1964

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Recommended Citation

Webb, Alice-Ann and Carlquist, Sherwin (1964) "Leaf Anatomy as an Indicator of *Salvia Apiana-mellifera* Introgression," *Aliso: A Journal of Systematic and Floristic Botany*. Vol. 5: Iss. 4, Article 4.
Available at: <https://scholarship.claremont.edu/aliso/vol5/iss4/4>

LEAF ANATOMY AS AN INDICATOR OF SALVIA APIANA-MELLIFERA INTROGRESSION

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INTRODUCTION

The hybridization of *Salvia apiana* and *S. mellifera* is a classical instance of introgressive hybridization, and has been described in detail by Epling (1947) and Anderson and Anderson (1954). These authors have described the circumstances of this hybridization and the characters (chiefly those of flowers and inflorescence) which serve to distinguish the two species and indicate degree and nature of hybridization. Other information on these species has been given by Epling (1942) and Epling and Lewis (1942). The clarity of the *S. mellifera*-*S. apiana* introgression invites anatomical study, especially because there are obvious differences between the species in texture and other fine structural points which suggested that anatomical study would be rewarding. Study of the *S. mellifera*-*S. apiana* introgression has thus far not featured leaf anatomy, which offers good material for analysis.

The potential usefulness of anatomy in analysis of plant hybrids does not seem to be generally appreciated. Anatomy may require more work in preparation, but as the results below show, certain anatomical features may be extremely decisive in establishing the nature of individuals and populations. A number of papers do emphasize the use of anatomical features, chiefly those of leaves and indument, in study of hybrids. These include Barua and Wight (1958), Cannon (1909), Cousins (1933), Heiser (1949), MacFarlane (1892), Pryor, Chattaway, and Kloot (1956), Rollins (1944), and Russell (1919).

Leaves alone are the focus of the present study not only because they have hitherto received little attention, but also because they offer a wide variety of anatomical features. Other portions of the plant are potentially interesting, but would tend to be, in some cases, anatomical expressions of facts already known and analyzed in terms of gross morphology (e.g., differences in floral venation would parallel differences in floral shape).

MATERIALS AND METHODS

The specimens of *Salvia apiana*, *S. mellifera*, and putative hybrids were collected in the field, mostly in regions near Claremont. Selection of representatives of these species and their hybrids was based on criteria for identification developed by Epling (1947) and Anderson and Anderson (1954). Leaves were fixed in formalin-propiono-alcohol (Johansen, 1940). Sections were prepared with the usual paraffin techniques and stained with safranin and fast green. Fixed leaves were also cleared in 2.5% sodium hydroxide followed by a 250% chloral hydrate solution to continue the clearing process yet inhibit

¹This paper is derived from a Senior Thesis presented by Miss Webb in partial satisfaction of the degree of Bachelor of Arts at Pomona College.

softening. Cleared leaves were dehydrated in an ethyl alcohol series, stained in safranin, destained and then stored in xylene, and finally mounted on slides in canada balsam. Specimens documenting this study are as follows: *S. mellifera*: Webb 768, 769, 774, 775, 777, 781, 785, 793, and 798; *S. apiana*: Webb 770, 771, 773, 779, 780, 782, 792, 794, 795, 796, and 799; *S. mellifera* × *S. apiana*: Webb 766, 767, 772, 783, 784, 786, 787, 789, 790, and 791. The specimens of *S. apiana* are mostly from the rocky alluvial areas between the Rancho Santa Ana Botanic Garden and the foothills of the San Gabriel Mountains north of Claremont. This alluvial fan is at an elevation of about 1200 feet, and is characterized by a scrub vegetation including *Eriogonum fasciculatum*, *Adenostoma fasciculatum*, *Lepidospartum squamatum*, *Sambucus callicarpa*, and *Rhus ovata*. The specimens of *Salvia mellifera* are mostly from the foothills to the north of this alluvial fan, in an area covered with *Quercus agrifolia*, *Q. dumosa*, *Artemisia californica*, *Rhus laurina* and other chaparral shrubs. Hybrids tend to occur in disturbed areas in these foothills. Herbarium specimens of all of the collections listed above were prepared and have been deposited in the Herbarium of the Rancho Santa Ana Botanic Garden.

Many anatomical and morphological features of the leaves were studied, and various qualitative and quantitative characteristics were analyzed. Those which have been selected for presentation here are believed to contain features which serve as the best anatomical criteria for differentiating the two species, and thus for analyzing putative hybrids. Some features (e.g., glandular trichomes) which do not differentiate the species have been included for their value in comparison with those which do, and to present a more complete picture of leaf anatomy.

The means which are presented in table 1 are the averages of the averages obtained from each plant in the three categories. Three leaves were usually studied in each individual, for each characteristic. Therefore, each mean is usually the average of 27–30 leaves for each species (or for the hybrid group).

The statistical interpretations of significance in table 2 are based on the "U" test of Siegel (1956). This simple test does not require the use of a calculator, and can therefore be used in the field. The formulae used are:

$$U = n_1 n_2 + \frac{n_1(n_1 + 1)}{2} - R_1$$

$$U = n_1 n_2 + \frac{n_2(n_2 + 1)}{2} - R_2$$

In these formulae, n = number of samples, n_1 = species 1 (e.g., *S. apiana*), n_2 = species 2 (e.g., *S. mellifera*) and R = rank. In this test, the species are ranked, with one being assigned to the lowest value; these ranks are then added for each species, equalling R . The calculated U must be smaller than the value found in the table of values of Siegel for any given dimension. If U is larger than the value at the 5% level of probability, this indicates that the variation is not significant. In other words, this much variation could be found within one species or even within the leaves on an individual plant. If the value of U is smaller than the value given in the table, then the variation or difference between the two species (or between one species and the hybrids) may be considered statistically significant.

The commentary of Dr. Edgar Anderson upon the potential value of leaf anatomy in analysis of the *S. apiana*-*S. mellifera* introgression inspired this study, and his interest is gratefully acknowledged.

ANATOMICAL FEATURES

LEAF MARGINS

As shown in fig. 1, three characteristics of margins were analyzed quantitatively: (1)

depth of sinuses between lobes; (2) length of lobes (parallel to the margin); and (3) the distance of the recurvature of the margin. Means for these are given in table 1.

Lobes.—Although the depth of sinuses does show a difference between the two species, as the photographs in fig. 3–4 would suggest, this difference does not prove statistically significant, as shown in table 2. Probably this difference could be significant if the length of the lobes (perpendicular to the leaf axis) including recurved portion of margins could have been measured. A separate calculation of recurvature of margins was more convenient and simpler, however, and is discussed below.

TABLE 1. *Quantitative leaf characteristics: means*

CHARACTERISTIC	S. MELLIFERA	S. APIANA	HYBRIDS
<i>Margins:</i>			
Depth of sinuses (1), μ	354.9	322.3	298.8
Length of lobes (2), μ	1186.5	1696.5	1230.3
Recurvature of lobes (3), μ	237.7	0.0	114.7
<i>Venation:</i>			
Area of areoles, mm ²	2.47	3.62	2.86
Vein-endings per areole	.668	.902	.745
<i>Sections:</i>			
Thickness of leaf through major veins (4), μ	300.7	322.8	289.8
Thickness of leaf between major veins (5), μ	163.9	198.3	164.7
Depth of pockets (6), μ	231.9	123.3	161.7
Diameter of pockets (7), μ	311.1	145.8	265.6
<i>Epidermis:</i>			
Thickness of outer wall of upper epidermis, μ	10.21	4.27	6.26
Length $\times$ width of larger epidermal cells, μ	161.1 $\times$ 123.6	100.3 $\times$ 72.0	151.5 $\times$ 100.8
Length $\times$ width of smaller epidermal cells, μ	123.0 $\times$ 110.5	66.6 $\times$ 55.8	106.2 $\times$ 94.8
Upper epidermis: per cent of cells bearing non-glandular trichomes	.89	68.69	35.87
Upper epidermis: per cent of cells bearing glandular trichomes	3.08	3.12	3.16
Upper epidermis: per cent of cells which are guard cells	0.0	3.16	2.09
Lower epidermis: per cent of cells bearing non-glandular trichomes	27.22	62.06	46.91
Lower epidermis: per cent of cells bearing glandular trichomes	12.62	3.35	10.18
Lower epidermis: per cent of cells which are guard cells	11.32	9.47	14.46

Length of lobes easily sets *S. mellifera* apart from *S. apiana*, as shown in table 1 and table 2. The short lobes of *S. mellifera* are manifestly distinct from the long lobes of *S. apiana*, as comparison of fig. 3 and fig. 4 suggests. In the hybrids (fig. 5, 6), the lobes are intermediate in length but tend to be very irregular, and longer lobes may alternate with shorter ones. The difference between *S. mellifera* and the hybrids is not significant, and might indicate the tendency of the hybrids to resemble *S. mellifera* more than *S. apiana*.

Recurvature.—The presence or absence of recurvature on the margins is a characteristic obvious upon superficial examination of the leaves. As shown in table 1, there is no

recurvature of margins in *S. apiana*, whereas lobes in *S. mellifera* are prominently revolute, forming conspicuous flaps on the lower surface of the leaf. The hybrids show a mean figure for recurvature which falls between that of *S. mellifera* and the negative figure for *S. apiana*. Most of the hybrids have a degree of marginal recurvature like the minimal condition in *S. mellifera*; three hybrid plants, however, showed no marginal curl. Thus, marginal recurvature clearly differentiates *S. apiana* from *S. mellifera* but would not be a good characteristic for determining the hybrid character of some putatively hybrid plants, despite the positive discrimination shown in table 2.

TABLE 2. Statistical significance of comparisons of data from table 1.

CHARACTERISTIC	S. MELLIFERA TO S. APIANA	S. MELLIFERA TO HYBRIDS	S. APIANA TO HYBRIDS
<i>Margins:</i>			
Depth of sinuses (1)	0	0	0
Length of lobes (2)	+	0	+
Recurvature of lobes (3)	+	+	+
<i>Venation:</i>			
Area of areoles	—	—	—
Vein-endings per areole	—	—	—
<i>Sections:</i>			
Thickness of leaf through major veins (4)	0	0	0
Thickness of leaf between major veins (5)	+	0	+
Depth of pockets (6)	+	+	0
Diameter of pockets (7)	+	0	+
<i>Epidermis:</i>			
Thickness of outer wall of upper epidermis	+	+	0
Length × width of larger epidermal cells	+	0 0	+
Length × width of smaller epidermal cells	+	+	+
Upper epidermis: per cent of cells bearing non-glandular trichomes	+	+	+
Upper epidermis: per cent of cells bearing glandular trichomes	0	0	0
Upper epidermis: per cent of cells which are guard cells	+	+	+
Lower epidermis: per cent of cells bearing non-glandular trichomes	+	+	+
Lower epidermis: per cent of cells bearing glandular trichomes	0	0	0
Lower epidermis: per cent of cells which are guard cells	0	0	0

Hydathodes.—The presence of hydathodes is related to subtle differences in the shape of lobes of *S. apiana* as compared with those of *S. mellifera*. As shown in fig. 4, a single hydathodic vein-ending per lobe is characteristic of leaves of *S. apiana*. Hydathodes are absent in *S. mellifera* (fig. 3). Some hybrids show weakly-developed hydathodes (fig. 5) whereas others do not (fig. 6). The presence of hydathodes in *S. apiana* is curious because hydathodes, typically, are to be expected in plants of moister environments. Although neither of the two *Salvias* can be said to grow in a mesic habitat, *S. apiana* does grow in drier sites than *S. mellifera* (Anderson and Anderson, 1954). The revolute margins of *S. mellifera* would probably not be expected to contain hydathodes on account of their morphology. Revolute margins characterize plants in chaparral-like habitats, and although

they may be more frequent in the *macchia* of the Mediterranean region than in the Californian chaparral, they may be found in some plants of the latter region, such as *Eriogonum fasciculatum*, *Trichostema lanatum*, *Diplacus* spp., *Ceanothus papillosus* and other species of *Ceanothus*, and various tarweeds. Although commonly distinguished from chaparral by ecologists, the coastal sage may be expected to contain plants with similar leaf characteristics.

Fig. 1-2. Fig. 1. Diagram of measurements used on leaf margins. The margin, above, is shown double because it is curled over (revolute). "1" = depth of sinus between lobes; "2" = length of lobe; "3" = width of curled-over portion of margin.—Fig. 2. Diagram of measurements used on leaf sections. "4" = thickness of leaf where major bundles occur, between pockets; "5" = thickness of leaf between main bundles, above pockets; "6" = depth of pockets; "7" = diameter of pockets.

VENATION PATTERN

Subtle differences were observed when comparing the venation patterns of cleared leaves of the two species; two measures were selected; both concern areoles. An areole, for purposes of this study, is considered the smallest area completely enclosed by veinlets. Within such areoles, terminal vein-endings may be present. As shown in table 1, there are differences between the two species with respect to areole size and number of vein-endings per areole. The statistical significance of these differences was not calculated because environmental modifiability of areole size, even within an individual, is so very great. Statistical significance is thus virtually ruled out in terms of the sampling in the present study. Sampling was not sufficiently exhaustive to rule out the possibility that the results in table 1 were achieved by chance. Nevertheless, these features deserve careful study in comparing species. The larger number of vein-endings in *S. apiana* may well correspond to the larger size of areoles characteristic of that species. There is still a possibility that these venation features would represent valid differences between the species if exhaustively studied under controlled conditions.

CHARACTERISTICS OF SECTIONED LEAVES

Many differences are apparent when sectioned material of *S. apiana*, *S. mellifera*, and the hybrids is compared (fig. 7-10). The basic composition of mesophyll in the two species is similar. Two layers of palisade and two or three layers of spongy tissue are generally present. Veins and veinlets are sheathed by thin-walled non-photosynthetic cells. Larger veins are associated with vein-sheath extensions or partial vein-sheath extensions. Resin-like deposits may be seen in mesophyll and epidermal cells. Crystalline sphaeroidal deposits were observed in epidermal cells of *S. mellifera* (fig. 11).

Thickness.—Four measures of portions of leaf transactions were devised. These, shown in fig. 2, are: (4) thickness of leaf between pockets, through veins; (5) thickness of leaves

above pockets, between major veins; (6) depth of the pockets on the abaxial surface of the leaf; and (7) diameter of pockets. Obviously, any given pocket may not be sectioned near its center, but in a large sample the measurements in sections will be equally distributed between median and edge of pockets. The measurements, as shown in tables 1 and 2, indicate that width of leaf if measured through a vein is rather constant and does not serve to differentiate the species. However, leaves of *S. apiana* are much thicker than those of *S. mellifera* if measured in areas between major veins. The hybrids tended to resemble *S. mellifera* more closely than *S. apiana* in this characteristic, indicating a possible dominance of the thinness of the *S. mellifera* leaf, at least insofar as hybrids as a whole tend to resemble *S. mellifera* more closely in most characteristics (Epling, 1947).

Pockets.—The pockets on leaves, as seen by comparing fig. 7 and 8, markedly differentiate *S. mellifera* and *S. apiana*. The pockets in *S. mellifera* leaves are large, arching up to form a bullate surface on the leaf. Although the upper surface of the leaf in *S. apiana* is relatively plane, not exhibiting a bullate characteristic, there are pockets on the lower surface of the leaf. Hybrids exhibit intermediate conditions: the hybrid shown in fig. 9 shows a tendency toward the bullate condition of *S. mellifera*, whereas the hybrid of fig. 10 resembles the plane upper surface and smaller pockets of *S. apiana*. As shown in table 2, measurements of pockets are significant in differentiating *S. apiana* from *S. mellifera*. Curiously enough, hybrids tend to have somewhat shallower pockets than one might expect, thus resembling *S. apiana* more closely. The pockets are, at the same time, wider, resembling *S. mellifera* more closely. These two measurements used conjunctively would enable one to distinguish hybrids from the parents.

EPIDERMAL CHARACTERISTICS

Epidermal Wall.—As can be seen by comparing fig. 11 and 12, the outer wall of adaxial epidermis cells of leaves of *S. mellifera* is much thicker than that of *S. apiana*. This feature is statistically significant in separating *S. mellifera* and *S. apiana*. The hybrids (fig. 13, 14) may be intermediate as a whole, but the mean tends to be within the potential variability of *S. apiana*.

The outer wall of the epidermis tends to be thicker at the margins. This is especially prominent in *S. mellifera* (fig. 15), in which the average thickness of the wall at margins is 12.8μ . The epidermal surface of leaves of *S. mellifera* tends to be prominently raised into grooves and valleys in areas at or near the margins (fig. 15). Such relief is absent from leaves of *S. apiana*. Some relief was observed on leaves of hybrids.

Dimensions of Epidermal Cells.—The dimensions—length and thickness (or width, if a two-dimensional view is taken)—of cells of the adaxial epidermis were measured. Only cells which did not bear a trichome were measured—no easy task in *S. apiana*. Two sizes of cells were visible on sections in all collections. This fact may in part result from sectioning (median and non-median) of cells, but these two sizes were computed separately, despite the fact that this probably is an artificial procedure. Nevertheless, the longer, wider size of cells of the upper epidermis in *S. mellifera* as compared with those of *S. apiana* is clearly evident. Hybrids were intermediate in size of these cells, as expected, but they tend to

Fig. 3–6. Cleared leaves of *Salvia* specimens, showing margin and venation.—Fig. 3. *S. mellifera*, Webb 775. Note relatively even size of lobes, short length, and rounded shape; although not clearly visible, margin of lobes is curled over.—Fig. 4. *S. apiana*, Webb 770. Margin is not revolute, and lobes are long; most lobes contain a prominent hydathode (dark tip on three of the lobes shown).—Fig. 5. Hybrid, Webb 766. Lobes are irregular in size and shape; inconspicuous hydathodes may be seen on some lobes.—Fig. 6. Hybrid, Webb 783. Lobes are rounded, like those of *S. mellifera*, and have revolute margins, but are irregular in shape and size. All, $\times 13$.

FIGURES 3-6

approximate *S. mellifera* more closely, as the significant differences in most cases between *S. apiana* and the hybrids tend to show.

Trichomes.—The characteristics of trichomes analyzed included frequency (number of cells bearing a trichome compared with total number of epidermal cells) on both upper and lower surfaces, and types of trichomes present.

The types of trichomes illustrated in fig. 16–22 characterize both species. Thus, in contrast with other instances of hybrids analyzed anatomically, such as *Parthenium* (Rollins, 1944) in which different trichomes characterize different species, distinctions between the two species of *Salvia* depend solely on distribution, not morphology. Non-glandular trichomes vary from short, with an appressed terminal cell (fig. 16) through intermediate types (fig. 17) to long “woolly” types (fig. 18). Glandular trichomes may be short, with a single terminal cell (fig. 19, 20) or with a pair of glandular cells (fig. 21) or several glandular cells, forming a peltate trichome (fig. 22) in which a broad subsessile head is formed. Frequency of glandular and nonglandular trichomes was computed independently, and these figures are given in table 1. These figures were computed as percentages of the total number of epidermal cells counted.

As can be seen from these figures, glandular trichomes are of about the same frequency in both species, as well as in the hybrids, on the upper epidermis. On the lower surfaces of leaves, however, glandular trichomes are more abundant in *S. mellifera* than in *S. apiana*.

Non-glandular trichomes on the upper surfaces of leaves proved a most distinctive feature, because the great frequency of non-glandular trichomes in *S. apiana* is in marked contrast to their infrequency in *S. mellifera*. This extreme difference permits intermediate conditions in the hybrids to stand out in bold relief, and they are statistically separable from both of the parental species. This reflects the fact that even a relatively small degree of hybridity would markedly reduce the abundance of trichomes as compared to *S. apiana*, or increase the abundance as compared to *S. mellifera*. Non-glandular trichomes are more frequent on the lower surface of *S. mellifera* leaves than on the upper surface, but on leaves of *S. apiana*, they are about equally frequent on both surfaces. Despite the greater frequency of non-glandular trichomes on lower surfaces of *S. mellifera*, they can still be used for discriminating between the two species, and between the hybrids and either of the parental species (see tables 1 and 2). Thus, abundance of non-glandular trichomes on either surface of leaves provides as clear a discriminant as could be desired.

Stomata.—Like trichomes, stomata were computed as a percentage of the total number of epidermal cells counted. As seen in table 1, stomata are totally absent on the upper surface of leaves of *S. mellifera* examined. They are relatively abundant (3.16%) on the upper surface of *S. apiana* leaves. These frequencies are doubtless related to the fact that trichomes are relatively dense on the upper surface of *S. apiana* leaves, infrequent on upper surfaces of *S. mellifera* leaves. This would seem to lend some support to those who believe that a woolly covering of non-glandular trichomes limits transpiration.

The greater frequency of stomata on the lower surface of *S. mellifera* leaves as compared with the lower surface of *S. apiana* leaves would seem to suggest that *S. mellifera* compensates for lack of stomata on the upper surface by a greater frequency of stomata on the lower surface, a frequency greater than in *S. apiana*. Nevertheless, the difference between

Fig. 7–10. Transections of *Salvia* leaves.—Fig. 7. *S. mellifera*, Webb 775. Note large pocket on lower surface of leaf, and nearly bare upper epidermis.—Fig. 8. *S. apiana*, Webb 770. Pockets are irregular and shallow; trichomes are abundant on upper epidermis.—Fig. 9. Hybrid, Webb 789.—Fig. 10. Hybrid, Webb 766. The two hybrids show intermediate conditions in leaf anatomy. All, $\times$ ca. 165.

FIGURES 7-10

FIGURES 11-14

the two species in abundance of stomata on lower surfaces of leaves did not prove sufficient to achieve statistical significance with the test used in this study.

Fig. 15. Margin, from transection of leaf of *Salvia mellifera*, Webb 776. Note partially revolute margin, and marked epidermal relief which tends to occur near margins of leaf. $\times 600$.

CONCLUSIONS

Of the features analyzed in this study, no statistical significance could be attributed to differences between *Salvia apiana* and *S. mellifera* in the following characteristics: depth of sinuses on leaf margins; thickness of leaves when measured through veins; abundance of glandular trichomes; abundance of stomata on abaxial surfaces of leaves. Since these characteristics did not statistically distinguish the two species, obviously they are of no use in detecting hybridity in individuals. The remaining characteristics all show statistical significance in separating *S. mellifera* and *S. apiana*. Presence or absence of hydathodes might be added, as might also venation features if extensive sampling could have been employed. Of these characteristics, however, only a few are consistently useful in discriminating between the hybrids and *both* parental species. These include: length of margin recurvature; abundance of non-glandular trichomes on upper surfaces of leaves; abundance of trichomes on lower surfaces of leaves; and abundance of stomata on upper surfaces of leaves.

Fig. 11-14. Adaxial epidermis of *Salvia* leaves, from leaf transections.—Fig. 11. *S. mellifera*, Webb 775. Note thick outer cell wall; crystalline sphaeroids are shown in epidermal cells.—Fig. 12. *S. apiana*, Webb 770. Outer wall is relatively thin.—Fig. 13. Hybrid, Webb 789.—Fig. 14. Hybrid, Webb 766. Fig. 13 shows resemblance to *S. mellifera*, fig. 14 simulates *S. apiana*. All, $\times 685$.

The hybrids tended toward *S. mellifera* (difference not statistically significant) in the following features: length of lobes on margins of leaves; size of epidermal cells; thickness of leaf (between major veins); and width of pockets in the lower epidermis. Features in which the hybrids were not significantly different from *S. apiana* included only depth of pockets on lower surfaces of leaves and thickness of outer epidermal cell wall on upper

Fig. 16-22. Types of trichomes, from leaf transections of *Salvia apiana*. All of these trichomes may be found on *S. mellifera* also. Fig. 16-18. Non-glandular trichomes; fig. 19-22. Glandular trichomes.—Fig. 16. Trichomes with terminal cell parallel to surface of leaf.—Fig. 17. Trichome with diagonal orientation of terminal cell.—Fig. 18. Long, erect trichome.—Fig. 19, 20. Trichomes with a single terminal cell.—Fig. 21. Trichome with a pair of terminal cells; cuticle has separated from the apical portion of the cells.—Fig. 22. A large peltate trichome. All, $\times 685$.

surfaces of leaves. Thus hybrids would tend, on anatomical grounds, to show more numerous resemblances to *S. mellifera* than to *S. apiana*.

This situation is interesting because it is in agreement with a conclusion reached by Epling (1947). Epling claimed that conformations of the corolla in *S. apiana* (with divergent stamens) and *S. mellifera* (which has slightly exerted stamens) are distinctive and a barrier to bee pollination. For reasons of corolla morphology, pollen would be less likely to be carried by bees from *S. mellifera* to *S. apiana* plants than from *S. apiana* to *S. mellifera*. *Salvia mellifera* would therefore more frequently be a maternal parent, and F_1 hybrids would tend to backcross to *S. mellifera*. Therefore, hybrids would more frequently resemble *S. mellifera* than *S. apiana*. Despite the existence of individuals with some *S. apiana*-like characteristics (e.g., Webb 766), leaf anatomy shows a preponderance of *S. mellifera*-like features.

The utility of the anatomical method in analyzing hybrids seems amply demonstrated by the present study. In spite of the additional effort involved in microtechnical preparations, fine discriminants of differences between species are available, and sensitive indicators of hybridity can be demonstrated.

LITERATURE CITED

- Anderson, E., and B. R. Anderson. 1954. Introgression of *Salvia apiana* and *Salvia mellifera*. *Ann. Mo. Bot. Gard.* 41: 329-339.
- Barua, P. K., and W. Wight. 1958. Leaf sclereids in the taxonomy of thea camellias. *Phytomorphology* 8: 257-264.
- Cannon, W. A. 1909. Studies in heredity as illustrated by the trichomes of species and hybrids of *Juglans*, *Oenothera*, *Papaver*, and *Solanum*. *Carnegie Inst. Wash. Publ.* 117: 1-67.
- Cousins, S. M. 1933. The comparative anatomy of the stem of *Betula pumila*, *Betula lenta* and the hybrid *Betula jackii*. *J. Arnold Arb.* 14: 351-355.
- Epling, C. 1942. California Salvias. *Ann. Mo. Bot. Gard.* 25: 95-188.
- . 1947. Natural hybridization of *Salvia apiana* and *S. mellifera*. *Evolution* 1: 69-78.
- , and H. Lewis. 1942. Centers of distribution of the chaparral and coastal sage associations. *Am. Midland Naturalist* 27: 445-462.
- Heiser, C. B., Jr. 1949. Study in the evolution of the sunflower species *Helianthus annuus* and *H. bolanderi*. *Univ. Calif. Publ. Botan.* 23: 157-208.
- Johansen, D. A. 1940. *Plant microtechnique*. McGraw Hill. New York. 523 p.
- MacFarlane, J. M. 1892. A comparison of the minute structure of plant hybrids with that of their parents and its bearing on biological problems. *Trans. Roy. Soc. Edinburgh* 37: 203-286.
- Pryor, L. D., M. Margaret Chattaway, and N. H. Kloot. 1956. The inheritance of wood and bark characters in *Eucalyptus*. *Austral. J. Botan.* 4: 216-239.
- Rollins, R. C. 1944. Evidence for natural hybridity between guayule (*Parthenium argentatum*) and mariola (*Parthenium incanum*). *Am. J. Botan.* 31: 93-99.
- Russell, A. M. 1919. The macroscopic and microscopic structures of some hybrid *Sarracenias* compared with that of their parents. *Contrib. Botan. Lab. Univ. Penn.* 5: 1-41.
- Siegel, S. 1956. *Non-parametric statistics for the behavioral sciences*. McGraw Hill. New York.