* it undergoes enlargement.

HISTOLOGY OF THE COROLLA

Outer (abaxial) epidermis : The cells on the lobes are tabular, sub-rectangular to polygonal with wavy anticlinal walls, but do not possess infoldings as in *H. niger* (Fig. 4, Ao); they measure approximately L and T=15 to 27 to 45 to 75 μ and R=24 to 36 to 70 μ . The anticlinal walls of the epidermal cells become progressively less wavy towards the base (Fig. 4, Bo, Co and Do), where they are nearly straight or slightly curved and resemble the corresponding cells of *H. niger*. The cells in the middle part of the tube are elongated and polygonal, measuring approximately L=60 to 90 to 210 μ , T=15 to 30 to 45 μ and R=30 to 36 to 60 μ . At the base of the tube, the cells have thick, refractive and lignified anticlinal walls and are almost rectangular in surface view; the dimensions are approximately L=15 to 45 to 78 μ , T=9 to 21 to 30 μ and R=30 to 36 to 54 μ . The cells of the tube, in dried specimens, have faint striations on the cuticle.

THE FLOWER OF *HYOSCYAMUS MUTICUS* LINN.

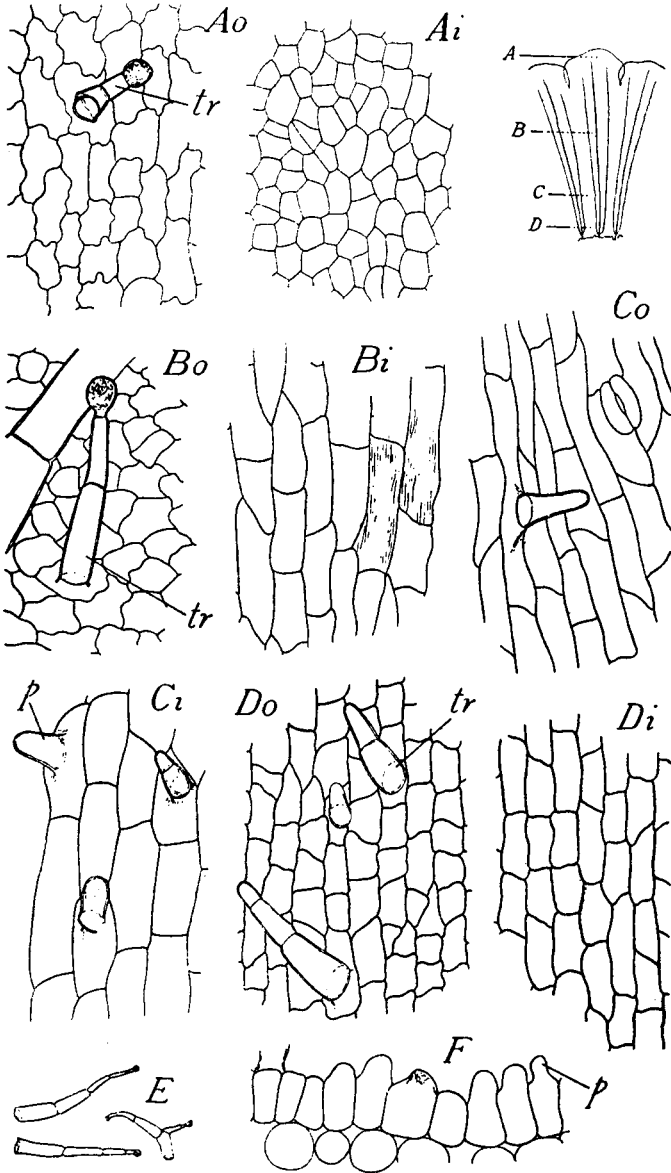


FIG. 4. *H. muticus* Linn. Corolla. Ao, outer epidermis at A (as marked on the diagram at top right-hand corner); Ai, inner epidermis at A; Bo, outer epidermis at B; Bi, inner epidermis at B, cuticular striations shown on two cells only. Co, outer epidermis at C; Ci, inner epidermis at C; Do, outer epidermis at D; Di, inner epidermis at D; E, trichomes from the outer epidermis of the lobes, $\times 30$; F, transverse section of the inner epidermis at the base of the tube. *p*, papilla; *tr*, trichome. All $\times 160$, except E.

The lobes bear abundant glandular *trichomes* which vary greatly in size and are occasionally branched. On the epidermis of the tube, the trichomes are smaller and less frequent; covering trichomes with a blunt and rounded apical cell are common on the lower part of the tube. Occasional epidermal cells of the tube possess papillae (Fig. 4, Co and Do).

Stomata are very rare on the outer epidermis of the lobes but are more frequent on the middle part of the tube.

Inner (adaxial) epidermis: The cells on the lobes are smaller in size than those of the outer epidermis and are sub-rectangular to polygonal with almost straight anticlinal walls which do not have infoldings as in the case of *H. niger* (Fig. 4, Ai). The cells measure approximately L and T=12 to 21 to 33 to 66 μ and R=15 to 18 to 27 μ . In the upper part of the tube, the epidermal cells are sub-rectangular and elongated, and the cuticle has well-marked striations (Fig. 4, Bi); the cells measure approximately L=60 to 105 to 195 μ , T=21 to 36 to 60 μ and R=24 to 30 to 45 μ . The anticlinal walls of the epidermal cells in the thin tubular part of the tube are lignified. *Trichomes* are rare on the lobes and on the upper part of the tube. 1 to 3-celled, short, tapering and bluntly-pointed covering trichomes occur on the lower half of the tube except at the very base (Fig. 4, Ci); the cells in this region also frequently bear papillae (Fig. 4, F) and faint cuticular striations are visible upon parts of some of the cells; the cell walls are not pitted as is the case of corresponding cells in *H. niger*. The cells are more regularly arranged at the base of the tube (Fig. 4, Di), where they are rectangular in shape, have thicker lignified walls and are covered with a finely striated cuticle; they measure approximately L=24 to 60 to 90 μ , T=15 to 24 to 30 μ and R=30 to 36 to 48 μ . *Stomata* are absent from the inner epidermis of the corolla.

Trichomes: Two types of trichomes are present on the corolla, glandular and covering. The glandular trichomes, present on the outer surface only, have a 1 to 7-celled uniseriate stalk and a sub-spherical, unicellular, glandular head which is filled with yellowish-brown granular contents; they are thinner walled than those on the calyx, and the cuticle is usually not striate, though in certain dried specimens, faint longitudinal striations were sometimes visible on the basal cells. The glandular trichomes measure approximately 70 to 280 to 630 μ in length, 18 to 54 to 75 μ in basal diameter, and the diameter of the head is about 15 to 30 μ ; occasional glandular trichomes are branched. The covering trichomes are slightly tapering, uniseriate, 1 to 3-celled and terminate in a small blunt cell which has a rounded or sub-spherical apex; the trichomes measure approximately 21 to 105 to 210 μ in length and 21 to 30 to 60 μ in diameter at the base. The papillae on the cells of the tube frequently resemble the very short covering trichomes.

The *mesophyll* consists of 4 to 11 layers of loosely arranged parenchymatous cells, the number of layers decreasing towards the apex with consequent thinning of the lamina in the lobes. Numerous *crystals* of

THE FLOWER OF *HYOSCYAMUS MUTICUS* LINN.

various forms, more commonly double pyramids or prisms, occur especially along the main veins. The cells adjacent to the main veins in the lower part of the tube contain minute starch grains.

Venation : The venation in *H. muticus* is similar to that in *H. niger*.

The midrib in transverse section shows spiral xylem elements surrounded by parenchyma with scattered groups of phloem.

HISTOLOGY OF THE STAMENS

Filament : The structure of the filament closely resembles that of *H. niger*. The chief points of difference are:

1. The longitudinally strongly striate cuticle (Fig. 5, B and C);

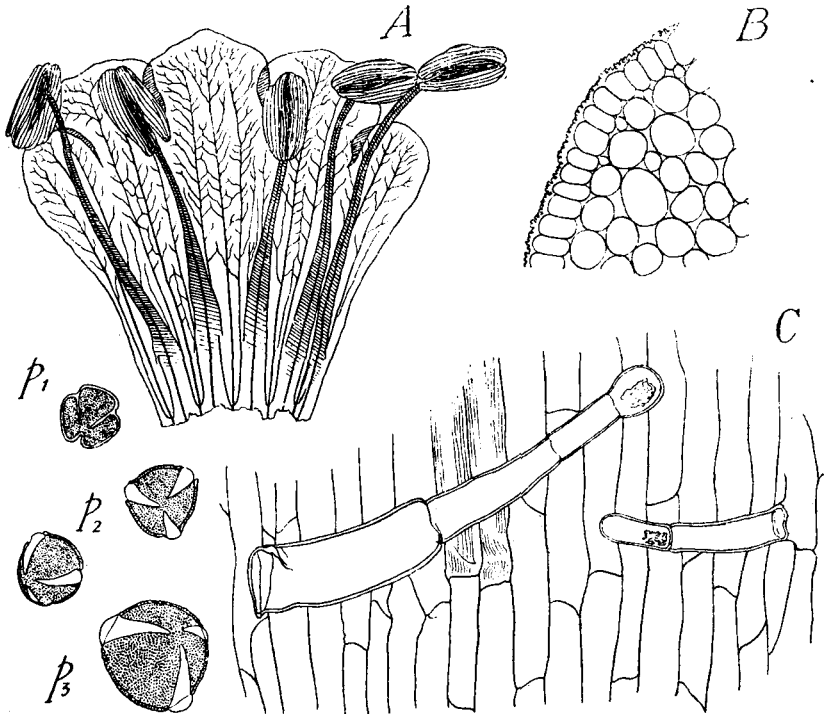


FIG. 5. *H. muticus* Linn., A, corolla spread out showing venation and epipetalous stamens $\times 3$. B, transverse section through a filament showing striations on the cuticle, $\times 200$. C, epidermis of a filament, striations shown on two cells only, $\times 200$. p 1, immature pollen grain, $\times 200$; p 2, mature pollen grains, $\times 200$; p 3, pollen grain $\times 350$ to show arrangement of the pits more clearly.

2. The dimensions of the cells are approximately L=60 to 120 to 240 μ , R=15 to 21 to 27 μ and T=9 to 15 to 30 μ ;

3. The trichomes (Fig. 5, C) are usually glandular with a sub-spherical or elongated unicellular head and a 1 to 6-celled uniseriate stalk; they are approximately 60 to 180 to 630 μ long and 24 to 30 to 90 μ in diameter at the base;

4. The few covering trichomes present have a cylindrical and rounded tip and are only slightly tapering.

Anther: The histology of the anther is in general quite similar to that of *H. niger*. The main points of difference are:

1. The *crystals* in the connective are prisms and double pyramids and some are dumb-bell or hour-glass shaped and others are of irregular shapes;

2. The epidermal cells on the lobes measure approximately L and T= 9 to 24 to 33 to 75 μ and R=25 to 45 μ , at the stomium, L and T=3 to 6 μ and R=6 to 9 μ ;

3. The cells of the fibrous layer which are also partly lignified, measure approximately L=15 to 30 to 45 μ , T=15 to 45 to 90 μ and R=15 to 36 to 90 μ .

Pollen grains: (Fig. 5, P₁, P₂, and P₃). The pollen grains closely resemble those of *H. niger* in shape and the situation of pores and furrows. They measure approximately 39 to 45 to 48 μ in diameter when mounted in lactophenol and 45 to 54 to 63 μ when boiled in chloral hydrate solution; the diameter of the pores is 6 to 9 to 15 μ . The exine is pitted with unequal rounded pits, the latter being frequently arranged in short longitudinal or inclined rows of 3 to 8 pits, unlike those in *H. niger* which are irregularly arranged. The pollen grains contain small oil globules and minute starch grains which measure approximately 1 to 5 μ in diameter after treatment with chloral-iodine reagent, being similar to those of *H. niger* in these respects.

HISTOLOGY OF THE STIGMA AND STYLE

The *stigma* (Fig. 6, D₁, D₂ and D₃) differs slightly from that of *H. niger* in not being so markedly capitate. It is either in the form of a small protuberance covered with papillae and a slight funnel shaped hollow in the centre, or occasionally the style ends in a stigma having the same diameter as the style with a funnel shaped depression. The papillae are rather cylindrical in form, the papillose cells measure approximately 30 to 45 to 90 μ in length and 15 to 24 to 30 μ in diameter at the base.

The *style* (Fig. 6, A), which is about 15 to 30 mm. in length and 0.1 mm. in diameter, is similar to that of *H. niger*. The cells of the epidermis (Fig. 6, B) resemble those of *H. niger* in shape; they measure approximately L=90 to 120 to 260 μ , T=9 to 24 to 45 μ and R=21 to 27 to 42 μ . In certain epidermal cells the anticlinal walls appear wavy, which is probably due to shrinkage. *Stomata* often occur on the style, but are not elevated as in case of *H. niger*. Slender 2- to 3-celled uniseriate covering *trichomes* with a rounded tip occur on the style but are rare (Fig. 6, tr); they measure about 54 to 165 μ in length and 15 to 165 μ in basal diameter.

The style in transverse section (Fig. 6, C) shows an arrangement identical with that of *H. niger*; the larger parenchymatous cells in the outer band measure approximately 15 to 42 μ in diameter, the smaller cells of the central core measure about 4 to 8 μ in diameter; the lacuna in the centre is usually larger than in *H. niger*, and occasionally a few

THE FLOWER OF *HYOSCYAMUS MUTICUS* LINN.

pollen grains may be observed in the lacuna. The cells of the fundamental parenchyma abound with minute starch grains measuring about 1 to 6 μ in diameter.

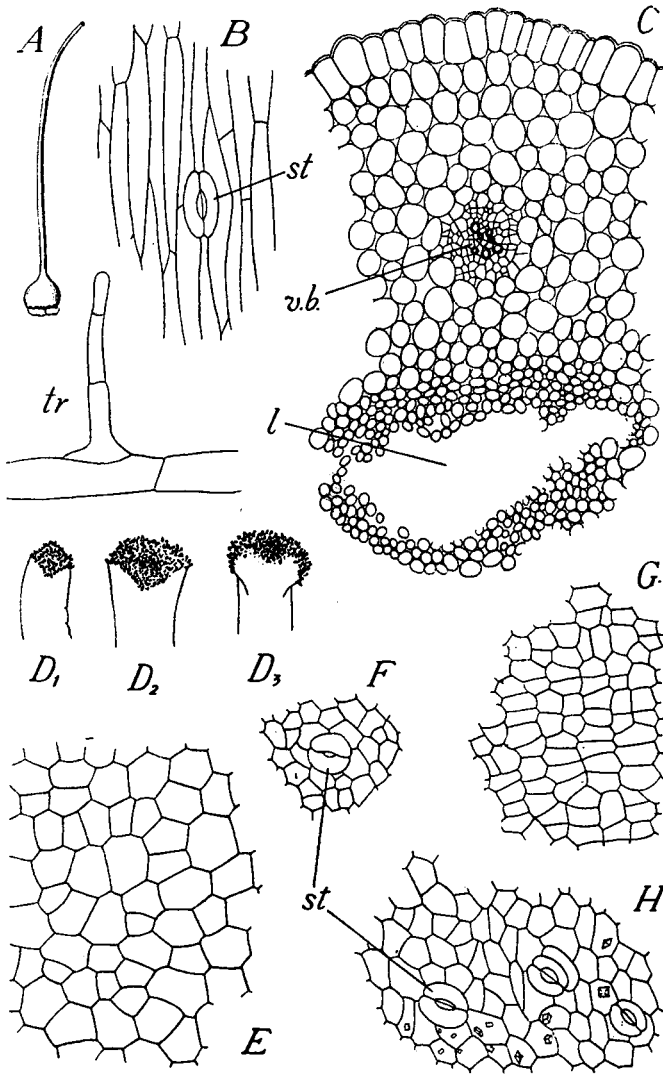


FIG. 6. *H. muticus* Linn., Carpels. A, entire gynoecium, $\times 1.2$; B, epidermis of style, C, transverse section of the style; D1, D2, D3, three forms of stigma $\times 11$; E, outer epidermis over apical part of the ovary. F, part of the upper epidermis at the base of the ovary showing a stoma. G, outer epidermis at the base of the ovary. H, inner epidermis of the ovary wall near the base, showing crystals in certain cells. *l*, lacuna; *st*, stoma; *tr*, trichome from the style; *v.b.*, vascular bundle. All $\times 160$, except A and D.

THE OVARY

The cells of the *outer epidermis* of the ovary wall are similar to those of *H. niger* (Fig. 6, E, F and G); the cells are polygonal tabular in form, being slightly larger in the upper region; they measure approximately L and $T=6$ to **24 to 30** to 45μ and $R=12$ to **24** to 30μ . *Stomata* are absent on the outer epidermis but very rarely occur at the base with the pore usually open (Fig. 6, F). The *inner epidermis* has cells similar to those on the outer but are slightly smaller (Fig. 6, H). *Stomata* are frequent and are usually open; they do not always show cruciferous arrangement of subsidiary cells.

The *mesophyll* consists of quite closely arranged parenchymatous cells traversed by about 30 vascular strands running vertically (about 50 in *H. niger*). The *septum* and *placenta* are similar to those of *H. niger* except that the *crystals* in *H. muticus* are usually double pyramids, prisms, cubes or other irregular shapes.

The *ovules* consist of undifferentiated thin-walled parenchymatous cells.

POWDERED FLOWERS

The dried flowers of *H. muticus* ground to No. 90 mesh, result in a greenish-grey powder with an unpleasant musty odour which differs markedly from that of *H. niger*. The following mountants were used for the examination of the powder; dilute glycerin, iodine solution, solution of chloral hydrate, lactophenol and phloroglucin and hydrochloric acid. For the determination of the structural features, the most useful of these mountants are (1) solution of chloral hydrate and (2) phloroglucin and hydrochloric acid. The following is a list of characters useful for the identification of the drug arranged in order of importance :

1. Pollen grains; mature grains measuring about 40 to 50μ in lactophenol and about 50 to 60μ after boiling in chloral hydrate solution. In general, they resemble the pollen of *H. niger* excepting for the sculpturing of the exine, which differs slightly in that the numerous pits are often arranged in short rows.
2. Broken pieces of simple or branched glandular trichomes from the calyx having unicellular globular heads and usually covered with a markedly striate cuticle.
3. Fragments of the inter-neural regions of the calyx having wavy-walled epidermal cells accompanied by cruciferous stomata and covered with a striated cuticle; crystals of calcium oxalate having the form of double pyramids, prisms or irregular shaped, present in the cells of the mesophyll and of the epidermises.
4. Fragments of the basal part of the corolla tube; the rectangular epidermal cells having lignified cell walls which do not bear pits on their anticlinal walls as in case of *H. niger*.

5. The other characters, e.g., the fibrous layer of the anther wall, show no distinction from those of *H. niger*.

Note: Probably due to the method of preparation of the drug in commerce, the colouring matter in the petals and anthers is destroyed and the colour does not show in patches around fragments of the corolla and anther walls in chloral hydrate mounts as it does in the case of *H. niger*.

SUMMARY

The chief histological characters of the flower of *H. muticus* of special diagnostic value, as compared with those of the flower of *H. niger*, are:

1. *Calyx*: Epidermal cells wavy-walled on lobes to almost straight-walled at the base; epidermis covered with thick striated cuticle; cruciferous (anisocytic) stomata frequent; numerous glandular trichomes often branched and terminating in unicellular heads, basal cells covered with striated cuticle; numerous crystals present in all parts of the calyx including the epidermises either singly or as aggregates; lateral veins branch before reaching the sinuses; marginal veins absent.

2. *Corolla*: Epidermal cells with straight walls or somewhat wavy on the outer surface of the lobes and with a finely striated cuticle; epidermal cells at the base of the tube lignified; glandular trichomes on outer epidermis and covering ones on both epidermises of the tube; many cells of the inner epidermis of the tube bear papillae; stomata present on the outer epidermis at the middle of the tube; small crystals of calcium oxalate present mainly along the veins.

3. *Stamens*: *Filament*, strongly striated cuticle; trichomes mostly glandular, more frequent towards the base. *Anther*, a few stomata on epidermis, fibrous layer slightly lignified; crystals in connective present as prisms, pyramids, dumb-bell or hour-glass shaped. *Pollen grains*, spherical, about 40 to 60 μ in diameter, with 3 pores and 3 furrows; pits on exine often arranged in short rows; contain oil globules and starch grains.

4. *Gynæcium*: *Stigma*, somewhat capitate with a slight depression in the centre; covered with comparatively short papillae. *Style*, stomata on epidermis, starch grains abundant in ground tissue. *Ovary*, bilocular, a few ranunculaceous stomata are present at the base of the outer epidermis and are frequent on the inner epidermis; starch grains and crystals present, the latter being single or in aggregates and occurring in all tissues excepting the outer epidermis. *Ovules* numerous.

5. *Powder*: The powder of the flowers of *H. muticus* may be identified by the characters of the pollen grains, the simple or branched glandular trichomes and the striated cuticle on the epidermal cells of the calyx.

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S. ROHATGI

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