A COMPARATIVE STUDY OF MARYLAND SENNAS

By

Frank J. Slama

Thesis submitted to the Faculty of the Graduate School of the University of Maryland in partial fulfillment of the requirements for the degree of Doctor of Philosophy.

1935
ACKNOWLEDGEMENT.

This investigation was begun under the supervision of the late Professor Plitt.

For his valuable criticism and direction, I wish to thank sincerely Dean DuMez.

I wish to express my appreciation to Professor Norton for his efforts and suggestions and also to Dr. Bamford, particularly for histological criticisms.
I. Introduction

The Maryland sennas* with the exception of Cassia Medsgeri, were studied by botanists as far back as the seventeenth century. At that time Tournefort (1656-1708), after a study of these sennas, placed them in the tribe Cassia, and later Linne, in his Species Plantarum left this classification unchanged. Willdenow, in his work also accepted the classification of his predecessors. Gray, in his Manual of Botany in which the four Maryland species are described, keeps them together and his classification is otherwise very similar to Linne's. However, in recent years some botanists have questioned this classification and have divided the

Chamaecrista camporum Greene, Fittonia 5:108. 1903.  
Chamaecrista fasciculata (Michx.) Greene, Fittonia 3:242. 1897.  

Cassia procumbens L. Sp. Pl. 380. 1753.  
Nictitella amena Raf. Sylva Tell. 128. 1838.  
Cassia nictitans commixta Millsp. West Va. Geol. Surv. 5:283. 1913.  

Cassia acuminata Moench, Meth. 273. 1794.  
Cassia reflexa Salisb. Prodr. 326. 1796.  
Ditremexa marilandica (L.) Britton and Rose.  

Cassia Medsgeri Shafer, Torreya 4:179. 1904.  
Ditremexa Medsgeri (Shafer) Britton and Rose.
Maryland sennas into two groups: Cassia, which includes Cassia Marylandica and Cassia Medsgeri; and Chamaecrista, which includes Cassia nictitans and Cassia Chamaecrista. Brittton and Rose also separated the Maryland sennas into two groups: Ditremexa and Chamaecrista, respectively. However, up to the present time no definite conclusions have been reached. Most of the work attempting to decide the question has been carried out only upon the flowers and pods.

In this paper I have made a pharmacognostic study of the various organs of the Maryland sennas and from this study I feel that I have obtained results which are of value in deciding this question.

II. History.

"Senna," according to Joensson, is one of the very oldest as well as most widely used of crude drugs. It was buried in the tombs of the Pharaohs and was widely used at the time of Alexander the Great. Senna first came into Europe about the 9th or 10th century." Martins and LaWall also agree that the Arabian physicians introduced the drug to Europe, and Mesue Senior (777-857), a celebrated physician, is credited by the latter with the introduction of these mild laxatives into medicine. When senna was
first used medicinally is unknown, as it was widely employed as a folk remedy before its introduction into medicine.¹

Senna has been officially recognized in this country as a therapeutic agent for more than a century. It was admitted to the first edition of the Pharmacopoeia of the United States, which appeared in 1820, and has been included in all subsequent editions, including the present. Senna is also included in the following foreign pharmacopoeias: Mexican Pharmacopoeia, 1925; Roumanian Pharmacopoeia, 1926; Norweigian Pharmacopoeia, 1913; Netherlands Pharmacopoeia, 1926; Swedish Pharmacopoeia, 1925; Italian Pharmacopoeia, 1920; Swiss Pharmacopoeia, 1907; German Pharmacopoeia, 1926; Argentine National Pharmacopoeia, 1928; Belgian Pharmacopoeia, 1906; British Pharmacopoeia, 1932; Russian Pharmacopoeia, 1929; French Codex, 1920; Danish Pharmacopoeia, 1907. For unknown reasons, the Spanish Pharmacopoeia, 1926, gives S. Hispanica as its official senna.

Of the Maryland sennas, C. Marilandica alone was official in the United States Pharmacopoeia, being recognized as such from 1820 to 1870. Perot⁹ showed by his researches that "C. Marilandica is very weak, if possessing any purgative properties at all, and entirely inferior to the imported senna" and as other workers obtained similar results, the reason for dropping
this senna from the later editions of the Pharmacopoeia of the United States is obvious.

No work has hitherto been carried out on C. Medsgeri. This is most likely due to the fact that its characteristics are so similar to those of C. Marilandica that it was generally identified as the latter. Both chemical and pharmacognostical work has been done on C. Marilandica.\textsuperscript{10,11,12,13}

C. nictitans and C. Chamaecrista also have not been studied extensively. Gallaher,\textsuperscript{14} in a chemical analysis of C. nictitans states: "Cathartic acid could not be prepared from it, but the powder was ascertained to have a laxative effect." This is the only important research that has been done on C. nictitans, and no work of any great value has been carried out on C. Chamaecrista.

In 1933, Slama\textsuperscript{15} reported a pharmacognostical study of the official sennas, Cassia Senna and C. angustifolia and the four Maryland sennas: C. Marilandica, C. Medsgeri, C. nictitans and C. Chamaecrista. The stomata, neighboring cells, epidermal cells and the distribution of epidermal hairs of the upper and lower surfaces of the leaflets were compared. A study was also made of the margins, apices, petiolules, and glands on the petioles and the cross sections of the leaflets. From the differences noted, the sennas were separated into three groups: Group I, the official
sennas; Group II, C. Marilandica and C. Medsgeri, and Group III, C. nictitans and C. Chamaecrista.

III. Habitat of the Maryland Sennas.

C. Marilandica is found near the Hillen Road, Baltimore and at Owings Mills, Spesutie Island, Havre de Grace and Allegheny County. It grows in soil which is somewhat rich and damp. Full-grown C. Marilandica plants are about four feet high.

C. Medsgeri is very similar in appearance and size to C. Marilandica and like the latter, it is found in somewhat rich, damp soil. C. Medsgeri is rather rare in Maryland and only one colony, situated about three miles east of Carney, Maryland, is known. However, specimens from Bladensburg and Redgate, Maryland were examined at the United States National Herbarium.

C. Chamaecrista and C. nictitans are found nearly always in dry, sandy soil. The former, which attains a height of about two feet when mature, is found in Maryland at Rosedale, Garland, South River, Mitchellsville, Chestertown, Snow Hill, College Park, Quantico, Brooklyn, Essex, and various scattered places in the close vicinity of Baltimore.

C. nictitans greatly resembles C. Chamaecrista, but it is smaller. This plant was gathered on a slope at Cromwell's Woods,
Brooklyn, Md., Gwynns Falls (Baltimore) and South River, Md.

It also grows at Emmitsburg, Salisbury, Preston, Chesapeake City, Mitchellsville and Newark. *C. nictitans*, like the other sennas, grows in the open where the sunlight is plentiful.

Outside of Maryland the distribution of the four species is as follows: *C. Marilandica* -- Massachusetts to Indiana, North Carolina and Tennessee; *C. Medsgeri* -- Pennsylvania to Iowa, Georgia, Kansas and Texas; *C. Chamaecrista* -- Massachusetts to Florida, Ohio, South Dakota, Kansas and Texas; and *C. nictitans* -- Maine to Georgia, Ohio, Kansas, Oklahoma and Texas.

The differences in appearance and habitat of these sennas are shown by the following photographs taken in the field.
Fig. 1 — C. Marilandica Plant.
Fig. 2 -- Flowering Top of C. Marilandica.
Fig. 3 -- C. Marilandica Flowers.

Fig. 4 -- C. Chamaecrista Flowers.
Fig. 5 — C. Chamaecrista Plant
Fig. 6 — *C. nictitans* Plant.
IV. Descriptive Studies

A. Macroscopic

1. Leaflets

a. Kind of leaves. All of the senna plants have paripinnately-compound leaves.

b. Shape of leaflets. The Maryland senna leaflets are inequilateral and of oblong-lanceolate or oval-oblong shape.

c. Average size of leaflets. The leaflets were pressed immediately after collecting. The average sizes of twenty leaflets of each species are as follows:

- C. Marilandica, 42.68 mm. by 15.48 mm., C. Medsgeri, 35.15 mm. by 15.25 mm., C. nictitans 8.45 mm. by 2.25 mm. and C. Chamacrista 10.75 mm. by 3.25 mm. The official senna leaflets were taken from stock, and their average sizes are C. angustifolia 26 mm. by 7.85 mm., C. Senna 22.6 mm. by 8.1 mm.

d. Shrinkage of leaflets. After the leaflets of C. Marilandica were completely dried out, the decrease in
length and width was found to be 4.74% and 7.05%, respectively. C. Medsgeri showed a decrease of 2.9% in length and 5.3% in width. C. Chamaecrista and C. nictitans showed a decrease of 13.7% and 6.6% in length and 25.8% and 13% in width, respectively.

e. Number of leaflets per rachis

C. Marilandica and C. Medsgeri exhibit from 5 to 9 pairs of leaflets per rachis. C. Chamaecrista and C. nictitans have from 5 to 10 and 10 to 20 pairs of leaflets per rachis, respectively.

f. Appearance of leaflets near the close of the growing season. Naturally, during the growing season, the plants are in a healthy, vigorous condition and as a consequence all the leaflets are green. As the growing season terminates, the green color of many leaflets is altered and because of this change, one can separate the Maryland sen- nas into two groups. The green color of the leaflets of C. Marilandica and C. Medsgeri plants, which are rather difficult to identify in the field, is changed to a deep yellow while the leaflets of C. Chamaecrista and C. nictitans exhibit a dark brown, almost black color. Besides being able to sep-
arate the Maryland sennas into two groups, one can easily differentiate between the C. Medsgeri and C. Marilandica plants, as the former shows a greater percentage of green leaflets per plant. However, in the case of the two remaining Maryland senna plants, the leaflets are of no value as a test for separating them, as both classes of plants exhibit leaflets of the same color.

**g. Venation**

Upon examining the leaflets, one may readily see that the venation occurring in Cassia nictitans and C. Chamaecrista differs greatly from that of the two remaining species (Figs. 7 to 10). In C. nictitans and C. Chamaecrista the midrib is closer to one margin and consequently it divides the leaflet into two unequal parts, the larger showing the veins more numerous and longer, and usually with three veins originating at the petiolule (Fig. 11). The average leaflet of C. Marilandica or C. Medsgeri has the midrib running practically through the center of the leaflet and has veins similar to each other in size and number on either side of the midrib.

Now if the leaflets of C. nictitans and C. Chamaecrista are examined with the naked eye or a 5X hand
Fig. 7 — C. Marilandica Leaflet. x 2½

Fig. 8 — C. Medsgeri Leaflet. x 2½

Fig. 9 — C. nictitans Leaflet. x 3

Fig. 10 — C. Chamaecrista Leaflet. x 3
Fig. 11 — Base of *C. Chamaecrista* Leaflet Showing Venation.
leaves, the veins are seen extending very close to the margins without any apparent division into veinlets; but on the contrary, the remaining senna leaflets show the terminal branches of the veinlets anastomosing near the margins. (Figs. 7 - 10).

2. Glands.

a. Description

Although C. Marilandica and C. Medsgeri exhibit glands which are green and which may or may not be situated at the base of the lowest pair of leaflets, nevertheless the former produces either conical or club-shaped glands, while C. Medsgeri presents only conical glands. C. Chamaecrista and C. nictitans have large and small cup-shaped glands, respectively, and in both cases the glands which are dark red or green tinted red, are always found beneath the lowest pair of leaflets.

With the exception of C. Marilandica, all the remaining sennas produce only single glands. C. Marilandica shows both single and double glands present and the latter are found on some of the petioles of the upper half of the plant. The remaining petioles of the plant bear single glands. (Table No. 1).
b. Location.

All the Maryland sennas exhibit glands which are found only on the petioles.

c. Distribution of glands.

The glands of C. Chamaecrista and C. nictitans are distributed rather uniformly on all petioles and are distant from the stem approximately 4 mm. and 3 mm., respectively. In C. Medsgeri and C. Marilandica, the glands are not distributed uniformly on the petioles as in each species the distance of the gland from the stem increases gradually toward the base of the plant. Furthermore, only the petioles on the lower portion of each plant have the glands situated at the base of the lowest pair of leaflets. C. Marilandica has more leaves per plant than C. Medsgeri, approximately twice as many.

d. Character of glands at the close of the growing season.

After the glands have ceased secreting, they gradually shrink and become smaller until at the end of the growing season they are dried up completely. At this time, the glands of C. Medsgeri and C. Marilandica are so brittle that very little effort is required to separate them from their respective petioles and, as a consequence, many glands drop off and disappear from the plants. On the other hand, the glands of C. Chamaecrista and
C. nictitans are always present on both living and dried plants.

e. Comparison of the glands.

A comparison was made of the characteristics exhibited by the glands, and the differences noted are shown in Table No. 1.

Table No. 1

Comparison of the Glands

<table>
<thead>
<tr>
<th>Plant</th>
<th>Shape of gland</th>
<th>Color of Glands</th>
<th>Remarks</th>
</tr>
</thead>
<tbody>
<tr>
<td>C. Marilandica</td>
<td>Conical (or club-shaped)</td>
<td>Green</td>
<td>Some glands are situated at base of lowest pair of leaflets.</td>
</tr>
<tr>
<td>C. Medsgeri</td>
<td>Conical</td>
<td>Green</td>
<td>Some glands are situated at base of lowest pair of leaflets.</td>
</tr>
<tr>
<td>C. Chamaecrista</td>
<td>Large cup-shaped glands</td>
<td>Dark red or green tinted red</td>
<td>Glands always beneath the lowest pair of leaflets.</td>
</tr>
<tr>
<td>C. nictitans</td>
<td>Small cup-shaped glands</td>
<td>Dark red or green tinted red</td>
<td>Glands always beneath the lowest pair of leaflets.</td>
</tr>
</tbody>
</table>

f. Conclusions

From this study of the glands it is concluded that:

(1) C. Medsgeri, C. Chamaecrista and C. nictitans produce only
single glands, whereas *C. Marilandica* exhibits both single and double glands.

(2) The glands are distributed rather uniformly only on the petioles of *C. Chamaecrista* and *C. nictitans*.

(3) *C. Marilandica* has a greater number of leaves per plant than *C. Medsgeri*.

(4) On the basis of data presented in Table No. 1 it is possible to separate the Maryland sennas into two groups: (1) *C. Marilandica* and *C. Medsgeri*, and (2) *C. Chamaecrista* and *C. nictitans* because of the differences shown in the shape, color and the distribution of the respective glands.

3. Flower Buds.

**Table No. 2 - Comparison of the Flower Buds**

<table>
<thead>
<tr>
<th>Plant</th>
<th>Appearance of buds</th>
<th>Position of buds</th>
<th>Shape of buds</th>
<th>Ratio of length to width of buds</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>C. Medsgeri</em></td>
<td>First week in July</td>
<td>In short axillary racemes at top of plants</td>
<td>Elliptical</td>
<td>1.5 to 1</td>
</tr>
<tr>
<td><em>C. Marilandica</em></td>
<td>&quot;</td>
<td>&quot;</td>
<td>Elliptical</td>
<td>1.5 to 1</td>
</tr>
<tr>
<td><em>C. Chamaecrista</em></td>
<td>Middle of July</td>
<td>In small clusters above the axils. Occasionally flower buds are found in the axils</td>
<td>Conical</td>
<td>3 to 1</td>
</tr>
<tr>
<td><em>C. nictitans</em></td>
<td>&quot;</td>
<td>&quot;</td>
<td>Conical</td>
<td>3 to 1</td>
</tr>
</tbody>
</table>
In Table No. 2, additional data are given which are of importance in the separation of the Maryland sennas into two groups.

4. Flowers

a. Color

The only characteristic common to all the flowers of the Maryland sennas is the yellow color.

b. Appearance of mature flowers.

C. Medsgeri and C. Marilandica produce the first full-grown flowers about the middle of July. Full-grown flowers are first seen on C. Chamaecrista and C. nictitans plants about August 1.

c. Height of flowering season.

C. Marilandica exhibits the greatest number of flowers the first week in August, and C. Medsgeri about the third week in July. With C. Chamaecrista and C. nictitans, the height of the flowering season occurs the first week in August.
d. Disappearance of flowers.

By this means one can again separate the Maryland sennas into two groups: one consisting of C. Medsgeri and C. Marylandica, whose last flowers of the season disappear about August 2 and August 13 respectively; and the second group comprising C. Chamaecrista and C. nictitans plants, about one-half of which exhibit flowers until the end of the growing season.

e. Size of flowers.

The full-grown flowers of C. Marylandica, C. Medsgeri and C. Chamaecrista are normally about 3/4 inch in diameter with occasional flowers 1 inch in diameter. C. nictitans has the smallest flowers, which rarely exceed 1/4 inch in diameter and which are borne on very short, barely perceptible, pedicels.

f. Size of floral organs.
Table No. 3. - Size of Floral Organs.

<table>
<thead>
<tr>
<th>Plant</th>
<th>Sepals</th>
<th>Petals</th>
<th>Stamens</th>
<th>Pistil length</th>
<th>Pedicel length</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>C. Marilandica</strong></td>
<td>Five</td>
<td>Five</td>
<td>Ten</td>
<td>10 mm.</td>
<td>15 mm.</td>
</tr>
<tr>
<td></td>
<td>Three small 3 mm. x 4 mm.</td>
<td>Two small 2 mm x 4 mm.</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>C. Medsgeri</strong></td>
<td>Five</td>
<td>Five</td>
<td>Ten</td>
<td>11 mm.</td>
<td>14 mm.</td>
</tr>
<tr>
<td></td>
<td>Three large 2½-3 mm. x 6 mm.</td>
<td>Two small 2½ mm. x 5 mm.</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>C. Chamaecrista</strong></td>
<td>Five</td>
<td>Five</td>
<td>Ten</td>
<td>13½ - 16 mm.</td>
<td>16 mm.</td>
</tr>
<tr>
<td></td>
<td>Three large 3½-4 mm. x 14-15 mm.</td>
<td>Four small 7-9 mm x 12-15 mm.</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>C. niititans</strong></td>
<td>Five</td>
<td>Five</td>
<td>Ten</td>
<td>3-4 mm.</td>
<td>1 mm.</td>
</tr>
<tr>
<td></td>
<td>Three large 1 mm. x 4 mm.</td>
<td>Four small 2½ x 3½ mm.</td>
<td>Five of nearly equal length</td>
<td>Five stamnodes</td>
<td>2½ - 3 mm.</td>
</tr>
</tbody>
</table>
From the results in Table 3, it is concluded that:

(1) All the flowers consist of five sepals, five petals, ten stamens and one pistil (5-5-10-1). However, C. nictitans flowers differ slightly from those of the other three species in that staminodes are present.

(2) C. nictitans flowers show the smallest pedicels. The pedicels of the remaining senna flowers are approximately of the same length.

(3) C. nictitans is the only Maryland senna in which the stamens are practically of equal length.

(4) The flowers of all the Maryland sennas show the outermost floral whorl made up of five sepals -- three large and two small.

(5) The corollas of C. Marilandica and C. Medsgeri flowers consist of 5 almost regular petals.

(6) In all flowers the styles are curved and point towards the top of the flowers. All stigmas are capitate.
g. Differentiation of the senna flowers.

If the flowers of C. Marilandica and C. Medsgeri are examined, it is immediately apparent that they resemble each other very closely. They are approximately of the same color and size; their construction is very similar, and the whorls of floral organs consist of the same number of parts. After a careful study of these flowers, it was found that either could be identified correctly by means of the pistils. The pistil of the C. Marilandica flower is villose, although the underside and the basal portion of the pistil are devoid of hairs. On the other hand, the C. Medsgeri flower shows a completely pubescent pistil, the style being slightly less pubescent than the ovary. However, in all other respects, the two flowers are identical, and in both cases the hairs which are present on the pistils always point towards the bottom of the flower.

It is a rather simple procedure to differentiate between C. nictitans and C. Chamaecrista flowers. The former flower is very small and is the only flower of the Maryland sennas to exhibit five fully-developed stamens of nearly equal length. Moreover, the C. Chamaecrista flowers may further
be identified by the presence of red patches at the base of several petals. The majority of the flowers show only three petals with red patches. In some flowers, all petals are so marked but in this case, two of the petals always exhibit the patches of a faint red color. The C. Chamaecrista petal is the only one that shows the red color present, but the petals of the remaining senna flowers are always uniformly yellow in color and are never marked with red.

Diagram A shows how the flowers C. Marilandica, C. Medsgeri and C. Chamaecrista are constructed; while diagram B shows the C. nictitans flower with ten stamens, five of which have not developed.
In Table No. 5, Size of Floral Organs, it was stated that the flowers of C. Marilandica, C. Medsgeri and C. Chamaecrista showed ten stamens which were of three different sizes. Actually, stamens of five different lengths make up the androecium of any one of the three flowers named. Stamens 1, 2, and 3 are the small ones and are of the same length. The medium-sized stamens are represented by stamens 4, 5, 9 and 10 of which 4 and 10 are slightly shorter than 5 and 9. Furthermore, of the three large stamens 6, 7, and 8, the middle stamen (7) is slightly shorter than the remaining two. Nevertheless the difference in the sizes of the stamens of any particular group is so slight that the stamens as a whole, were divided into three groups, and the average size for each group was tabulated.

In the same table, it was further stated that all the Maryland senna flowers have irregular calyxes, made up of five sepals (three large and two small). One of the two small sepals is always found at the bottom of the flower in position 4, while the remaining small sepal may occupy either position 1 or 2. However, the majority of
the flowers show the small sepals in positions 4 and 1. In all flowers the sepals are scarcely united at the base.

5. Stems

a. Size

The results obtained from a study of the sizes of the Maryland senna stems, point once more to their separation into two groups. C. Medsgeri and C. Marylandica stems have an average diameter of 5 mm. as compared to a diameter of 1.5 mm. for C. Chamaecrista and C. nictitans. Naturally, the stems at the top and bottom of the plants differ slightly in their diameter; and this is also true of any stem at the beginning and the end of the vegetative cycle. Therefore, all the stem measurements were made at a point midway between the base and top of the plants. The results which were obtained at the time of flowering and again at the close of the growing season when compared, differed only very slightly from the average diameters given above.

b. Color
At the start of the growing season all the senna plants have green stems. The stems of *C. Marylandica* and *C. Medsgeri* plants remain green until the approach of fall when they turn yellow. However, *C. Chamaecrista* and *C. nictitans* stems behave differently. About the latter part of May, these stems have some of their green color replaced gradually by red. The majority of the latter plants show the entire inner face of any stem (the half of the stem which faces the central axis of the plant) colored red, often dark red while the remainder of the stem is dark green. The *C. Chamaecrista* and *C. nictitans* stems do not change their color until the approach of fall and, at this time, about one-half of the plants have brown stems. By the end of October, all the senna plants have stems that have become dark brown or black.

It is also possible to separate the Maryland sennas into two groups by the color of the fractured ends of the stems. If the stems of *C. Marylandica* and *C. Medsgeri* plants are broken, the exposed central cylinders are light orchid and deep orchid, respectively. On the other hand, *C. Chamaecrista* and *C. nictitans* stems, if treated in a similar manner, always show white, or cream-
colored central cylinders.

c. Arrangement

If a study is made of the arrangement of the stems on the Maryland senna plants, it may be readily seen that two kinds of stem arrangements are present and that they differ to such an extent that it is possible to separate the Maryland sennas into two groups (Figs. 1, 5, 6). In the case of C. Marylandica and C. Medsgeri plants there is only one stem present from which all the leaves arise. On the other hand with C. Chamaecrista and C. nictitans plants, the main stem branches near the ground, and secondary branching also occurs.

d. Epidermal hairs

With the exception of C. Medsgeri, the remaining senna stems show the presence of many hairs which are arranged differently on each. For example, C. Marylandica has the largest hairs distributed irregularly over the stem, although their number seems to increase
as the top of the plant is approached. However by making use of the distribution of the hairs on the stems, one may differentiate between C. Chamaecrista and C. nictitans plants. The stems of the former produce hairs only on the inner face, or the dark red portion of the stem, while the dark green portion is devoid of hairs.

Hairs are distributed uniformly over the entire stems of the C. nictitans plants. In all the plants which produce stem trichomes, the hairs increase in number as one approaches the top of the plants.

The results of a study of the distribution of the hairs on the stems of the Maryland senna plants are shown in the following chart.

<table>
<thead>
<tr>
<th>Trichomes on the Epidermis of the stems</th>
<th>Long Hairs</th>
<th>Rarely found, stems practically devoid of hairs -- C. Medageri</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Short Hairs</td>
<td>Hairs present only on inner face of stems -- C. Chamaecrista</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Stems completely encircled by hairs -- C. nictitans</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Straight hairs many, irregularly arranged -- C. Marylandica</td>
</tr>
</tbody>
</table>

Note: During the entire growing season, only two hairs
were seen on the stems of C. Medsgeri plants. The hairs were long and very similar to the C. Marilandica hairs in all respects.


a. Shape and distribution.

Once more the Maryland sennas can be divided into two groups by means of the different characteristics exhibited by their stipules. (Figs. 12-15). The stipules of C. Marilandica and C. Medsgeri are the largest. They are spatulate or lanceolate, usually with parallel margins and apices which terminate in short, acute points. On the contrary, C. Chamaecrista and C. nictitans stipules are smaller; they are triangular and are terminated by gradually-tapering, sharp points.

b. Color and duration.

The stipules of C. Marilandica and C. Medsgeri plants are green until a short time before they disappear. All the leaves are subtended by two stipules at the beginning of the growing season, but gradually the lower stipules become yellow and disappear until, by the middle of July, only the upper half of the plant shows stipules present. Near the end of the growing
season only the uppermost portions of the plants bear stipules, the majority being yellow. The persistent stipules of C. Chamaecrista and C. nictitans are green on the very young plants, but in a short time those situated at the bottom of the plants show their tips and margins colored red. However, as one passes up the plant, the red color gradually disappears from the margins until at the growing tip, the stipules exhibit only their tips of a red color.

7. Roots

a. Description

The roots of C. Marilandica and C. Medsgeri are perennial and grow horizontally in the soil. They are rough, almost black, somewhat knotty; stem scars are present and the roots have many rootlets which are stiff and leathery (Figs. 16, 17). The fracture is tough and splintery.

The roots of C. Chamaecrista and C. nictitans are annuals and grow vertically in the ground. They are reddish brown, smooth, soft and pliable, and all the rootlets
Fig. 16 — C. Marilandica Root

Fig. 17 — C. Medsgeri Root
Fig. 18 -- C. Chamaecrista Roots Showing Tubercles

Fig. 19 -- C. nictitans Roots Showing Tubercles
Fig. 20 -- *C. Chamaecrista* Tubercles

Fig. 21 -- *C. nictitans* Tubercles
arise from the tap root. Both the tap root and the rootlets bear tubercles (Figs. 18-21). Only the C. Chamaecrista and C. nictitans roots and rootlets show the presence of the cream-colored tubercles, and the larger tubercles are found on the former plants. The tubercles appear solid and firm throughout the greater portion of the growing season, but at the close of the growing season the roots show only the dried tubercles present. Their probable function is to house the bacteria which play an important role in the nitrogen cycle.

In the case of C. Marilandica and C. Medsgeri only the roots of the mature plants were examined for tubercles. It is possible that the roots of the seedlings or the young plants of these species would exhibit tubercles. However, attempts to germinate these seeds proved more or less unsuccessful. Furthermore, seedlings could not be found growing in their natural habitat.

8. Pods

a. General characteristics

The results of a study of the pods of the Maryland Sennas are shown in the following table.
Table No. 4 — Comparison of the Pods.

<table>
<thead>
<tr>
<th>Plant</th>
<th>Shape of undehisced pod</th>
<th>Average size of mature pods</th>
<th>Average no. of segments per pod</th>
<th>Average no. of pods per plant</th>
<th>Aver. size of segments</th>
<th>Ratio of length &amp; width of segments</th>
<th>Color of undehisced pods</th>
</tr>
</thead>
<tbody>
<tr>
<td>C. Marilandica</td>
<td>slightly curved</td>
<td>10.1 cm. long x 7 mm. wide</td>
<td>13.8</td>
<td>29</td>
<td>5 mm. long x 7 mm. wide</td>
<td>1:1.4</td>
<td>chocolate to almost black</td>
</tr>
<tr>
<td>C. Medsgeri</td>
<td>curved</td>
<td>8.5 cm. long x 9 mm. wide</td>
<td>16</td>
<td>10½</td>
<td>8 mm. long x 9 mm. wide</td>
<td>1:1.1</td>
<td>chocolate</td>
</tr>
<tr>
<td>C. Chamaecrista</td>
<td>straight</td>
<td>36.5 mm. long x 5 mm. wide</td>
<td>10.2</td>
<td>many</td>
<td>4 mm. long x 5 mm. wide</td>
<td>1:1.25</td>
<td>Yellowish green to red</td>
</tr>
<tr>
<td>C. nictitans</td>
<td>straight</td>
<td>22.7 mm. long x 3.8 mm. wide</td>
<td>5.6</td>
<td>many</td>
<td>3.75 mm. long x 4 mm. wide</td>
<td>1:1.07</td>
<td>Yellowish green to red</td>
</tr>
</tbody>
</table>

The pods of the four Maryland senna species are shown in Figures 22 to 25.
Fig. 22 — *C. Marilendica* Pods

Fig. 23 — *C. Medageri* Pods
Fig. 24 -- C. Chamaecrista Pods

Fig. 25 -- C. nictitans Pods
Under the heading of "average number of pods per plant" in Table No. 4, no definite number is placed opposite C. Chamaecrista and C. nictitans because of an oversight. However, it can be definitely that the majority of these plants produce more pods per plant than the two remaining sennas. I should roughly estimate that the average number of pods on the C. Chamaecrista and C. nictitans plants would be about 40, although one exceptionally large C. Chamaecrista plant showed 220 pods present.

Table No. 4 shows that it is also possible to separate the Maryland sennas into two groups when the pods are compared in the following ways:

1. Shape and size of pods
2. Average number of pods per plant
3. Average number of segments per pod
4. Color of pods
5. Size of the segments

b. Dehiscence of the Pods

1. C. Marilandica pods 9/30/33

The majority of the pods have dehisced and many are still
attached to the plants. Although the valves have separated sufficiently to allow the seeds to escape, nevertheless many pods show all the seeds present. The pods of C. Marylandica may be opened with little effort.

2. C. Medsageri Pods 9/30/33. At this time, all the plants showed pods that have not dehisced. These pods seem to lack the power of dehiscing and the valves adhere so tightly that, in several cases, a knife was needed to separate them.

3. C. Chamaecrista Pods 10/1/33. Numerous pods have dehisced and they are found in their characteristic twisted fashion. Other pods that have not dehisced as yet can be opened easily but one must be careful not to lose any seeds as the valves twist in order to eject them.

4. C. nictitans Pods 10/1/33. These behave almost the same as the C. Chamaecrista pods. However, the valves of the C. nictitans pods twist to a greater degree.
The following chart shows how the pods may be of value in recognizing the individual senna species after the growing season has terminated.

Mature Pods

- Large, curved (valves do not twist)
  - Chocolate to almost black, segments of pods (average 13.8) 1.4 as wide as long (7 mm x 5 mm) -- *C. Marilandica*
  - Chocolate, segments of pods (average 16) approximately as wide as long (9 mm x 8 mm) -- *C. Medageri*

- Small, straight (Valves twist)
  - Yellowish green to red, segments of pods (average 10.2) 1.25 as wide as long (5 mm x 4 mm) -- *C. Chamaecrista*
  - Yellowish green to red, segments of pods (average 5.6) almost as wide as long (4 mm x 3.75 mm) -- *C. nictitans*

9. Seeds

The results obtained from a study of the seeds of the Maryland sennas are shown in the following table.
Table No. 5 -- Comparison of the Maryland Senna Seeds

<table>
<thead>
<tr>
<th>Name of Plant</th>
<th>Color of Seeds</th>
<th>Shape of Seeds</th>
<th>Average Size of Seeds</th>
<th>Average number of seeds per pod</th>
<th>Fully develop seeds per pod (average)</th>
<th>Number of pods examined</th>
</tr>
</thead>
<tbody>
<tr>
<td>C. Marylandica</td>
<td>Brown</td>
<td>Fan-shaped</td>
<td>4.3 mm. wide x 4.9 mm. long</td>
<td>13.7</td>
<td>87.6%</td>
<td>30</td>
</tr>
<tr>
<td>C. Medsgeri</td>
<td>Brown or Olive</td>
<td>Elliptical</td>
<td>3 mm. wide x 4.6 mm. long</td>
<td>15.5</td>
<td>32.6%</td>
<td>30</td>
</tr>
<tr>
<td>C. Chamaecrista</td>
<td>Black</td>
<td>Somewhat rectangular</td>
<td>2.8 mm. wide x 4 mm. long</td>
<td>9.4</td>
<td>77.7%</td>
<td>30</td>
</tr>
<tr>
<td>C. niottitans</td>
<td>Black</td>
<td>Somewhat rectangular</td>
<td>2.6 mm. wide x 3.3 mm. long</td>
<td>5.4</td>
<td>77.7%</td>
<td>30</td>
</tr>
</tbody>
</table>

Table No. 5 shows that the physical characteristics of the seeds differ sufficiently to warrant the separation of the Mary-
land sennas into two groups.

C. Medsgeri shows the smallest yield of mature seeds and all mature pods of the plant show the presence of many brown, shrivelled, undeveloped seeds. In this case only the fully-developed seed is of an olive color and each flat surface shows a dark green elliptical depression which indicates the position and the shape of the enclosed embryo (Fig. 27). The C. Marilandica seeds also exhibit these depressions, but in this case the entire testa is of a uniform brown color (Fig. 26). However, with the C. Chamacrista and C. nictitans seeds these characteristic depressions are absent, but the testa instead is pitted.

Furthermore, the pits arrange themselves in longitudinal rows which make the unmagnified seeds appear striated or furrowed (Fig. 28). By means of these characteristics one, with the aid of a 10 X hand lens, can separate the Maryland sennas as shown in the following chart.

<table>
<thead>
<tr>
<th>Maryland Senna Seeds</th>
<th>Pits arranged in prominent longitudinal rows—</th>
<th>C. Chamaecrista.</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Longitudinal pit rows not quite so prominent—</td>
<td>C. nictitans.</td>
</tr>
<tr>
<td>Not shiny, Brown or Olive</td>
<td>Seed of olive color, depression dark green—</td>
<td>C. Medsgeri.</td>
</tr>
<tr>
<td></td>
<td>Depression present but entire seed of uniform brown color—</td>
<td>C. Marilandica.</td>
</tr>
</tbody>
</table>
Fig. 26 — C. Marilandica Seed
Fig. 27 — C. Medsgeri Seed

Fig. 28 — C. Chamaecrista Seed

NOTE: Seeds Magnified Approximately 25 times.
Behavior of the Maryland Sennas During the Growing Season.

A comparative study of the Maryland senna plants was made during the 1933 growing season. The purpose of this study which extended from the first week in May until the latter part of October was to ascertain whether the plants exhibited any important differences which would point to their separation. In this work first the vegetable cycle of each plant is fully described and finally, a comparison is made of the differences noted.

C. Marilandica. The first evidence that these plants have started to grow is shown during the first week in May, when young shoots sprout from the underground roots. Gradually the plants increase in size and by June 1 they are about two feet high. About July 1, they have not only attained a height of 3 or 4 feet, but flower buds have appeared on several plants. On July 18 the plants were again examined and a few full-grown flowers were first seen. Gradually the flowers increased in number until the first
week in August when the height of the flowering season occurred. However, after this period, the number of flowers began to decrease and by August 13, all the flowers had disappeared. In the meantime pods were being formed, very small ones appearing for the first time on July 23. On August 5, the largest pods were about 1/3 natural size, by August 10 they had attained two-thirds of their natural size and it was not until August 15 that full-grown, green pods could be obtained. From August 15 until the middle of September, very few changes occurred in these plants, the most important being the increase in the size of the pods. However, by the first of October approximately one-half of the plants were devoid of both leaflets and pods, and only the bare yellow stems remained. The remaining plants still showed pods which had begun to dehisce and also bore a majority of yellow leaflets. Nevertheless, in two or three weeks the plants practically disappeared and only occasionally a dried stem bearing a cluster of pods at its summit was to be found.
C. Medsgeri. These plants behaved like those of C. Marilandica during the first part of the growing season, that is, from the first week in May until the middle of July. In this case, the flower buds were first seen on July 2, and in about a week they started to open. By July 18, many plants exhibited several full-grown flowers, the greatest number appearing about July 24. During the latter part of July the number of flowers gradually decreased until the last flower disappeared on August 2. In this case very small pods appeared about July 15. However, on July 23, the pods were 2/3 natural size, and by August 2, many plants bore pods of natural size. From early August until the middle of September the only important change was shown by the pods, most of which had matured. During the first week of October, few yellow leaflets were present and the plants as a whole, were in a much better condition than those of C. Marilandica. In about three weeks, the plants of C. Medsgeri practically disappeared and only the bare stems, several of which still bore pods, remained.
C. Chamaecrista. The seeds of these plants start germinating the first week in May, and by the latter part of June seedlings, as well as plants 12 inches high, are to be found. However, flower buds first appeared on July 17, and within a week the number of flower buds increased greatly, with several blossoms present. On August 2, several plants were nearly 2 feet high, and all of them exhibited various amounts of full-grown flowers, which continued to increase in number until the middle of August when the maximum number was produced. As the growing season progressed the flowers gradually decreased in number, and pods in different degree of development occupied their places. However, all the plants exhibited many flowers until late in September. On October 1, some plants were devoid of green leaflets and only the stems bearing the pods were seen. Nevertheless, these latter plants were outnumbered by those having green leaflets and flowers, and quite a few C. Chamaecrista plants exhibited flowers and green leaflets almost until the end of the growing season, about October 15. The pods were first seen on August 10 and by August 18 all the plants exhibited many pods, several of average size.
Gradually the number of pods increased, and by the middle of September some of them had dehisced. On October 15 these plants were very inconspicuous, a great majority of the pods had dehisced and the leaflets that remained on the stems were shrivelled and dark brown. In about a week, plants had practically disappeared.

*C. nictitans*. These plants behaved to a great extent like those of *C. Chamaecrista*, although they are smaller and produce full-grown flowers somewhat later. The seeds of *C. nictitans* plants start germinating during the first week in May, and by June 23 the plants have attained a height of eight inches. The first flower buds were seen on July 19, and about a week later many flower buds and an occasional blossom were seen. However, by August 1 a few full-grown flowers were present on several plants, while the remaining plants showed an increase only in the number of flower buds. The greatest number of flowers was produced during the latter part of August, and flowers were still present on many plants as late as October 1.
By this time, however, several plants had become brown, they were devoid of leaflets, and only the pods, the greater portion which had dehisced, remained. Upon the whole, the *C. nictitans* plants were in a better condition on October 1 than those of *C. Chamaecrista*, the majority of the former exhibiting green leaflets, many flowers and a few unripe pods. But, by the middle of October, the *C. nictitans* plants changed greatly. All of the plants turned brown, many leaflets disappeared, and nearly all the pods had dehisced. Nevertheless, within a week or ten days, the plants had almost entirely disappeared. As for the pods, a few very small ones were seen on August 15, and they were present until the end of the growing season. In this case, several pods of natural size appeared toward the end of August, and dehiscence was first noted about September 20.

The most important characteristics shown by each plant during the growing season are given in the following table.
Table No. 6. — Comparison of the Maryland Sennas During Their Growing Season (1933)

<table>
<thead>
<tr>
<th>Name</th>
<th>Growing Season (Start)</th>
<th>First Flower Buds</th>
<th>First Flowers</th>
<th>Height of Flowering Season</th>
<th>Time of Last Flowers</th>
<th>First young Pods</th>
<th>Mature Pods</th>
<th>Growing Season ends</th>
</tr>
</thead>
<tbody>
<tr>
<td>C. Marylandica</td>
<td>First week in May</td>
<td>July 7</td>
<td>July 18</td>
<td>August 4</td>
<td>August 13</td>
<td>July 23</td>
<td>August 13</td>
<td>About the middle of Oct</td>
</tr>
<tr>
<td>C. Medsgeri</td>
<td>First week in May</td>
<td>June 29</td>
<td>July 10</td>
<td>July 24</td>
<td>August 2</td>
<td>July 15</td>
<td>August 2</td>
<td>About the middle of Oct</td>
</tr>
<tr>
<td>C. Chamaecrista</td>
<td>First week in May</td>
<td>July 17</td>
<td>July 27</td>
<td>August 15</td>
<td>Flowers present until the end of the growing season</td>
<td>August 10</td>
<td>August 18</td>
<td>About the middle of Oct</td>
</tr>
<tr>
<td>C. nic-titans</td>
<td>First week in May</td>
<td>July 19</td>
<td>August 1</td>
<td>August 25</td>
<td>Flowers present until the end of the growing season</td>
<td>August 15</td>
<td>August 26</td>
<td>About the middle of Oct</td>
</tr>
</tbody>
</table>
From the descriptions and Table No. 6, it is readily seen that,

(1) The Maryland sennas begin growing at approximately the same time.

(2) The roots of C. Marilandica and C. Medsgeri are perennials, while those of C. Chamaecrista and C. nictitans are annuals.

(3) C. Medsgeri is about a week to ten days ahead of C. Marilandica in its growth.

(4) All the flowers of C. Marilandica and C. Medsgeri have disappeared by the middle of August. The flowers of C. Chamaecrista and C. nictitans do not disappear during the summer, and many plants bear flowers until the end of the growing season.

(5) The plants of C. Chamaecrista and C. nictitans produce mature flowers and pods about two weeks later than C. Medsgeri and C. Marilandica plants.

(6) The Maryland sennas grow to different heights; thus C. Marilandica and C. Medsgeri attain a height of about 4½ feet, C. Chamaecrista when full grown is about two feet high, while C. nictitans, the smallest, never exceeds one foot in height.
B. Microscopic Studies

This portion of the work includes the study of various differing characters as the epidermal cells and the venations of petals, the vein islets of sepals, petals and stipules, the palisade ratio of the leaflets and the pollen grains. Although the various organs with the exception of pollen grains were collected from about the same part of the plants (middle) and were placed in the chloral hydrate reagent at approximately the same time (midday), nevertheless it is possible for these characters to fluctuate, as the Maryland senna plants were not grown under the same biological conditions. That these conditions are imperative in studying leaf anatomy has been shown previously by various workers. Zalenski, as a result of his work in 1904 on the anatomical differences in leaves inserted at different levels on the stem, formulated the "Zalenski law" which states that "the anatomical structure of the individual leaves of one and the same shoot is, so to speak, a function of their distance from the root system." Yapp, in 1912, came to the same conclusion as Zalenski, namely that "the upper leaves of
a plant are more xeromorphic in structure than the lower ones." Hence,\textsuperscript{19} and Rippel\textsuperscript{20} also confirmed Zalenski's results. Also it has been shown that leaf structure is definitely influenced by light, by environmental moisture (atmospheric and soil) and by periodic wilting.\textsuperscript{21} As a result, Zalenski, Yapp, Rippel, Alexandrov and many other investigators have come to the conclusion that "all influences which result in a greatly increased loss of water by a plant, or a restricted supply of water to the developing leaves, lead to essentially similar changes of leaf structure."

Nevertheless in these microscopical studies, results have been obtained in many instances which differed sufficiently to be of pharmacognostical value.

1. Sepals
   a. Distribution of hairs

   The hairs present on the Maryland senna sepals exhibit characteristics which differ to such a degree that it is possible first to separate the sennas into two groups and finally into their four respective species.
Although both species have only marginal hairs which are one-celled and papilllose (Fig. 29), nevertheless the distribution of the hairs plays an important role in separating them. Thus, C. Marilandica always shows hairs present only along the margin of the upper half of the sepal, whereas the hairs of C. Medsgeri extend along the entire periphery of the sepal. During this study, one C. Marilandica sepal showed large hairs present on the lower surface, and this was the only sepal of the several examined which did not exhibit the upper and lower epidermises devoid of hairs.

In the case of C. Chamaecrista and C. nictitans, the hairs are distributed differently from those of C. Marilandica and C. Medsgeri. Marginal hairs are absent but in each case multicellular non-papilllose hairs are present on the lower epidermis in the neighborhood of the midrib (Fig. 30). Furthermore, the upper epidermis is devoid of hairs as well as are the apices and the margins of the sepals. However, hairs may be found occasionally on the lower epidermis, a short distance from the margin, but the majority are found
along the midrib where they increase somewhat in number towards the base of the sepal. It is not difficult to differentiate between C. Chamaecrista and C. nictitans sepals, the former exhibiting the larger hairs which are two- to four-celled, while the latter possess hairs made up of from two to eight cells. They are lanceolate shaped and each sepal possesses an acuminate apex.

The results obtained from the study of the hairs on the Maryland senna sepals are given in the following chart.

```
Trichomes of Senna Sepals

- Minutely papillose, unicellular and marginal
  - Hairs restricted to upper half of sepal. Average length of hairs 147.2\mu Largest hair 197.9\mu—C. Marilandica.

- Hairs along entire periphery of sepal, Average length of hair 85.2\mu Largest hair 149.1\mu—C. Medsgeri.

- Not papillose, Hairs in neighborhood of midrib and multicellular
  - Hairs 2 to 4 celled, usually 3 celled. Average length of hairs 554\mu Largest hair 775.6\mu—C. Chamaecrista.

- Hairs 2 to 8 celled, usually 4 celled. Average length of hairs 395.75\mu Largest hair 552.4\mu—C. nictitans.
```
Fig. 29 — Marginal Hairs Near Apex of C. Mar­ilandica Sepal

Fig. 30 — Hairs along Midrib of C. Cha­maecrista Sepal
b. Stomata of the upper and lower epidermises.

A study of the sepals of Maryland senna flowers revealed that in every case, the lower and upper epidermises showed the majority of the stomata in the neighborhood of the midrib. The C. Chamaecrista and C. nictitans sepals differ from those of C. Marilandica and C. Medsgeri, since the former show the greatest number of stomata along the midribs near the apices; the stomata are generally absent near the margin, although occasionally a stoma may be found as close as six epidermal cells from the margin; and further, very few stomata are found at the base of these sepals. On the contrary, C. Marilandica and C. Medsgeri sepals exhibit the greatest number of stomata in the central portion of the sepal in the neighborhood of the midrib. Also, in the latter plants, very few stomata are found near the margins and at the bases of the sepals. Although, in any species, it is possible to obtain occasionally a sepal whose upper and lower epidermises show stomata distributed somewhat uniformly, nevertheless all species, as a whole, show the majority of the stomata restricted to the neighborhood of the midrib.
In Table No. 7 the stomata of the sepals are compared.

**Table No. 7 - Stomata of the Maryland Senna Sepals.**

<table>
<thead>
<tr>
<th>Species</th>
<th>Number of stomata measured</th>
<th>Sepals collected</th>
<th>Average size of guard cells (microns), U.E.</th>
<th>Average size of guard cells (microns), L.E.</th>
<th>Ratio of short diameter to shorter larger, U.E. dia. to larger L.E.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cassia Marilandica</td>
<td>200</td>
<td>Midday</td>
<td>20.41 x 26.00</td>
<td>22.54 x 26.60</td>
<td>0.785 to 1</td>
</tr>
<tr>
<td>C. Medsgeri</td>
<td>200</td>
<td>Midday</td>
<td>24.85 x 33.01</td>
<td>25.91 x 33.19</td>
<td>0.753 to 1</td>
</tr>
<tr>
<td>C. Chamaecrista</td>
<td>300</td>
<td>Midday</td>
<td>13.85 x 20.59</td>
<td>14.2 x 23.43</td>
<td>0.672 to 1</td>
</tr>
<tr>
<td>C. nictitans</td>
<td>200</td>
<td>Midday</td>
<td>14.33 x 18.1</td>
<td>14.58 x 20.94</td>
<td>0.79 to 1</td>
</tr>
</tbody>
</table>

Conclusions:

(1) The stomata of C. Medsgeri and C. Marilandica sepals are larger than those of C. Chamaecrista and C. nictitans.

(2) The foregoing is a significant characteristic in identifying the Maryland sennas.

c. Neighboring cells.

The number of neighboring cells which accom-
panied each stoma were studied, and the results are
given in Table No. 8.

Table No. 8 -- Comparison of the Neighboring
Cells (N.C.)

<table>
<thead>
<tr>
<th>Species</th>
<th>Number of stomata examined</th>
<th>2 N.C.</th>
<th>3 N.C.</th>
<th>4 N.C.</th>
<th>5 N.C.</th>
</tr>
</thead>
<tbody>
<tr>
<td>C. Marilandica</td>
<td>100</td>
<td>56</td>
<td>32</td>
<td>12</td>
<td></td>
</tr>
<tr>
<td>C. Medsgeri</td>
<td>150</td>
<td>89</td>
<td>43</td>
<td>14</td>
<td>4</td>
</tr>
<tr>
<td>C. Chamaecrista</td>
<td>200</td>
<td>200</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>C. nictitans</td>
<td>200</td>
<td>174</td>
<td>23</td>
<td>3</td>
<td></td>
</tr>
</tbody>
</table>

From this table it may be concluded that:

(1) It is possible to separate the four species
into two main groups, as C. Marilandica and C. Medsgeri
sepals show a greater number of stomata accompanied by three
and four neighboring cells.
d. Epidermal cells.

The larger epidermal cells appeared on C. Medsgeri and C. Marilandica sepals. The cells which comprised the upper epidermis of the former sepal were the largest of those studied. Both species showed the upper epidermal cells larger than those on the lower epidermis, and in the case of C. Marilandica the upper epidermal cells were also slightly more undulate. (Figs. 31-34)

The upper epidermal cells of C. nictitans sepals were the smallest studied and appear very undulate (Fig. 37); while the lower epidermal cells show very little sinuosity and are somewhat larger. (Fig. 38). The C. Chamaecrista sepal can be recognized by the fact that it has epidermal cells larger than those present on C. nictitans sepals, and also by the undulate upper epidermal cells. Both epidermises of C. Chamaecrista sepals exhibit epidermal cells which differ slightly in size. (Figs. 35, 36)

The following chart shows the sepals can be differentiated by their epidermal cells.
Largest cells, cells with undulate outlines absent — C. Medsgeri sepal.

Large

Cells smaller than those of C. Medsgeri
(14.4-31.9μ wide x 24.8-56.8μ long)
slightly undulate, especially the upper
epidermal cells — C. Marilandica sepal.

Epidermal Cells
of Sepals

Cells larger than those of C. nictitans
(12.43-24.8μ wide x 35.5-92.3μ long);
upper epidermal cells somewhat undulate —
C. Chamaecrista sepal.

Small

Smallest cells (12.4-21.3μ wide x 31.9-67.4μ long); upper epidermal cells greatly un-
dulate — C. nictitans sepal.

e. Vein islet number of sepals.

By means of the vein islets (page 66), it is possible to separate the sepals into two groups. Table No. 9 gives the results obtained in this study.
Table No. 9. — Vein Islet Numbers of Sepals

<table>
<thead>
<tr>
<th>Species</th>
<th>Vein islet number</th>
<th>Remarks</th>
</tr>
</thead>
<tbody>
<tr>
<td>C. Medsgeri</td>
<td>8.5</td>
<td></td>
</tr>
<tr>
<td>C. Marilandica</td>
<td>8.0</td>
<td></td>
</tr>
<tr>
<td>C. Chamaecrista</td>
<td>11.5</td>
<td></td>
</tr>
<tr>
<td>C. nictitans</td>
<td></td>
<td>Vein islet number undetermined. Under low power, sepal was too narrow to cover the entire field.</td>
</tr>
</tbody>
</table>

Figures 39 -- 42 show the vein islets of the sepal.
Fig. 31 — Upper Epidermis of C. Marilandica Sepal

Fig. 32 — Lower Epidermis of C. Marilandica Sepal
Fig. 33 — Upper Epidermis of C. Medsgeri Sepal

Fig. 34 — Lower Epidermis of C. Medsgeri Sepal
Fig. 35 — Upper Epidermis of C. Chamaecrista Sepal

Fig. 36 — Lower Epidermis of C. Chamaecrista Sepal
Fig. 37 — Upper Epidermis of C. nictitans Sepal

Fig. 38 — Lower Epidermis of C. nictitans Sepal
Fig. 39 — Vein Islets of C. Marilandica Sepal

Fig. 40 — Vein Islets of C. Medsgeri Sepal
**Fig. 41** — Vein Islets of C. Chamaecrista Sepal

**Fig. 42** — Vein Islets of C. nictitans Sepal
2. Corollas

a. Preparation.

During the summer I picked flowers from the various plants and placed them immediately in a strong solution of chloral hydrate (5-2). The specimens remained in this clearing agent at room temperature for about five months, and at this time the petals were cleared so well that a comparative study could be made. The petals were separated from the flowers and were mounted on a slide in a few drops of the clearing solution. Finally a cover slip was applied and the specimen was examined under the microscope at a magnification of 450.

b. Epidermal cells

An examination of the petals revealed that all petals showed epidermal cells with undulate outlines for the most part. Also, the size of the epidermal cells is practically of no diagnostic value in the case of C. Marylandica, C. Medsgeri and C. Chamaecrista as is shown in the following table. (Figs. 43 - 45)
Table No. 10 Epidermal Cells of Maryland Senna Flowers.

<table>
<thead>
<tr>
<th>Species</th>
<th>Width of Epidermal Cells</th>
<th>Length of Epidermal Cells</th>
</tr>
</thead>
<tbody>
<tr>
<td>C. Marylandica</td>
<td>26.63 to 42.6 microns</td>
<td>35.5 to 71 microns</td>
</tr>
<tr>
<td>C. Medsgeri</td>
<td>21.3 to 35.5 microns</td>
<td>35.5 to 60.4 microns</td>
</tr>
<tr>
<td>C. Chamaecrista</td>
<td>21.3 to 32 microns</td>
<td>30.18 to 81 microns</td>
</tr>
<tr>
<td>C. nictitans</td>
<td>12.43 to 28.4 microns</td>
<td>28.4 to 49.7 microns</td>
</tr>
</tbody>
</table>

On the other hand, it is possible that the epidermal cells of C. nictitans petals can be recognized by their smaller size. (Fig. 46).

c. Marginal venations.

If the venations of the Maryland senna petals are studied, the veins appear to leave the midrib in a somewhat pinnate fashion, and after traversing the petals they usually form terminal branches a short distance from the margin.
This tendency to produce terminal branches is most pronounced at and near the apices of the petals.

An examination of the apices of the petals (Figs. 47—50) shows that the terminal branches of the veinlets near the margins differ sufficiently in size to be employed in the identification of each petal. C. Marilandica and C. Medsgeri petals show the medium-sized terminal branches; the C. Marilandica petal may be differentiated from that of C. Medsgeri, as the former exhibits the smaller-sized terminal endings. C. Chamaecrista petal, on the other hand, has terminal branches of veinlets which are long and narrow and the largest of all. The C. nictitans petal is characteristic in that many small terminal branches are present near the margin.

The following chart shows how the four petals may be differentiated.
Terminal branches 237.4\(\mu\) - 775.6\(\mu\)--
C. Marilandica.

Terminal branches 395\(\mu\) - 1380\(\mu\)--
C. Medsgeri

Terminal branches long (601.5\(\mu\) - 2137\(\mu\))
and narrow -- C. Chamaecrista petal

Terminal branches 156.3\(\mu\) - 612.4\(\mu\)--
C. nictitans.
Fig. 43 — Epidermal Cells of C. Marilandica Petal

Fig. 44 — Epidermal Cells of C. Medsgeri Petal

Fig. 45 — Epidermal Cells of C. Chamaecrista Petal

Fig. 46 — Epidermal Cells of C. nictitans Petal
Fig. 47 — C. Marilandica Petal (Apex)

Fig. 48 — C. Medsgeri Petal (Apex)
Fig. 49 — C. Chamaecrista Petal (Apex)

Fig. 50 — C. nictitans Petal (Apex)
3. Pollen

A study was made of the pollen grains to determine whether they differed sufficiently in their physical characteristics to be of any value in the identification of the four senna species. An examination of the four pollens revealed that they were similar in that all of the grains were elliptical and practically of same size; and each grain is divided into two almost-equal portions by a pronounced longitudinal ridge as the result of tetrad division. (Fig. 51)
The average sizes of the pollen grains are given in Table No. 11.

Table No. 11 -- Pollen Grains

<table>
<thead>
<tr>
<th>Pollen</th>
<th>Number of pollen grains examined</th>
<th>Average size of pollen grains in microns</th>
<th>Ratio of width to length</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Width</td>
<td>Length</td>
</tr>
<tr>
<td>C. Medsgeri</td>
<td>50</td>
<td>25.66</td>
<td>39.42</td>
</tr>
<tr>
<td>C. Marilandica</td>
<td>50</td>
<td>24.41</td>
<td>40.22</td>
</tr>
<tr>
<td>C. Chamaecrista</td>
<td>50</td>
<td>21.64</td>
<td>39.94</td>
</tr>
<tr>
<td>C. nictitans</td>
<td>50</td>
<td>22.84</td>
<td>37.03</td>
</tr>
</tbody>
</table>
4. Vein Islets

Zufall and Burlage\textsuperscript{22} state: "If one traces the vein of a leaf as it dwindles in size, he finds that it becomes too small to be seen with the naked eye and finally as he views it under the microscope he finds that it is composed of only one or two vessels which connect with similar branches of other veins." For the minute leaf area included within the meshes of the veinlets Benedict\textsuperscript{23} coined the term "vein islet."

Zalenski\textsuperscript{24} was the first to show that a mathematical relationship existed between venation and leaf surfaces, and from his work he concluded that: "In different plants, grown under exactly the same biological conditions, the total length of veinlets per unit area of leaf surface is a constant which shows only the slightest variation. This figure bears no relation to the systematic position of the plant." Shuster\textsuperscript{25}, in 1908, published a paper in which he confirmed Zalenski's work. Later Benedict in comparing many leaves used the area of the vein islet instead of Zalenski's method, in which "the com-
bined length of the veins in 1 square millimetre of leaf surface" was measured. Benedict used an enlarging camera to photograph leaves at an enlargement of three diameters and counted the number of vein islets. Ensign reviewed Benedict's work and found it inaccurate because the chlorophyll in uncleared leaves hides from 17 to 62 per cent of the vein islets. As a result, Ensign made the following conclusion: "The above data show that any study of leaf venation made from uncleared leaves is wholly unreliable. The varying thickness and chlorophyll content of leaves render many of the smaller veinlets entirely invisible." Benedict also showed that the venations of leaves obtained from young and old plants of the same species differed, and concluded that "while the size of the vein islets always diminished with age, the actual size of the islets at any age was a specific character, relatively constant in each species." However, Ensign studied the vein islets of cleared leaves of grape and grape fruit and showed Benedict's methods and results to be inaccurate and unreliable. Levin, in 1929, made a thorough study of several species of drugs
belonging to the Barosma, Cassia, Erythroxylon and Digitalis genera in order to determine the pharmacognostical value of the vein islet areas in distinguishing between drugs of closely related species. In 1932, Zufall and Burlage published their work, in which vein islets were employed to differentiate between the leaves of Atropa Belladonna and Phytolacca decandra; Stramonium and Hyoscyamus; and Mentha piperita and Mentha spicata.

Method of Determining the Average Area of the Vein Islets.

Levin used a modification of Ensign's method to calculate the average vein islet area. Levin's method was as follows: "At a definite magnification the image of the leaf was projected by means of a series of prisms on to a horizontally placed sheet of paper, upon which a rectangular area representing 4 square millimetres of leaf surface had been traced. The islets which fell within this area were counted, and from this figure the average area of one islet was determined." Levin also found that "the results obtained show that the vein islets are most constant in the
central portion of the lamina, excluding the regions near the midrib and margin." He stated further that "all the counts recorded, therefore were made in the central portion of the lamina, excluding the midrib and margin."

Zufall and Burlage used the method of Levin in a modified form to determine the average area of the vein islet. Their method is: "Two pieces of the leaf were obtained in the following way: Each end of the leaf was cut off at a point one-fourth of the distance from tip to base and discarded; the piece left was cut down the midrib and from each side a piece was obtained by cutting away one-fourth of the outer edge and one-fourth next to the midrib. These areas were used because in them the vein islets were found to be more nearly uniform in size. The vein islets next to the midrib are larger than in the other regions, while in the margins of some leaves no vein islets are found." They determined the vein islet area by this method: "The area of the vein islet may be determined by counting the number of vein islets within the field of the 16 mm. objective. The number of vein islets in the field divided by the area of the field in
millimetres gives the area of the vein islet."

The Maryland and the official senna leaflets showed vein islets arranged according to the way Levin described previously and, as a result, only the vein islets in the central portion of each leaflet were examined. Since the leaflets were of such size that they could be studied conveniently under the microscope, a modification of the Zufall-Burlage method was used to determine the vein islet areas. In each case the leaflet was removed from the clearing solution and mounted on a slide in glycerin. Then, by means of a mechanical stage, a field was obtained which represented the mid-points between the apex and the base, and the midrib and the margin of the leaflet, respectively. In this method, only two determinations were made on each C. Marilandica and C. Medsgeri leaflet. The two positions are shown by the small crosses in Figure 7. With C. Marilandica and C. Medsgeri it is possible to make two determinations on each leaflet as, in each case, the midrib divides the leaflet into two portions which exhibit approximately the same size and physical characteristics. However, the C. Chamaecrista and C. nictitans leaflets show
the midrib dividing each leaflet into two portions of unequal size. Therefore, in these leaflets, the vein islet areas were determined only on the larger portion and as a result, each leaflet gave rise to one determination (Figure 9). The average areas of the vein islets of the senna leaflets are given in Table 12.

Vein islet numbers.

Levin was the first to use the term "vein islet number" to represent "the number of vein islets per square millimetre." Levin's method for obtaining the vein islet number has been given. Zufall and Burlage obtained their vein islet number by using "a microscope whose 16 mm. objective gave a field whose area was 2.01 square millimetres." They further stated that this field was approximately 2 square millimetres in area. In this method it is necessary only to determine the average number of vein islets per field, as "this number divided by 2 (the area of our field was 2 square millimetres) gives the number of vein islets per square millimetre or the "vein-islet number."
The vein islet numbers of the six senna leaflets were determined by a modification of the Zufall-Burlage method (page 86).

The following table gives the results obtained in the study of the vein islets of the official and the Maryland senna leaflets.

Table No. 12 — Vein Islet Numbers

<table>
<thead>
<tr>
<th>Specie</th>
<th>Number of leaflets examined</th>
<th>Average Vein Islet Area in sq. mm.</th>
<th>Vein Islet numbers</th>
<th>Average Vein Islet number</th>
<th>Type of Vein Islets</th>
</tr>
</thead>
<tbody>
<tr>
<td>C. Senna</td>
<td>14</td>
<td>0.035</td>
<td>25 - 31</td>
<td>28.3</td>
<td>Definite</td>
</tr>
<tr>
<td>C. angustifolia</td>
<td>10</td>
<td>0.047</td>
<td>19 - 23.5</td>
<td>21.5</td>
<td>Definite</td>
</tr>
<tr>
<td>C. Medsgeri</td>
<td>15</td>
<td>0.111</td>
<td>6 - 13</td>
<td>9.0</td>
<td>Definite</td>
</tr>
<tr>
<td>C. Marylandica</td>
<td>16</td>
<td>0.045</td>
<td>20 - 25</td>
<td>22.25</td>
<td>Definite</td>
</tr>
<tr>
<td>C. Chamaecrista</td>
<td>12</td>
<td>0.036</td>
<td>24 - 32</td>
<td>27.5</td>
<td>Reduced</td>
</tr>
<tr>
<td>C. nictitans</td>
<td>14</td>
<td>0.055</td>
<td>15.5 - 23</td>
<td>18.1</td>
<td>Reduced</td>
</tr>
</tbody>
</table>
Table No. 12 shows:

(1) A study of the vein islet numbers of the official senna leaflets showed results very similar to those given by Levin. Levin's statement that "the vein islet numbers for C. acutifolia and C. angustifolia provide a character by which these two types of sennas can be accurately and easily distinguished" was checked and found true.

(2) Making use of the average vein islet area, one can differentiate C. Marilandica from C. Medsgeri, and C. nictitans from C. Chamaecrista.

(3) It is possible to separate the Maryland sennas into their respective species by means of the vein islet numbers.

The vein islets of the official and the Maryland senna leaflets are shown in Figures 52 to 57.
Fig. 52 — Vein Islets of Cassia Senna Leaflet

Fig. 53 — Vein Islets of C. angustifolia Leaflet
Fig. 54 — Vein Islets of C. Marilandica Leaflet

Fig. 55 — Vein Islets of C. Medsgeri Leaflet
Fig. 56 — Vein Islets of C. Chamaecrista Leaflet

Fig. 57 — Vein Islets of C. nictitans Leaflet
5. Palisade Ratio

Zornig and Weiss in their anatomical study of the leaves of certain composite plants found that the average number of palisade cells beneath an upper epidermal cell was of diagnostic value. They also stated that the ratio of the number of palisade cells included within the area of any upper epidermal cell remained almost constant. Wallis and Dewar later made a thorough study of the comparative anatomy of leaves belonging to several species of Barosma, and they also concluded "that variation in the palisade ratio is independent of the position on the leaf." Furthermore, they first used the term "palisade ratio," stating in their paper that "the total number of palisade cells beneath four upper epidermal cells when divided by four, gave the recorded figure for the "palisade ratio", this being the term which we have adopted to express the average number of palisade cells beneath an upper epidermal cell."

In this work, the palisade ratio was obtained
by examining various upper epidermal cells regardless of their positions on the leaflets. The term "palisade ratio" as used in this work differs somewhat from the definition given previously before by Wallis and Dewar. Only one epidermal cell was studied instead of four, and the number of palisade cells under any individual epidermal cell was determined. All determinations were made on cells which were separated by at least two epidermal cells; never on adjacent cells. Throughout this work the term will therefore designate the average number of palisade cells, both entire and fractions thereof, that are included beneath an upper or lower epidermal cell, as the case may be. For example, the number of palisade cells beneath the epidermal cell A, in Figure 67 was determined to be 20.

The palisade ratios of six senna leaflets were studied; namely, those of C. Senna, C. angustifolia and the four Maryland sennas, (Figs. 58--63 ). Since the official senna leaflets exhibit the palisade tissue adjacent to both the upper and lower epidermises, it was therefore necessary to determine the palisade ratios for the upper and lower epidermal cells of these leaflets.
The results obtained from this study are as follows:

**Table No. 13 -- Palisade Ratio**

<table>
<thead>
<tr>
<th>Species</th>
<th>Source of Leaflets</th>
<th>Number of leaflets examined</th>
<th>Number of determinations made</th>
<th>Range of Palisade Ratio</th>
<th>Diameter of Palisade Cells</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cassia Senna</td>
<td>Leaflets obtained from stock and placed in a saturated solution of chloral hydrate (5-2) and boiled until cleared.</td>
<td>3</td>
<td>50</td>
<td>5 - 23</td>
<td>7.1 - 15.97</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>average 12.4</td>
<td>average 11.4</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>3 - 13</td>
<td>8 - 24.85</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>average 7.1</td>
<td>average 14.48</td>
</tr>
<tr>
<td>Cassia angustifolia</td>
<td></td>
<td>3</td>
<td>50</td>
<td>5 - 17</td>
<td>7.1 - 21.3</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>average 10.0</td>
<td>average 13</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>3 - 12</td>
<td>7.1 - 19.53</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>average 7.0</td>
<td>average 13.1</td>
</tr>
<tr>
<td>Cassia Medagerti</td>
<td>Fresh leaflets gathered August, 1933 and placed immediately in a saturated solution of chloral hydrate until November, 1933.</td>
<td>3</td>
<td>25</td>
<td>6 - 14</td>
<td>6.9 - 19.53</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>average 9.1</td>
<td>average 13.3</td>
</tr>
<tr>
<td>Cassia Marylandica</td>
<td>Fresh leaflets gathered August, 1933 and placed immediately in a saturated solution of chloral hydrate until November, 1933.</td>
<td>3</td>
<td>25</td>
<td>6 - 13</td>
<td>6.9 - 17.75</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>average 8.1</td>
<td>average 12</td>
</tr>
<tr>
<td>Cassia Chamaecrista</td>
<td>Leaflets collected Sept. 1933 and dried at room temperature, they were cleared in chloral-phenol by boiling</td>
<td>3</td>
<td>25</td>
<td>12 - 23</td>
<td>6.2 - 16</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Average 18.4</td>
<td>average 10.3</td>
</tr>
<tr>
<td>Cassia nictitans</td>
<td></td>
<td>3</td>
<td>25</td>
<td>10 - 16</td>
<td>7.1 - 15.1</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>average 12.5</td>
<td>average 9.9</td>
</tr>
</tbody>
</table>
From Table No. 13 it may be concluded that:

(1) In the official senna leaflets, the upper epidermis in each case is easily differentiated from the lower epidermis; it is impossible to differentiate between the lower epidermises of the official senna leaflets; and finally, it is possible to identify each species of the official senna leaflets by means of the palisade ratios of the upper epidermises.

(2) The Maryland sennas can be separated into two groups - C. Marilandica, C. Medmgeri; and C. Chamaecrista, C. nictitans.

(3) It is possible to differentiate between C. Chamaecrista and C. nictitans leaflets, as the former shows a larger palisade ratio. Furthermore, C. nictitans has the smallest palisade cells.

(4) The official senna leaflets show two rows of palisade tissue present. The Maryland senna leaflets have only one row of palisade tissue which is adjacent to the upper epidermis.
Fig. 58 — Palisade Cells of Cassia Senna Leaflet (Upper Epidermis)

Fig. 59 — Palisade Cells of Cassia angustifolia Leaflet (Lower Epidermis)

Fig. 60 — Palisade Cells of C. Medsgeri Leaflet

Fig. 61 — Palisade Cells of C. Marilandica Leaflet

Fig. 62 — Palisade Cells of C. Chamaecrista Leaflet

Fig. 63 — Palisade Cells of C. nictitans Leaflet
6. Stipules

a. Distribution of hairs

Although the stipules of the Maryland sennas show their upper and lower epidermises devoid of trichomes; nevertheless, marginal hairs are always present. C. Marilandica stipules have marginal hairs that are straight or slightly curved and distributed rather uniformly along both margins from the base to the apex of the stipule. Again, the hairs are very long, and many of them occur approximately at right angles to the margins (Fig. 64). C. Medsgeri stipules, on the other hand, often exhibit curved marginal hairs whose apices point towards the midrib of the stipule (Fig. 65). In this case the hairs are arranged irregularly along the margins and usually one margin shows itself almost devoid of trichomes. However, with C. Chamaecrista and C. nictitans stipules the hairs are distributed uniformly along both margins from a point just below the apex to the base (Fig. 66). The smallest hairs occur near the
apex of the stipule and gradually increase in length, so that the largest are found at the base.

The stipules of *C. Marilandica* and *C. Medgeri* have unicellular and minutely papillose marginal hairs, whereas the two remaining stipules always show multicellular hairs which are not papillose, or only slightly so.

The characteristics exhibited by the marginal hairs of the stipules are given in the following chart:

| Hairs large, unicellular and minutely papillose | Hairs usually straight and at right angles to margins. Uniformly distributed along both margins. Average size of hairs 761.4 μ, largest hair 1100 μ — *C. Marilandica*. |
| Marginal trichomes of Maryland senna stipules | Hairs occasionally curved, with their tips pointing towards the midrib. Not uniformly distributed along both margins. Average size of hairs 225 μ, largest hair 319.5 μ long — *C. Medgeri*. |
| Hairs small, multicellular, not papillose, or slightly so. | Hairs 2 to 4 celled, usually 2 celled, Average size of hairs 149.8 μ, largest hair 195.3 μ — *C. Chamaecrista*. |
| | Hairs 2 to 4 celled, usually 3 celled, Average size of hairs 124.7 μ, largest hair 177.5 μ — *C. nictitans*. |
Fig. 64 — Marginal Hairs of C. Marilandica Stipule

Fig. 65 — Marginal Hairs of C. Medsgeri Stipule

Fig. 66 — Marginal Hairs of C. Chamaecrista Stipule
This chart shows that it is possible by means of the trichomes present on the stipules to separate the Maryland sennas into two groups (C. Marilandica, C. Medsgeri and C. Chamaecrista, C. nictitans) and further to separate them into, and to identify, the respective species.

b. Stomata of the upper and lower epidermises.

The upper and lower epidermises of the stipules exhibit characteristics which differ to such an extent that the species may be separated either into the two groups previously mentioned, or finally into their individual species. For example, the stomata on the upper and lower epidermises of C. Chamaecrista and C. nictitans stipules are distributed almost uniformly (Figs. 71-74) whereas the stipules of C. Marilandica and C. Medsgeri have stomata on the upper epidermises restricted to the neighborhood of the midrib; the lower epidermises have the stomata distributed more uniformly (Figs. 67-70).

The results obtained from a study of the stomata of the stipules are given in the following table.
Table No. 14 -- Comparison of the Stomata of the Maryland Senna Stipules

<table>
<thead>
<tr>
<th>Species</th>
<th>Stomata per sq.mm. U.E.</th>
<th>Stomata per sq.mm. L.E.</th>
<th>Average size of guard cells (microns) U.E.</th>
<th>Average size of guard cells (microns) L.E.</th>
<th>Ratio of shorter diam. to larger U.E.</th>
<th>Ratio of shorter diam. to larger L.E.</th>
</tr>
</thead>
<tbody>
<tr>
<td>C. Marylandica</td>
<td>---</td>
<td>80.63</td>
<td>23.61 x 33.55</td>
<td>25.98 x 34.15</td>
<td>.704 to 1</td>
<td>.758 to 1</td>
</tr>
<tr>
<td>C. Medsgeri</td>
<td>---</td>
<td>76</td>
<td>18.99 x 28.20</td>
<td>20.94 x 29.09</td>
<td>.673 to 1</td>
<td>.72 to 1</td>
</tr>
<tr>
<td>C. Chamaecrista</td>
<td>131.65</td>
<td>165.53</td>
<td>17.40 x 20.23</td>
<td>17.57 x 20.77</td>
<td>.86 to 1</td>
<td>.846 to 1</td>
</tr>
<tr>
<td>C. nictitans</td>
<td>93.9</td>
<td>100.67</td>
<td>15.69 x 19.95</td>
<td>16.93 x 20.41</td>
<td>.787 to 1</td>
<td>.829 to 1</td>
</tr>
</tbody>
</table>

Since stomata are not distributed uniformly upon the upper epidermises of C. Marylandica and C. Medsgeri stipules, therefore the number of stomata per square millimetre for these surfaces was not included in Table 14. Nevertheless, in both stipules, twenty guard cells on the upper epidermis were measured and the results appear in the fourth column.
An examination of Table No. 14 shows that:

(1) The senna species can be separated into two groups since the stomata are distributed differently upon the epidermises of the stipules.

(2) The largest stomata are found on the *C. Marilandica* stipules, the smallest on *C. nictitans* stipules.

(3) In all species, the smaller stomata are found upon the upper epidermis of the stipule.

Throughout the study of the sepals and stipules the terms upper epidermis and lower epidermis were used to designate the adaxil and abaxil surfaces, respectively.

c. Epidermal cells.

The results obtained from a study of the epidermal cells of the stipules are shown in the following chart.
Epidermal cells of Maryland senna stipules

- **Large**
  - (Cell wall not undulate, or slightly so)
  - Largest cells — C. Medsgeri stipule.

- **Small**
  - (Cell wall undulate)
  - Cells smaller than C. Medsgeri, often appearing somewhat square-like — C. Marilandica stipule.

- Outline of cell very undulate, stomata accompanied by 2 to 4 neighboring cells — C. nictitans stipule.

- Outline of cell not so undulate, all stomata accompanied by two neighboring cells — C. Chamæcrista stipule.

During this study, stomata with from two to five neighboring cells were found on the epidermises of C. Marilandica and C. Medsgeri stipules. Thus, the neighboring cells can also play an important role in the identification of the epidermises.
Fig. 67 — Upper Epidermis of C. Marilandica Stipule (near midrib)

Fig. 68 — Lower Epidermis of C. Marilandica Stipule
Fig. 69 — Upper Epidermis of C. Medsgeri Stipule (near margin)

Fig. 70 — Lower Epidermis of C. Medsgeri Stipule
Fig. 71 — Upper Epidermis of C. Chamaecrista Stipule

Fig. 72 — Lower Epidermis of C. Chamaecrista Stipule
Fig. 73 — Upper Epidermis of C. nictitans Stipule

Fig. 74 — Lower Epidermis of C. nictitans Stipule
V. Summary of the characters examined upon which the separation of the Maryland sennas into two groups is based.

A. **Genetical characters** (stable, not subject to ecological modifications)

<table>
<thead>
<tr>
<th>C. <em>Marylandica</em> and C. <em>Medsgeri</em></th>
<th>C. <em>Chaamaecrista</em> and C. <em>nictitans</em></th>
</tr>
</thead>
<tbody>
<tr>
<td>2. Pods large and curved.</td>
<td>2. Pods small and straight.</td>
</tr>
<tr>
<td>3. Greater number of segments and seeds per pod.</td>
<td>3. Smaller number of segments and seeds per pod.</td>
</tr>
<tr>
<td>4. Flower buds in short axillary racemes at the top of the plants. Large, elliptical buds.</td>
<td>4. Flower buds in small clusters above the axils and occasionally in the axils. Small, conical buds.</td>
</tr>
<tr>
<td>5. Larger seeds. Seeds brown or olive, not shiny and elliptical or fan-shaped.</td>
<td>5. Smaller seeds. Seeds black, shiny and rectangular.</td>
</tr>
<tr>
<td>6. Testa exhibits large depressions which correspond to the position of the embryo.</td>
<td>6. Testa is pitted, and pits are arranged in longitudinal rows. No large depressions present.</td>
</tr>
<tr>
<td>8. Terminal branches of veinlets anastomose near the margins of leaflets.</td>
<td>8. Terminal branches of veinlets end abruptly near the margins. Veinlets do not anastomose.</td>
</tr>
<tr>
<td>9. Leaflets divided into two nearly equal parts by the midrib</td>
<td>9. Midrib divides leaflets into two unequal portions.</td>
</tr>
<tr>
<td>10. Plants show one main stem.</td>
<td>10. Plants branch close to base.</td>
</tr>
<tr>
<td>11. Large, conical or club-shaped glands.</td>
<td>11. Small, cup-shaped glands.</td>
</tr>
</tbody>
</table>
13. Stipules green and of spatulate or lanceolate shape.


15. Hairs of large size present along the margins of the leaflets.

16. Marginal hairs of stipules large, unicellular and minutely papillose.

17. Sepals exhibit marginal hairs which are minutely papillose and unicellular.

18. Mucrons of the leaflets taper only slightly.

19. Mucrons of stipules terminate in short, acute points.

20. Large leaflets.

13. Stipules of triangular shape. They are green with red margins and tips.

14. Sepals lanceolate, with acuminate apices.

15. Hairs absent from the margins of the leaflets. Margins microscopically serrate.

16. Marginal hairs of stipules small, multicellular and not papillose, or slightly so.

17. Hairs are found only in the neighborhood of the midrib of the sepal. Hairs are not papillose and are multicellular.

18. Mucrons of leaflets taper to a sharp, slender point.

19. Mucrons of stipules terminate by gradually-tapering sharp points.

20. Small leaflets.
B. Ecological characters. (subject to ecological modifications).

<table>
<thead>
<tr>
<th>C. Marilandica and C. Medsgeri</th>
<th>C. Chamaecrista and C. nictitans</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. Plants found in rich, damp soil.</td>
<td>1. Plants grow in dry, sandy soil.</td>
</tr>
<tr>
<td>2. Flower buds appear the first week in July.</td>
<td>2. Flower buds appear about the middle of July.</td>
</tr>
<tr>
<td>3. Full-grown flowers produced about the middle of July.</td>
<td>3. Full-grown flowers produced about August 1.</td>
</tr>
<tr>
<td>4. Flowers lose their sepals, petals and stamens about August 13 and August 2, respectively.</td>
<td>4. About one-half of the plants exhibited flowers until the end of the growing season.</td>
</tr>
<tr>
<td>5. Plants about four feet high.</td>
<td>5. Plants are from one to two feet in height.</td>
</tr>
<tr>
<td>6. Smaller number of pods per plant.</td>
<td>6. Many pods per plant.</td>
</tr>
<tr>
<td>7. Stomata absent from the upper surfaces of the leaflets. Stomata large and elliptical.</td>
<td>7. Both leaflet surfaces show stomata present. Stomata small and tend to be circular.</td>
</tr>
<tr>
<td>8. Stomata not distributed uniformly upon the upper epidermises of the stipules. Stomata large and elliptical.</td>
<td>8. Stomata distributed rather uniformly upon both surfaces of the stipules. Stomata small and somewhat circular.</td>
</tr>
<tr>
<td>9. Leaflets have smaller palisade ratios.</td>
<td>9. Leaflets have larger palisade ratios.</td>
</tr>
<tr>
<td>10. Stipules have large epidermal cells.</td>
<td>10. Stipules have small epidermal cells.</td>
</tr>
<tr>
<td>11. Large, slightly curved hairs present on the stems.</td>
<td>11. Stems exhibit small, curved or coiled hairs.</td>
</tr>
</tbody>
</table>
12. Petioles large and green.
13. Average diameter of stems 5 mm.
14. Fractured stems show their pith to be either a light or deep orchid color.
15. Glands always green.
16. Pods varying in color from chocolate to black.
17. At the end of the growing season the glands are brittle and many are missing on the petioles.
18. Stems green throughout the growing season.

12. Petioles small, red or tinted red.
13. Average diameter of stems 1.5 mm.
14. Fractured stems always show white or cream colored pith.
15. Glands dark red, or green tinted red.
16. Yellowish green or red pods.
17. Glands always present on the petioles.
18. Stems not green throughout the growing season.
VI. Identification of the Powdered Maryland Senna Leaflets.

If an occasion should arise whereby it would be necessary to identify the powdered Maryland sennas, especially a mixture of all the species, there are several ways in which this could be accomplished.

However, before the examination could be made, several important steps are necessary in the preparation of the powders to be examined. First, if possible, a no. 20 or a no. 40 powder should be used in order that the fragments would be of sufficient size to show all, or a majority of the characteristics of each plant; and second, the specimens should be cleared by treating them with chloral hydrate (5-2), or some other such clearing agent as chloral phenol, lactic acid, or nitric acid. Finally, after clearing, the powder should be mounted on a slide either in the clearing agent used previously or in glycerin, a cover slip applied and the specimen examined under the microscope.

Even if the powdered specimen to be examined should consist of a mixture of the four senna plants, there would be sufficient number of characteristics of each
plant present so that each species could be easily identified.

For simplicity, each organ of the senna plants will be dis-
cussed under a separate heading, and the methods for the iden-
tification of each species will be given. In the following
identification key, only the surface characteristics are
employed to differentiate the four species.

Key for the identification of the powdered official and Mary-
land senna leaflets.

1. Margins.
   a. Serrated.
      1°. Average size of serrations—$61.3\mu$ long —
         C. Chamaecrista.

      2°. Average size of serrations—$64.4\mu$ long —
         C. nictitans.

         (Note: C. nictitans leaflet usually has
         one margin serrated only about 1/5
         of the distance from the base to
         the apex.)

   b. Not serrated.
      1°. Only large hairs always present.
         a°. Marginal hairs up to $933\mu$ in length
         and almost perpendicular to the mar-
         gins — C. Marilandica.
b. Marginal hairs up to 587μ in length and parallel to the margin — C. Medsgeri.

2. Only small hairs present. Hairs point towards the apex.
   a¹. Hairs up to 52μ in length — C. angustifolia.
   b¹. Hairs slightly smaller than (a¹). — C. Senna.

2. Apices.

a. Tapering slightly
   1. Mucrons about three times as long as wide — C. Marilandica.
      2¹. Mucrons about twice as long as wide — C. Medsgeri.

b. Apices tapering to a sharp, slender point.
   1. Apices about three times as long as wide — C. nictitans.
      2¹. Apices about twice as long as wide — C. Chamaecrista.

c. Apices from 1 to 1½ as long as wide, usually as long as wide and of ball or conical shape.
   1¹. Apices usually devoid of hairs — C. angustifolia.
      2¹. Apices usually showing hairs present — C. Senna.
3. Petiolules.

a. Large.

1. Petiolules rarely without hairs, hairs numerous -- *C. Medsgeri*.

2. Petiolules occasionally with hairs,

hairs not so numerous -- *C. Marilandica*.

b. Medium.

1. Hairs numerous on petiolules and along outline of petiolules -- *C. Senna*.

2. Hairs not so numerous on petiolules and along outline of petiolules -- *C. angustifolia*.

c. Small.

1. Either *C. nictitans* or *C. Chamaecrista*.

4. Epidermal hairs.

a. Epidermal hairs are absent in all Maryland senna leaflets.

b. Epidermal hairs present in the official senna leaflets. (50 - 250 μ long).
1. C. angustifolia.
   a. Upper epidermis -- average number of hairs per field -- .06.
   b. Lower epidermis -- average number of hairs per field -- 1.83

2. C. Senna
   a. Upper epidermis -- average number of hairs per field -- 1.72
   b. Lower epidermis -- average number of hairs per field -- 12.1

5. Palisade ratio.
   a. C. Mariandica -- 8.1
   b. C. Medsgeri -- 9.1
   c. C. Chamaecrista -- 18.4
   d. C. nictitans -- 12.5
   e. C. Senna -- Upper epidermis -- 12.4
      Lower epidermis -- 7.1
   f. C. angustifolia -- Upper epidermis -- 10.0
      Lower epidermis -- 7.0
6. Vein islet number

a. C. Marilandica —— 22.25
b. C. Medsgeri ——— 9.0
c. C. Chamaecrista —— 27.5
d. C. nictitans ——— 18.1
e. C. Senna ———— 26.83
f. C. angustifolia —— 20.8
VII. Conclusions.

From this study it is concluded that:

1. The Maryland sennas exhibit genetical and ecological characters which differ sufficiently to warrant the separation of the four species into two groups: Detremexa, which includes D. Marilandica and D. Medsgeri; and Chamaecrista, which includes Chamaecrista fasciculata and Chamaecrista procumbens.

2. As a result of this work the following diagnostic (new) characters were found:

(a) Genetical characters.

<table>
<thead>
<tr>
<th>D. Marilandica and D. Medsgeri</th>
<th>C. fasciculata and C. procumbens</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. Terminal branches of veinlets anastomose near the margins of leaflets.</td>
<td>1. Terminal branches of veinlets end abruptly near the margins. Veinlets do not anastomose.</td>
</tr>
<tr>
<td>2. Seeds of Brown or olive color, not shiny and elliptical. Testa exhibits a large depression which corresponds to the position of the embryo.</td>
<td>2. Seeds are smaller, black, shiny, and rectangular. Testa is pitted and the pits are arranged in longitudinal rows. Depressions are absent.</td>
</tr>
<tr>
<td>3. Sepals exhibit small, minutely papillose, unicellular marginal hairs.</td>
<td>3. Sepals exhibit large multicellular hairs only in the neighborhood of the midrib. Hairs not papillose.</td>
</tr>
<tr>
<td>4. Stipular hairs large, unicellular and minutely papillose.</td>
<td>4. Stipular hairs small, multicellular, rarely papillose.</td>
</tr>
</tbody>
</table>
(b) Ecological characters.

<table>
<thead>
<tr>
<th>D. Marilandica and D. Medsgeri</th>
<th>C. fasciculata and C. procumbens</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. Stomata absent from the upper surfaces of leaflets. Stomata large and elliptical.</td>
<td>1. Both leaf surfaces show stomata present. Stomata small and tend to be circular.</td>
</tr>
<tr>
<td>2. Stomata not distributed uniformly upon the upper epidermises of the stipules.</td>
<td>2. Stomata distributed rather uniformly upon both epidermises of the stipules.</td>
</tr>
<tr>
<td>3. Flower buds appear the first week in July. Full-grown flowers produced about the middle of July. Flowers lose their sepals, petals and stamens about August 13 and August 2, respectively.</td>
<td>3. Flower buds appear about the middle of July. Full-grown flowers produced about August 1. About one-half of the plants exhibited flowers until the end of the growing season.</td>
</tr>
<tr>
<td>4. Fractured stems show the pith to be either a light or deep orchid color.</td>
<td>4. Fractured stems always show white or cream colored pith.</td>
</tr>
</tbody>
</table>

(3) It has been shown that the adulteration of the official senna leaflets with the Maryland senna leaflets can be detected microscopically.

(4) This work is of value taxonomically as it shows the method used to find characteristics of diagnostic importance.
VIII. References.


5. Joensson, Pharm. Era, 56, 607 (1923); through Y.B.A. Ph. A., 12, 106.


7. La Wall, Four Thousand Years of Pharmacy, 102-103.


27. Ensign, Amer. J. Botany, 8, 433 (1921).

