TECHNICAL BULLETIN NO. 82 JANUARY 1971

# Morphological Variation and Interspecific Hybridization among *Desmodium intortum, Desmodium sandwicense,* and *Desmodium uncinatum*

PETER P. ROTAR and KUAN-HON CHOW

HAWAII AGRICULTURAL EXPERIMENT STATION, UNIVERSITY OF HAWAII

## CONTENTS

## THE AUTHORS

PETER P. ROTAR is Associate Professor of Agronomy, College of Tropical Agriculture, University of Hawaii, and Associate Agronomist, Hawaii Agricultural Experiment Station. KUAN-HON CHOW is a former graduate student, Department of Agronomy and Soil Science.

# Morphological Variation and Interspecific Hybridization among *Desmodium intortum, Desmodium sandwicense,* and *Desmodium uncinatum*[1]

PETER P. ROTAR and KUAN-HON CHOW

## INTRODUCTION

*Desmodium intortum* (Mill.) Urb. 'Greenleaf' and *D. uncinatum* (Jacq.) DC. 'Silverleaf' are important pasture legumes in Hawaii, the wetter coastal areas of northeastern Australia, and elsewhere in the tropics. *D. sandwicense* E. Mey. has also been tried with varying degrees of success. *D. intortum* × *D. sandwicense* hybrids have been obtained in Hawaii when the two species have been grown side by side.

The purpose of this study was to investigate the breeding behavior and morphological variation among the three species and their $F_1$ and $F_1 \times F_1$ hybrids.

## REVIEW OF LITERATURE

Hutton and Gray *(2)* reported the first successful fertile crosses from the combinations *D. sandwicense* × *D. uncinatum* and *D. intortum* × *D. uncinatum*. McWhirter *(3)* first reported successful crosses between *D. intortum* × *D. sandwicense*. He reported a high degree of compatibility from his crosses, whereas Hutton and Gray found wide differences in compatibility among their crosses. Morphological characters observed in the crosses of Hutton and Gray appeared to segregate in a normal Mendelian fashion.

Park and Rotar *(5)*, in detailed studies of *D. sandwicense*, found varying shades of red-colored flowers to be dominant to near-white-colored flowers; red stem color to be dominant to green stem color; the silver-colored leaflet

[1] Part of a thesis submitted by the junior author to the Graduate Division of the University of Hawaii in partial fulfillment of the requirements for the Master of Science degree.

marking to be dominant to green-colored leaflet; and that the flower color and stem color were linked to each other. They also observed considerable variation in the color of the flowers and in the redness of the stems.

Hutton *(1)* and Rotar et al. *(8)* have reported on the breeding behavior of the three species. Self- and cross-fertilization occurred in all three species. Hutton *(1)* indicated that they would set seed only after tripping, whereas Rotar et al. indicated that they would set seed without being tripped. The observed variations were probably due to the environmental differences between the two locations.

McWhirter *(4)* found a male sterility factor in *D. sandwicense*. When *D. sandwicense* was used as the male parent in crosses with *D. intortum*, the $F_1$ hybrids were uniformly male sterile. Progenies from the cross *(D. intortum* ♀ × *D. sandwicense* ♂*)* ♀ × *(D. intortum)* ♂ were completely fertile, indicating that *D. intortum* acted as a fertility restorer.

The genus *Desmodium* has been shown to comprise of diploid species with $2n = 20$ or $2n = 22$ chromosomes *(7)*. The three species reported here are diploids with $2n = 22$ chromosomes.

## MATERIALS AND METHODS

Five plants of *D. sandwicense*, numbered S11, S21, S31, S41, and S51; eight plants of *D. uncinatum*, numbered U12, U22, U32, U42, U52, U62, U72, and U82; and six plants of *D. intortum*, numbered I13, I23, I33, I43, I53, and I63, were used for interspecific hybridization in this study. Several plants were lost during the study due to virus disease. A tabular summary of the morphological characteristics of the surviving 15 plants is presented in Appendix Table A.

Stem internode length, leaflet width, and leaflet length were measured to the nearest millimeter; and raceme length was measured to the nearest 0.5 centimeter. Leaflet pubescence was measured by counting the number of hairs observed under a microscope within a circle of 3.2 mm diameter, and this in turn was multiplied by 12.5 to obtain the number of hairs per $cm^2$. Stem color was grouped into three categories: red, brown, or green. Growth habit was classified as upright, intermediate, or spreading. Plant vigor was rated as excellent, good, or poor. The leaflet length multiplied by its width was used as an index of leaflet size, and the ratio of leaflet length to width was obtained from the leaflet length divided by the leaflet width.

Crosses were made according to the method described by Rotar et al. *(8)*. Flower buds were emasculated in the late afternoon the day before they opened. Pollinations were carried out the next morning. The racemes were kept covered with fiber pollination bags containing wet cotton to maintain high relative humidity.

The following sets of crosses were made:

(a) Crosses among the three species.

(b) Crosses among the $F_1$ progenies of the three species:

*(D. sandwicense* ♀ × *D. intortum* ♂*)* ♀
× *(D. sandwicense* ♀ × *D. uncinatum* ♂*)* ♂

*(D. sandwicense* ♀ × *D. uncinatum* ♂*)* ♀
× *(D. sandwicense* ♀ × *D. intortum* ♂*)* ♂

*(D. sandwicense* ♀ × *D. intortum* ♂*)* ♀
× *(D. uncinatum* ♀ × *D. intortum* ♂*)* ♂

(c) The $F_1$ progenies of (a) and (b) were selfed and advanced to the $F_2$ generation. Successful crosses were difficult to obtain. No backcrosses were made. $S_1$ seeds were obtained by bagging the racemes of $F_1$ plants and allowing the flowers to set seeds.

Flowering responses to daylength, germination of hybrid seed, percentage of and relationship of pollen abortion, and pod formation were compared among the parent plants and their $F_1$, $F_1 \times F_1$, and $F_2$ progenies.

Observations on flowering behavior were made on (i) number of flowers opened per raceme per day, (ii) number of days per raceme to complete flowering, (iii) number of flowers opened per raceme, and (iv) length of racemes. Percentage of pollen abortion was determined by counting at least 500 pollen grains from each of two flowers per plant. Pollen grains were classified as either normal (full) and stained with acetocarmine or shriveled and unstained.

Characteristics used in genetic studies were stem color, internode length, leaflet size, leaflet marking on the midrib, rugose leaflet, raceme length, and seed weight. Stem color was segregated into two classes: (i) red or brown, or (ii) green. The 5th internode from the stem tip of 10 different stems was measured to the nearest 0.5 cm for internode length. The middle leaflet of the 5th or 6th leaf from each of 10 stems was measured for length and width to the nearest millimeter. Raceme length was an average of 5 racemes per plant. Raceme measurements were made when the last few terminal flowers were opening. To determine seed weight, 100 seeds were weighed to the nearest milligram and then multiplied by 10 to obtain 1000-seed weight.

## RESULTS AND DISCUSSION

### Flowering behavior of the parent plants and their hybrids

Twenty-seven plants including 6 parent plants, 8 two-species hybrids, and 11 three-species hybrids were observed for flowering behavior during February and March 1967. Observations on number of flowers per raceme, flowering period in days per raceme, and number of flowers opened per day are presented in Table 1.

**TABLE 1. Flowering behavior of three *Desmodium* species and their $F_1$ and $F_1 \times F_1$ hybrids, February–March 1967**

| Parent plant, $F_1$ or $F_1 \times F_1$ hybrid no. | No. of flowers opened per raceme | | Flowering period per raceme, in days | | No. of flowers opened per raceme per day | |
|---|---|---|---|---|---|---|
| | Average | Range | Average | Range | Average | Range |
| *D. sandwicense* | | | | | | |
| S31 | 50.8 | 42–76 | 8.8 | 6–12 | 5.7 | 3.9–7.7 |
| *D. uncinatum* | | | | | | |
| U32 | 46.8 | 28–81 | 10.8 | 8–14 | 4.3 | 3.0–5.8 |
| U62 | 46.0 | 34–63 | 14.0 | 9–18 | 3.3 | 2.6–4.2 |
| *D. intortum* | | | | | | |
| I23 | 27.8 | 19–41 | 5.2 | 4–7 | 5.3 | 4.0–6.8 |
| I43 | 43.3 | 28–61 | 8.4 | 6–11 | 5.1 | 4.4–6.1 |
| I53 | 30.8 | 24–41 | 7.2 | 6–8 | 4.2 | 3.3–6.8 |
| $F_1$ hybrids | | | | | | |
| S11 × I23 | 36.8 | 28–48 | 6.6 | 5–8 | 5.6 | 4.0–7.4 |
| S31 × I23 | 58.4 | 36–77 | 9.0 | 7–11 | 6.5 | 4.5–7.7 |
| S11 × U22 | 40.8 | 28–57 | 10.8 | 8–13 | 3.8 | 2.5–5.2 |

| | | | | | | |
|---|---|---|---|---|---|---|
| S11 × U62 | 43.8 | 26–62 | 8.0 | 6–11 | 5.5 | 3.1–8.7 |
| S21 × U82 | 46.4 | 28–56 | 7.2 | 5–9 | 6.4 | 5.3–8.0 |
| S31 × U22 | 54.6 | 33–75 | 12.2 | 7–20 | 4.5 | 3.7–6.3 |
| S51 × U42 | 29.8 | 27–33 | 6.0 | 5–7 | 5.0 | 3.5–5.6 |
| $F_1 \times F_1$ hybrids | | | | | | |
| (S11 × I23) × (S31 × U22) | 29.0 | 19–36 | 7.8 | 4–10 | 3.7 | 3.2–4.7 |
| (S21 × I23) × (S11 × U22) | 71.0 | 56–101 | 25.8 | 23–29 | 2.7 | 2.0–4.3 |
| (S21 × I23) × (S11 × U22) | 53.0 | 44–67 | 15.0 | 11–20 | 3.5 | 2.2–5.0 |
| (S21 × I53) × (S31 × U22) | 51.8 | 31–63 | 12.0 | 8–19 | 4.4 | 3.3–6.3 |
| (S21 × I23) × (S21 × U72) | 29.0 | 21–48 | 7.4 | 4–11 | 3.9 | 2.1–7.0 |
| (S21 × I23) × (S21 × U22) | 52.0 | 41–66 | 15.4 | 12–19 | 3.4 | 2.6–4.7 |
| (S21 × I53) × (U52 × I33) | 43.6 | 29–59 | 14.6 | 8–15 | 3.0 | 1.9–3.9 |
| (S31 × I23) × (U52 × I33) | 40.0 | 26–66 | 15.0 | 9–19 | 2.7 | 1.7–3.7 |
| (S51 × I23) × (S31 × U22) | 49.0 | 31–60 | 12.0 | 10–15 | 4.1 | 2.1–6.0 |
| (S51 × I23) × (U52 × I23) | 62.2 | 44–78 | 27.2 | 21–32 | 2.3 | 1.9–3.2 |
| (S51 × I23) × (S11 × U22) | 30.0 | 21–38 | 10.0 | 6–17 | 2.9 | 2.0–4.7 |

**Number of flowers per raceme:** Among the parent plants, the average number of flowers per raceme varied from 27.8 to 50.8 with a range of 19 to 81 flowers. *D. intortum* had fewer flowers per raceme than the other two species. Among the $F_1$ hybrids, the average number of flowers per raceme varied from 29.8 to 58.4 with a range of 26 to 77 flowers. Among the three-species hybrids ($F_1 \times F_1$), the average number of flowers per raceme varied from 30.0 to 71.0 with a range of 19 to 101.

**Number of flowers opened per raceme per day:** Among the parent plants, the average number of flowers opened per day per raceme varied from 3.3 to 5.7 with a range of 2.6 to 7.7 flowers. $F_1$ hybrids had an average of from 3.8 to 6.5 flowers with a range of 2.5 to 8.7. The three-species hybrids averaged 2.3 to 4.4 flowers with a range of 1.7 to 7.0. The three-species hybrids had fewer flowers opened per raceme per day than the $F_1$ hybrids or their parents.

**Length of flowering per raceme:** The parent plants averaged from 5.2 to 14.0 days per raceme with a range of 6 to 18 days. $F_1$ hybrids averaged from 6.6 to 12.2 days with a range of 5 to 20 days. Three-species hybrids averaged 7.4 to 27.2 days with a range of 4 to 32 days. Three-species hybrids took much longer to complete flowering than the $F_1$ hybrids or their parents.

**Sensitivity to daylength:** Investigations of flowering sensitivity to daylength of the parent plants and some of their three-species hybrids were made regularly during the period September 1966 through July 1967. *D. sandwicense* flowered throughout the period. *D. uncinatum* started flowering in October and ended in April. *D. intortum* started flowering in December and ended in March. *D. uncinatum* started flowering 2 months earlier and ended 1 month later than *D. intortum*. Rotar et al. *(8)* have shown that *D. sandwicense* is indeterminate. Wang *(9)* has shown that *D. intortum* is a short-day plant. From general observations on *D. uncinatum*, it can be concluded that it is also a short-day plant. The largest number of hybrids were flowering during the short-day period, when 53 to 56 of the 64 plants were in flower. During the summer months, 24 to 30 of the 64 plants were in flower. Eight of the 64 failed to flower. These were all dwarfed with shortened internodes and few stems and leaves. It appears possible to extend the flowering period of *D. uncinatum* or *D. intortum* into the summer season by hybridization and selection procedures.

## Crosses and environmental effects on crossing behavior

**Seed set from crosses:** Results of crosses among the three-species are presented in Table 2. In making two-species combinations, 149 pods were obtained from 1623 pollinations. Pod formation was 9.1 percent, with an average of 3.7 seeds per pod. Among the parent plants, open-pollinated pod formation varied from 16.9 to 51.1 percent, with average number of seeds per pod ranging from 3.5 to 6.3. Percentage of pod formation and average number of seeds

**TABLE 2. Pod set, seed set, and seed germination among interspecific hybrids of three *Desmodium species***

| Cross | No. of flowers pollinated | No. of pods formed | Pod set (percent) | No. of seeds per pod | No. of seeds germinated | Seed germination (percent) |
|---|---|---|---|---|---|---|
| *D. sandwicense* × *D. intortum* | 507 | 72 | 14.2 | 3.8 | 190 | 70 |
| *D. sandwicense* × *D. uncinatum* | 372 | 57 | 15.3 | 4.0 | 82 | 36 |
| *D. intortum* × *D. uncinatum* | 306 | 7 | 2.3 | 2.7 | 7 | 37 |
| *D. uncinatum* × *D. intortum* | 102 | 5 | 4.9 | 3.2 | 15 | 94 |
| *D. intortum* × *D. sandwicense* | 336 | 8 | 2.4 | 3.5 | 12 | 43 |
| *(D. sandwicense* ♀ × *D. intortum* ♂*)* ♀ × *(D. sandwicense* ♀ × *D. uncinatum* ♂*)* ♂ | 1593 | 87 | 5.6 | 2.8 | 127 | 51 |
| *(D. sandwicense* ♀ × *D. uncinatum* ♂*)* ♀ × *(D. sandwicense* ♀ × *D. uncinatum* ♂*)* ♂ | 220 | 10 | 4.6 | 2.3 | 8 | 35 |
| *(D. sandwicense* ♀ × *D. intortum* ♂*)* ♀ × *(D. uncinatum* ♀ × *D. intortum* ♂*)* ♂ | 933 | 37 | 4.0 | 3.0 | 39 | 36 |

per pod declined considerably from the two-species hybrids to the three-species hybrids. In making three-species combinations, 2746 pollinations were made and 134 pods were obtained, with an average of 2.7 seeds per pod. Percentage of pod formation of three-species hybrids varied from 4.0 to 5.6 percent, with average number of seeds per pod ranging from 2.3 to 3.0.

Percentage of pod formation from crosses was always lower than from the parent. Cross compatibility varied among the types of combinations made. If *D. sandwicense* was used as the female parent in crosses with *D. intortum* and *D. uncinatum*, the percentage of pod formation was relatively high, 14.2 percent and 15.3 percent, respectively. If *D. sandwicense* was used as the male parents the percentage of pod formation in crosses was low, 2.4 percent.

Percentage of pod formation from crosses among *D. intortum* and *D. uncinatum* was low, from 2.3 to 4.9 percent. The variability observed from one cross to the next suggested that physiological factors were influencing percentage of crosses.

**Environmental effects on crosses:** The relationship between pod formation from crosses and monthly temperatures is presented in Fig. 1. Higher temperatures resulted in poorer pod set. Correlation between percentage of pods set from crosses and the monthly average temperatures during the 1965–1966 winter season was negative and statistically highly significant, $r = -0.997$, $df = 3$. Over 500 crosses were made each month, from December to March, and 200 were made in April. The results are in agreement with those of Park and Rotar *(5)* and Rotar et al. *(8)* who indicated that the best results were obtained during cool weather with high relative humidity. This is also in agreement with Hutton *(1)* who found that pollen germination of *D. uncinatum* was poor when the relative humidity was low.

**Seed germination:** Percentage germination of open-pollinated seeds from the parent plants was usually higher than 95 percent (all seeds were scarified by pricking the seed coat with a needle). Percentage germination of hybrids was much lower. In two- and three-species hybrids the percentage germination varied from 37 to 94 percent, and from 35 to 51 percent, respectively. Although the three species are closely related as was indicated by pod set and seed set, the buildup of incompatibility differences due to possible chromosomal differences resulted in a lowering of seed viability in going from two-species to three-species hybrids.

Results of germination tests from open-pollinated seeds of the parent plants and their two- and three-species hybrids are presented in Table 3. The results are comparable to those presented in Appendix Table A. Percentage germination decreased considerably, falling from over 97 percent for parents, to 56 percent for two-species hybrids, and to 41 percent for three-species hybrids.

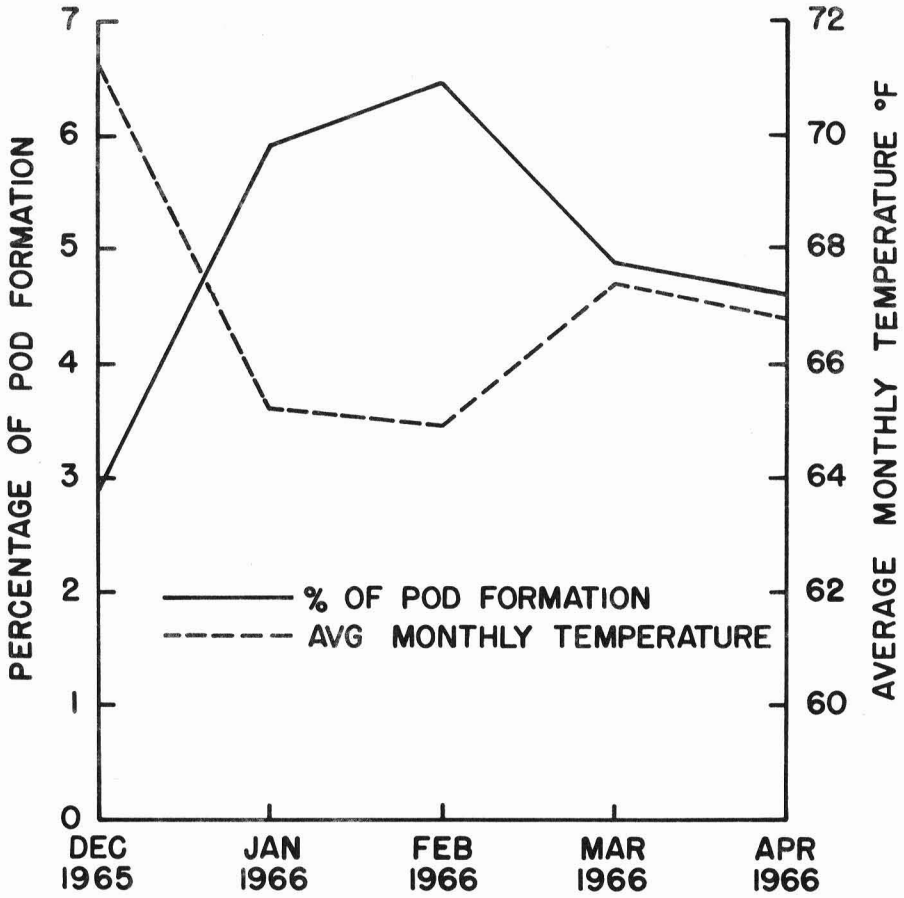


FIG. 1. Relationship between pod formation from *Desmodium* crosses and monthly temperatures, December 1965 through April 1966.

**TABLE 3. Percentage germination of open-pollinated seeds among three *Desmodium* species as compared with seeds from their $F_1$ and $F_1 \times F_1$ hybrids**

| Plant or cross | Seed germination (percent) |
|---|---|
| Parent plants | |
| *D. sandwicense* | 99 |
| *D. uncinatum* | 96 |
| *D. intortum* | 96 |
| $F_1$ hybrids | |
| *D. sandwicense* × *D. intortum* | 70 |
| *D. sandwicense* × *D. uncinatum* | 36 |
| *D. intortum* × *D. uncinatum* | 37 |
| *D. uncinatum* × *D. intortum* | 94 |
| *D. intortum* × *D. sandwicense* | 43 |
| $F_1 \times F_1$ hybrids | |
| *(D. sandwicense* × *D. intortum)* × *(D. sandwicense* × *D. uncinatum)* | 51 |
| *(D. sandwicense* × *D. uncinatum)* × *(D. sandwicense* × *D. intortum)* | 35 |
| *(D. sandwicense* × *D. intortum)* × *(D. uncinatum* × *D. intortum)* | 36 |

**TABLE 4. Average percentage pollen abortion among three *Desmodium* species and their $F_1$ and $F_1 \times F_1$ hybrids**

| Plant or cross | Average pollen abortion (percent) |
|---|---|
| Parent plants | |
| *D. sandwicense* | 2.5 |
| *D. uncinatum* | 4.5 |
| *D. intortum* | 2.6 |
| $F_1$ hybrids | |
| *D. sandwicense* ♀ × *D. uncinatum* ♂ | 2.5 |
| *D. sandwicense* ♀ × *D. intortum* ♂ | 22.1 |
| $F_1 \times F_1$ hybrids | |
| *(D. sandwicense* ♀ × *D. intortum* ♂*)* ♀ × *(D. sandwicense* ♀ × *D. uncinatum* ♂*)* ♂ | 19.3 |
| *(D. sandwicense* ♀ × *D. uncinatum* ♂*)* ♀ × *(D. sandwicense* ♀ × *D. intortum* ♂*)* ♂ | 1.8 |
| *(D. sandwicense* ♀ × *D. uncinatum* ♂*)* ♀ × *(D. uncinatum* ♀ × *D. intortum* ♂*)* ♂ | 10.9 |

**Pollen abortion:** Averages of pollen abortion studies are presented in Table 4. Parent plants had less than 7 percent aborted pollen. *D. uncinatum* had a higher percentage of pollen abortion than *D. sandwicense* or *D. intortum*. In the $F_1$ generation, hybrids of *D. sandwicense* × *D. uncinatum* had a low percentage of aborted pollen, ranging from 0.8 to 4.6 percent, whereas hybrids of *D. sandwicense* × *D. intortum* had a high percentage of aborted pollen, ranging from 2.9 to 36.8 percent. This would indicate that the relationship between *D. sandwicense* and *D. uncinatum* is closer than that between *D. sandwicense* and *D. intortum* or that there is some form of incompatibility occurring. In the three-species hybrids, the percentage of aborted pollen varied from 0.4 to 62.1 percent. Fifteen hybrids from the combination *(D. sandwicense* ♀ × *D. intortum* ♂*)* ♀ × *(D. uncinatum* ♀ × *D. intortum* ♂*)* ♂ averaged 10.9 percent aborted pollen; the 31 hybrids of the combination of *(D. sandwicense* ♀ × *D. intortum* ♂*)* ♀ × *(D. sandwicense* ♀ × *D. uncinatum* ♂*)* ♂ averaged 19.3 percent aborted pollen. Greater incompatibilities were evident in the latter cross than in the former. Pod set in the three *Desmodium* species was statistically highly significantly and negatively correlated with the percentage of aborted pollen, $r = -0.736$, $df = 21$.

## Genetic studies among the three species

**Stem color:** All plants of *D. uncinatum* had red stems; *D. sandwicense* had red or green stems; and *D. intortum* had red, brown, or green stems, depending upon the plant. In determining genetic ratios, the stems were segregated into two classes: (i) red or brown or (ii) green. There were differences in intensity of the red stem color. Results of the stem color studies are presented in Table 5. These indicate that stem color in the three species is controlled by a single gene with dominance for red or brown stems and recessiveness for green stems. Crosses among red- or brown-stemmed parents produced red- or brown-stemmed $F_1$ progenies. $F_2$ progenies were also red or brown stemmed. Crosses among green-stemmed parents produced green-stemmed $F_1$ progenies and the $F_2$ progenies were also green stemmed. Crosses among red- or brown-stemmed and green-stemmed parents produced only red- or brown-stemmed $F_1$ plants. The $F_2$ progenies segregated into red- or brown-stemmed and green-stemmed plants, with 3:1 ratios whose chi-square values had probabilities from 0.25 to 1.00. From the three-species hybrids, the $F_1$ progenies were red or brown stemmed. The $F_2$ progenies segregated into red- or brown-stemmed and green-stemmed plants, with 3:1 ratios whose chi-square values had small and nonsignificant probabilities. These results are in agreement with Park and Rotar *(5)* who found that red stem color in *D. sandwicense* was dominant to green. In interspecific hybrids, McWhirter *(3)* and Hutton and Gray *(2)* indi-

**TABLE 5. Stem color of the parental *Desmodium* species, their $F_1$ hybrids and $F_2$ progenies, and chi-squares for goodness of fit to 3:1 ratios of red to green stem**

| Cross | $F_1$ or $S_1$ stem color | No. of $F_2$ progenies | | | | Chi-square | P |
|---|---|---|---|---|---|---|---|
| | | Observed | | Calculated | | | |
| | | Red or brown | Green | Red or brown | Green | | |
| Crosses among red or brown | | | | | | | |
| S21 × U22 | red or brown | 35 | | 35 | | | |
| S21 × U82 | red or brown | 36 | | 36 | | | |
| Crosses among green | | | | | | | |
| S11 × I33 | green | | 34 | | 34 | | |
| S11 × I43 | green | | 36 | | 36 | | |
| S31 × I43 | green | | 36 | | 36 | | |
| Crosses of red or brown × green | | | | | | | |
| S11 × U22 | red or brown | 27 | 9 | 27 | 9 | 0.00 | 1.00 |
| S21 × U72 | red or brown | 25 | 11 | 27 | 9 | 0.60 | 7.25 |
| S31 × U82 | red or brown | 12 | 3 | 11.25 | 3.75 | 0.20 | 7.50 |
| Three-species hybrids | | | | | | | |
| (S51 × I23) × (U52 × I33) | red or brown | 25 | 11 | 27 | 9 | 0.60 | 0.25 |
| (S21 × I53) × (U52 × I33) | red or brown | 20 | 5 | 18.75 | 6.25 | 0.34 | 0.50 |
| (S21 × U82) × (S21 × I53) | red or brown | 31 | 5 | 27 | 9 | 2.36 | 0.10 |

**TABLE 6. Tests of independence between internode length and growth habit of plants from the crosses (*D. sandwicense* ♀ × *D. intortum* ♂) ♀ × (*D. sandwicense* ♀ × *D. uncinatum* ♂) ♂**

| Cross | Internode length (cm) | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | 1–2 | 2–3 | 3–4 | 4–5 | 5–6 | 6–7 | 7–8 | 8–9 | Total |
| Spreading f | 3 | 2 | 5 | 11 | 6 | 4 | 1 | 2 | 34 |
| F | 2.66 | 2.12 | 5.84 | 8.50 | 8.50 | 4.25 | 1.06 | 1.06 | |
| f-F | 0.34 | —0.12 | —0.84 | 2.50 | —2.50 | —0.25 | —0.06 | 0.94 | |
| Intermediate f | 0 | 0 | 0 | 1 | 10 | 3 | 1 | 0 | 15 |
| F | 1.17 | 0.94 | 2.58 | 3.75 | 3.75 | 1.88 | 0.46 | 0.46 | |
| f-F | —1.17 | —0.94 | —2.58 | —2.75 | 6.25 | 1.12 | 0.54 | —0.46 | |
| Upright f | 2 | 2 | 6 | 4 | 0 | 1 | 0 | 0 | 15 |
| F | 1.17 | 0.94 | 2.58 | 3.75 | 3.75 | 1.88 | 0.46 | 0.46 | |
| f-F | 0.83 | 1.06 | 3.42 | 0.25 | —3.75 | 0.88 | —0.46 | —0.46 | |
| Total | 5 | 4 | 11 | 16 | 16 | 8 | 2 | 2 | 64 |

Chi-square = 32.79, df = 14, $P < 0.01$.

cated that stem color was controlled by a single pair of genes with dominance for the red stem.

**Internode length:** Measurements of internode lengths are presented in Fig. 2. These varied among the parents as follows: 6.0 to 8.0 cm for *D. uncinatum*, 3.4 to 6.3 cm for *D. intortum*, and 1.1 to 2.9 cm for *D. sandwicense*. Internode lengths of the two-species hybrids were intermediate to those of the parents. In the three-species hybrids, many gradations from one extreme to the other of the three parents were observed. Results indicated that the genetic behavior of internode length of these species was controlled by multiple genes.

A test of independence of internode length with stem growth habit of 64 three-species hybrids from the combination *(D. sandwicense* ♀ × *D. intortum* ♂*)* ♀ × *(D. sandwicense* ♀ × *D. uncinatum* ♂*)* ♂ is presented in Table 6. Results indicated that these two characteristics are closely related. It was concluded that plants with spreading and intermediate growth habits had significantly longer internodes than plants with an upright growth habit.

**Silver marking on midrib of leaflet:** There are three types of leaflet markings observable in the leaflets of these species. One is a reddish-brown fleck scattered throughout the leaflets of certain *D. intortum* plants. Another is the shiny appearance of the midrib and veins of the leaflet, observed only in *D. intortum*. This trait varies in its expression from nonappearance to nearly covering the leaflet. The third is the silver-gray marking on the midrib of the leaflet, which is observed in all three species but not in all plants.

All plants of *D. sandwicense* and *D. uncinatum* used in this study had silver marking on the leaflet midrib. Four of the *D. intortum* plants were marked. Results of midrib leaflet marking studies are presented in Table 7. In crosses among marked plants, the $F_1$ progenies were marked and only marked $F_2$ progenies were produced. In crosses among marked and nonmarked plants, only marked $F_1$ progenies were observed, and marked and nonmarked $F_2$ progenies were observed to segregate into 3 (marked) :1 (nonmarked) ratios whose chi-squares had probabilities varying from 0.10 to 0.75, depending upon the crosses. From this evidence it was concluded that the silver-gray marking on the midrib of the leaflet was controlled by a single pair of genes, with dominance for the marked and recessiveness for the nonmarked. This was in agreement with Park and Rotar *(5)* and with Hutton and Gray *(2)* who concluded that this leaflet marking was controlled by a single pair of genes.

**Rugose leaflet:** Plants having this characteristic had distinctively shaped leaflets which were also curved backward. This characteristic was observed only among the $F_2$ progenies from the combination *(D. sandwicense* ♀ × *D. intortum* ♂*)* ♀ × *(D. uncinatum* ♀ × *D. intortum* ♂*)* ♂, where *D.*

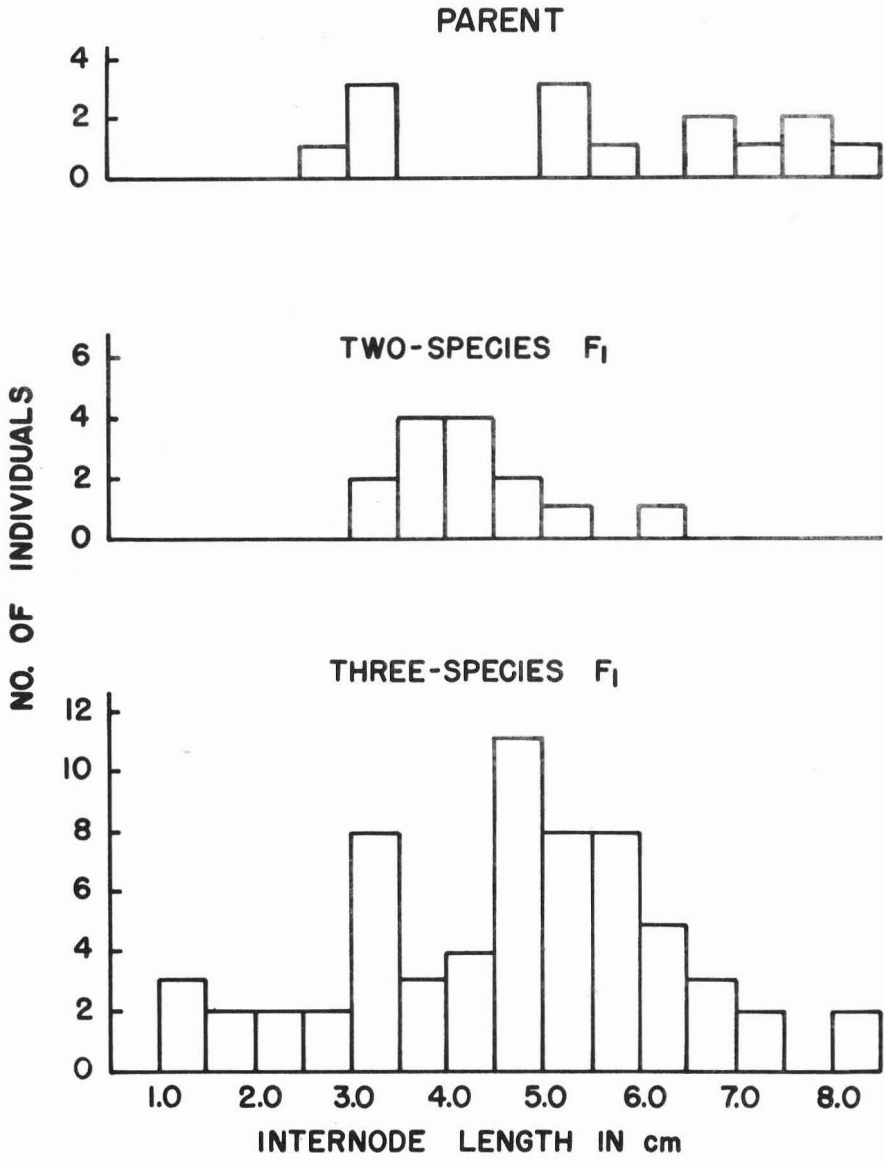


FIG. 2. Internode lengths of parents and two- and three-species hybrids of *D. sandwicense*, *D. intortum*, and *D. uncinatum*.

**TABLE 7. Inheritance of leaflet marking on the midrib of the leaflet of three *Desmodium* species**

| Cross | $F_1$ hybrids and $F_2$ progenies | | | | Chi-square for 3:1 ratio | P |
|---|---|---|---|---|---|---|
| | Observed | | Calculated | | | |
| | Marked | Nonmarked | Marked | Nonmarked | | |
| Marked × marked | | | | | | |
| S11 × U22 | 36 | 0 | 36 | 0 | | |
| S21 × U22 | 35 | 0 | 35 | 0 | | |
| S21 × U82 | 36 | 0 | 36 | 0 | | |
| S31 × U22 | 36 | 0 | 36 | 0 | | |
| S31 × U82 | 15 | 0 | 15 | 0 | | |
| Marked × nonmarked | | | | | | |
| S11 × I33 | 27 | 7 | 25.5 | 8.5 | 0.36 | 0.50–0.75 |
| S11 × I33 | 30 | 6 | 27.0 | 9.0 | 1.33 | 0.10–0.25 |
| S31 × I43 | 29 | 7 | 27.0 | 9.0 | 0.59 | 0.25–0.50 |
| Three-species hybrids | | | | | | |
| (S51 × I23) × (U52 × I33) | 34 | 2 | 36 | 0 | | |
| (S21 × I52) × (U52 × I33) | 24 | 1 | 25 | 0 | | |
| (S21 × U82) × (S21 × I52) | 36 | 0 | 36 | 0 | | |
| (S21 × I23) × (S11 × U22) | 20 | 0 | 36 | 0 | | |
| (S21 × I53) × (S31 × U22) | 17 | 8 | 18.75 | 6.25 | 0.65 | 0.10–0.25 |

*intortum* occurred twice in the combination. Four out of 19 plants from this cross had the rugose leaflet characteristic. This produces a fair fit for a 3 (normal) :1 (rugose leaflet) ratio with a chi-square of 3.55 the probability of which falls between 0.10 and 0.05.

These four plants flowered during the period November to April as did *D. uncinatum*. Their seed set was good, and their percentage of shriveled seed was low, 5.0 to 9.5 percent. The 1000-seed weights ranged from 3.73 to 3.76 grams, midway between *D. uncinatum* and *D. sandwicense*. Their internodes were short, with length varying from 3.1 to 5.5 cm. The stems were densely covered with long, uncinate hairs. Stem color varied from green to red. Leaflet-size indices varied from 12 to 16. The leaflets, covered with long, dense hairs, varied in color from green to brown. The rugose character was not observed in any of the parent plants nor was it observed among any of the $F_1$ hybrids from the three species.

**Raceme length:** Raceme length is one of the distinctive features of the three species. As the florets open from base to apex, the raceme elongates and reaches maximum length when the terminal flowers open. The raceme decreases in size as the seeds mature. Average raceme length among the three parent species was 24.4, 14.4, and 12.1 cm for *D. uncinatum*, *D. sandwicense*, and *D. intortum*, respectively. Raceme lengths of $F_1$ hybrids among the three species were as short as those of the short raceme parent. In the $F_1 \times F_1$ hybrids, raceme lengths exceeded the range of the parents. Results obtained were difficult to interpret and no attempt was made to determine the genetic behavior of raceme length.

Raceme length was statistically highly significantly correlated with length of flowering period per raceme and with total number of flowers per raceme, $r = 0.651$, df $= 22$, and $r = 0.542$, df $= 22$, respectively. This is expected, inasmuch as raceme length will influence the number of flowers that are present per raceme, and the length of the flowering period per raceme is a function of the number of flowers per raceme.

**Seed weight:** There were distinct differences in 1000-seed weights among the three species and their hybrids. Results are presented in Table 8. *D. uncinatum* had the heaviest 1000-seed weights, 4.08 grams/1000 seeds; *D. sandwicense* had the next heaviest, 3.54 grams/1000 seeds; and *D. intortum* the lightest, 1.68 grams/1000 seeds. These results are in agreement with Rotar and Urata *(6)*. Differences in seed size were obtained among the hybrids from the three species. None of the hybrids had 1000-seed weights which exceeded the means of the heaviest or lightest parents.

Percentage of shriveled seed from the $F_1$ generation of two-species crosses was not significantly greater than that of the parents ($t = 1.80$, df $= 24$,

**TABLE 8. Thousand-seed weights and percentage of shriveled seed from three *Desmodium* species and their $F_1$ and $F_1 \times F_1$ hybrids**

| Plant no. | 1000-seed weight (grams) | Shriveled seed (percent) |
|---|---|---|
| Parent | | |
| *D. sandwicense* | | |
| S31 | 3.54 | 2.5 |
| *D. uncinatum* | | |
| U12 | 3.99 | 2.5 |
| U32 | 4.01 | 3.0 |
| U42 | 4.08 | 2.5 |
| *D. intortum* | | |
| I33 | 1.68 | 4.5 |
| I63 | 1.99 | 5.0 |
| $F_1$ of two species | | |
| S11 × U22 | 2.81 | 5.0 |
| S11 × U62 | 3.46 | 4.5 |
| S21 × U82 | 3.00 | 3.0 |
| S31 × U22 | 3.31 | 6.0 |
| S31 × U42 | 3.35 | 4.5 |
| S51 × U42 | 3.25 | 4.0 |
| S31 × I42 | 2.53 | 4.0 |
| $F_1$ of three species | | |
| (S11 × I23) × (S31 × U22) a | 2.97 | 15.5 |
| b | 2.91 | 12.0 |
| c | 3.06 | 8.0 |
| (S11 × I23) × (S41 × U22) | 2.54 | 14.0 |
| (S11 × I23) × (S11 × U22) | 2.06 | 17.5 |
| (S21 × I23) × (S11 × U22) | 2.52 | 22.0 |
| (S21 × I23) × (S21 × U72) | 2.48 | 16.0 |
| (S21 × I53) × (S31 × U22) | 1.74 | 19.0 |
| (S51 × I23) × (S11 × U62) | 2.45 | 9.5 |
| (S21 × U82) × (S21 × I53) | 3.28 | 9.5 |
| (S21 × I53) × (U52 × I33) | 3.73 | 9.5 |
| (S21 × I53) × (U52 × I33) | 3.76 | 5.0 |

$P > 0.10$). Percentage of shriveled seed from the three-species hybrids was significantly greater than that of the parent ($t = 10.53$, df $= 38$, $P < 0.01$). Although the percentage of shriveled seed was not high in the $F_1$ generation from two-species hybrids, there was a trend in this direction. Evidently the

three species are still quite closely related, and it is only when they are combined that larger numbers of incompatibilities occur. This same trend was observed in percentage of pollen abortion, Table 4.

## SUMMARY AND CONCLUSIONS

These experiments were undertaken to study the breeding and flowering behavior, genetics, and relationships among the three species: *D. sandwicense*, *D. uncinatum*, and *D. intortum*. *D. sandwicense* was insensitive to daylength and flowered all year. *D. uncinatum* and *D. intortum* flowered only in the short-day season, from October through April, and from December through March, respectively.

In controlled crosses, percentage of pod formation was low, an average of 9.2 percent for any two of the three species crossed together, and an average of 4.9 percent when $F_1$ progenies of two species were combined to obtain three-species crosses. When *D. sandwicense* was used as the female parent in crosses with *D. uncinatum* or *D. intortum*, percentage of pod formation was relatively high, 15.3 percent and 14.2 percent, respectively. When *D. sandwicense* was used as the male-parent, percentage of pod formation was very low, 2.4 percent.

Germination percentages of hybrid seeds were low, 54 percent for seeds from two-species crosses and 45 percent for $F_1 \times F_1$ seeds. Hybrids of *D. sandwicense* and *D. uncinatum* had an average of 2.5 percent aborted pollen; hybrids of *D. uncinatum* and *D. intortum* had an average of 22.1 percent aborted pollen.

Stem color of the species was confirmed to be under the control of a single pair of genes with red or brown dominant to green.

Midrib leaflet marking was observed in all three species. The marking was confirmed to be due to a single pair of genes with dominance for the marked and recessiveness for the unmarked leaflets.

Rugose leaflet character was found in the three-species hybrids of the combination *(D. sandwicense* ♀ × *D. intortum* ♂*)* ♀ × *(D. uncinatum* ♀ × *D. intortum* ♂*)* ♂. This appeared to be due to a single pair of genes with the rugose leaflet recessive to the normal leaflet.

Raceme length, internode length of stems, and 1000-seed weight appeared to be controlled by multiple genes, inasmuch as those of the $F_1$ plants were intermediate to their parents and those of the $F_2$ progenies were nearly the same as or exceeded the ranges of the parents.

*D. intortum* and *D. uncinatum* had leaflet-size indices (leaflet length × leaflet width) which were approximately twice as large as those of *D. sandwicense*.

Percentages of pods set from crosses were negatively correlated with the mean monthly temperatures during the short-day season. The higher the mean monthly temperatures the lower the percentages of pods set.

## LITERATURE CITED

1. HUTTON, E. M. 1960. Flowering and pollination in *Indigofera spicata, Phaseolus lathyroides, Desmodium uncinatum* and some other tropical pasture legumes. Empire J. Exp. Agr. 28:235–243.

2. _____ and S. G. GRAY. 1967. Hybridization between the legumes *Desmodium intortum, D. uncinatum,* and *D. sandwicense.* J. Australian Inst. Agr. Sci. 33:122–123.

3. MCWHIRTER, K. S. 1963. Annual Report 1962–63. C.S.I.R.O. Division of Tropical Pastures, Brisbane, Queensland, Australia. P. 32.

4. _____ 1969. Cytoplasmic male sterility in *Desmodium.* Australian J. Agr. Res. 20:227–241.

5. PARK, SOON JAI, and P. P. ROTAR. 1968. Genetic studies in Spanish clover, *Desmodium sandwicense* E. Mey. I. Inheritance of flower color, stem color, and leaflet markings. Crop Sci. 8:467–470.

6. ROTAR, P. P., and U. URATA. 1966. Some agronomic observations in *Desmodium* species: seed weights. Hawaii Agr. Exp. Sta. Tech. Prog. Rep. No. 147. 14 p.

7. _____ and _____ 1967. Cytological studies in the genus *Desmodium;* some chromosome counts. Amer. J. Bot. 54:1–4.

8. _____ S. J. PARK, A. BROMDEP, and U. URATA. 1967. Crossing and flowering behavior in Spanish clover, *Desmodium sandwicense* E. Mey., and other *Desmodium* species. Hawaii Agr. Exp. Sta. Tech. Prog. Rep. No. 164. 13 p.

9. WANG, C. C. 1961. Growth, flowering and forage production of some grasses and legumes in response to different photoperiods. J. Agr. Assoc. China. N. S. 36:27–52.

## APPENDIX

### Morphological observations on the parent plants

Stem, leaflet, flower, pod, and seed characteristics are presented in tabular form in Appendix Table A.

**Stem variation:** Stem growth habit varied from upright to decumbent. Stem cross section varied from round, obtuse angled to acute angled. Stem internode length varied from 2.9 to 8.1 cm, depending upon the species and the environmental conditions. Stem color varied from light green, green, brown, and red to dark red, depending upon the plant or the species or both. Pubescence differed greatly in length, texture, density, and shape from plant to plant; some plants were also glandular.

*D. sandwicense:* The five plants of *D. sandwicense* used in this study had an upright habit of growth, and the stems were round in cross section. Stem internode length varied from 2.9 to 3.2 cm, the shortest of the three species. Stem color varied from green to red, depending upon the plant. *D. sandwicense* had few short stem hairs, which were sometimes glandular.

*D. uncinatum:* All eight plants of *D. uncinatum* had decumbent and spreading growth habit. The stems were round in cross section. The stem internode length varied from 6.2 to 8.1 cm. All of the stems of the *D. uncinatum* were either red or brown. *D. uncinatum* had long, dense, uncinate stem hairs.

*D. intortum:* The stems of the six *D. intortum* plants were multifarious in morphology. Growth habit varied from upright to spreading, and cross section of stems from nearly round to acute angled. Internode length varied from 3.4 to 6.4 cm. The stems' pubescence showed great diversity in amount and texture. Stem color varied from red to green.

**Leaf variation:** Leaves of the three species were trifoliate. Leaflet size varied from species to species; mature leaves were larger in summer, smaller in winter. The center leaflet was always larger than the two lateral leaflets. Leaflet form varied from lanceolate elliptic to ovate, depending upon the species or the plant or both, within the species. Leaflet apices were either acuminate or acute and leaflet bases were either truncate or obtuse.

Leaflet margins were entire and often provided with a row of hairs. The silver leaflet marking on the midrib of the leaflet was observed in each species; although some of the parent plants of *D. intortum* were unmarked, carrying only a brownish fleck on the leaflets, and others were entirely unmarked in any fashion. Leaf color varied from light green to green, and brown. Pubescence differed considerably in length, density, and shape between species and among

plants within species. The lower leaf surfaces usually were more pubescent than the upper surfaces.

*D. sandwicense:* Leaflet-size indices (leaflet length × leaflet width) of *D. sandwicense* ranged from 8 to 12, and the leaflet length to width ratios varied from 1.74 to 1.89. The *D. sandwicense* plants in this study had green leaflets. Leaflet form was lanceolate; leaf apices were acute; leaflet bases were obtuse, and constricted abruptly to petiolules.

*D. uncinatum:* Leaflet-size indices of *D. uncinatum* varied from 21 to 27; the leaflet length to width ratios varied from 1.77 to 1.86. All of the *D. uncinatum* plants used in this study had light-green leaves with a silver marking along the midrib of each leaflet. Leaflet form was elliptic. The width was about one-half the length. Leaflet apices were acute; leaflet bases were truncate.

*D. intortum:* Leaflet-size indices varied from 18 to 32 and leaflet length to width ratios varied from 1.49 to 1.69. *D. intortum* plants varied in leaf color and leaflet marking. I23 had brown leaflets and a silver leaflet marking along the leaflet midrib. The other plants were green and lacked the midrib marking. Leaflet form was ovate, with the broadest part below the middle. Leaflet apices were acute to acuminate and leaflet bases were truncate.

**Flowers and flowering habit:** Flower color varied from nearly white, through pink and red, and light bluish white, depending upon the species and plant or both. Raceme length varied among the three species. About 2 weeks were required for flowering to be completed on any one raceme; flower number per raceme varied among species and among plants within species.

The three species flowered in a similar manner. The day before the flowers opened, the closed petals expanded and were observed projecting beyond the sepals. In the late afternoon of the day before the flowers opened, the anthers were white or very light yellow. By about 8 PM, the anthers had turned yellow, indicating pollen maturation. About midnight the anthers had dehisced. By dawn the petals had completely expanded but the flower was still closed. Shortly after daylight, the standard petal became erect and the flower was receptive to pollinating insects. Pollen was usually discharged in a cloud around the stigma when the flower was tripped. The stigma did not strike the standard petal as in alfalfa. Once tripped, the flowers quickly wilted, and the standard petals were folded over the stigmas in a short time. Two flowers usually occurred at each node, and the pairs were arranged in a spiral on the raceme. Only a few flowers opened per raceme per day.

*D. sandwicense:* Flower color of *D. sandwicense* varied from nearly white, through pink to red, depending upon the plant. Racemes were about 15 cm

long with about 50 flowers per raceme. Flowering period per raceme averaged 8.8 days with 5.5 flowers opened per raceme per day.

*D. uncinatum:* Flower color of the six *D. uncinatum* plants was a bluish pink with white. Racemes were from 21 to 26 cm long with about 47 flowers per raceme. Flowering period per raceme varied from 10 to 14 days with 3 to 4 flowers opened per raceme per day.

*D. intortum:* Flower color of *D. intortum* varied from pink to deep red, depending upon the plant. Racemes varied from 10.8 to 14.6 cm in length with 27.8 to 43.4 flowers per raceme. Flowering period per raceme varied from 5.2 to 8.4 days with 4.3 to 5.3 flowers opened per raceme per day.

**Pods and seeds:** The three species studied produced serrated pods with 4 to 12 seeds per pod. The pods were covered with dense, uncinate hairs. The seeds were kidney shaped, from 1.5 to over 3.0 mm long, and from 1.0 to over 2.0 mm wide. Seed weight per 1000 seeds varied among the three species.

*D. sandwicense:* Percentage of pod set from open pollination was 47.2 percent. Each pod produced an average of 6.3 seeds. The 1000-seed weight was 3.54 grams. Seed color was light brown. Seed germination was about 99 percent for normal, mature seeds.

*D. uncinatum:* Percentage of pod set from open pollination varied from 16.9 to 32.6 percent. Each pod produced about 4 seeds. The 1000-seed weight was 4 grams. Seed color varied from green to light brown. Seed germination was about 96 percent for normal, mature seeds.

*D. intortum:* Percentage of pod set from open pollination varied from 23.3 to 51.1 percent. Seeds per pod varied from 4.4 to 5.7. The 1000-seed weight was 1.8 grams. Seed color was brown. Seed germination was about 96 percent for normal, mature seeds.

**TABLE A. Morphological characteristics, flowering behavior, number of seeds per pod, and percentage seed germination of the parent plants of three *Desmodium* species**

| Parent plant no. | Stem characteristics | | | | | |
|---|---|---|---|---|---|---|
| | Color | Internode length (cm) | Cross section | Pubescence | Growth habit | Vigor |
| *D. sandwicense* | | | | | | |
| S11 | green | 3.2 | round | few, short | upright | fair |
| S21 | red | 3.0 | round | few, short | upright | fair |
| S31 | green | 2.9 | round | few, short | upright | fair |
| *D. uncinatum* | | | | | | |
| U12 | red | 6.2 | round | dense, hooked | spreading | good |
| U22 | red | 7.0 | round | dense, hooked | spreading | good |
| U32 | red | 6.6 | round | dense, hooked | spreading | good |
| U42 | red | 7.5 | round | dense, hooked | spreading | good |
| U62 | red | 7.3 | round | dense, hooked | spreading | good |
| U82 | red | 7.3 | round | dense, hooked | spreading | good |
| *D. intortum* | | | | | | |
| I13 | red | 4.1 | obtuse angled | dense, hooked | intermediate | good |
| I23 | brown | 3.4 | obtuse angled | dense, hooked | spreading | fair |
| I33 | green | 4.8 | obtuse angled | dense, hooked | upright | good |
| I43 | green | 6.3 | acute angled | dense, hooked | spreading | good |
| I53 | green | 4.7 | acute angled | dense, hooked | intermediate | vigorous |
| I63 | green | 5.5 | acute angled | few, short | upright | good |

| Parent plant no. | Inflorescence characteristics: Flower color[1] | Raceme length (cm) | No. of flowers per raceme | Flowering period per raceme (days) | Flowers opened per day | Pods set[2] (percent) | Seeds per pod | 1000-seed weight (grams) | Seed germination (percent) |
|---|---|---|---|---|---|---|---|---|---|
| *D. sandwicense* | | | | | | | | | |
| S11 | Venetian pink | — | — | — | — | — | — | — | — |
| S21 | fuschine pink | — | — | — | — | — | — | — | — |
| S31 | dawn pink | 14.9 | 50.8 | 8.8 | 5.8 | 47.2 | 6.3 | 3.54 | 99 |
| *D. uncinatum* | | | | | | | | | |
| U12 | mallow purple | 24.5 | 47.8 | 11.3 | 4.2 | 27.6 | 4.4 | 3.99 | 97 |
| U22 | mallow purple | — | — | — | — | — | — | 4.05 | — |
| U32 | mallow purple | 26.0 | 46.8 | 10.8 | 4.3 | 16.9 | 4.5 | 4.01 | 95.5 |
| U42 | mallow purple | 21.2 | 48.0 | 12.0 | 4.0 | 21.3 | 4.1 | 4.08 | — |
| U62 | mallow purple | 22.6 | 46.0 | 14.0 | 3.3 | 32.6 | 3.9 | 4.11 | 96.5 |
| U82 | mallow purple | — | — | — | — | — | — | — | — |
| *D. intortum* | | | | | | | | | |
| I13 | phlox pink | — | — | — | — | — | — | — | — |
| I23 | roseine purple | 10.8 | 27.8 | 5.2 | 5.3 | 23.3 | 5.7 | — | — |
| I33 | phlox purple | — | — | — | — | — | — | — | — |
| I43 | phlox pink | 14.6 | 43.4 | 8.4 | 5.2 | 51.1 | 4.4 | 1.82 | 96.0 |
| I53 | —— | 11.0 | 30.8 | 7.2 | 4.4 | 49.6 | 4.7 | 1.78 | 97.0 |
| I63 | —— | — | — | — | — | — | — | 1.99 | 96.5 |

[1] Flower color according to British Colour Council, Horticultural Colour Chart, Vol. I and II. 1940. Henry Stone & Son, Ltd., England.
[2] Pods set under open pollination.

—Continued

**TABLE A. Morphological characteristics, flowering behavior, number of seeds per pod, and percentage seed germination of the parent plants of three *Desmodium* species—Continued**

| | Leaflet characteristics | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | | | | Pubescence | | | | |
| Parent plant no. | Color | Form | Apex | Base | Upper surface ($cm^2$) | Lower surface ($cm^2$) | Length × width ($cm^2$) | Ratio of length to width | Silver leaflet marking present |
| *D. sandwicense* | | | | | | | | | |
| S11 | green | lanceolate | acute | obtuse | 72 | 1331 | 10.51 | 1.74 | yes |
| S21 | green | lanceolate | acute | obtuse | 53 | 402 | 8.04 | 1.76 | yes |
| S31 | green | lanceolate | acute | obtuse | 266 | 1413 | 12.58 | 1.89 | yes |
| *D. intortum* | | | | | | | | | |
| U12 | light green | elliptic | acute | obtuse | 135 | 258 | 26.96 | 1.86 | yes |
| U22 | light green | elliptic | acute | obtuse | 188 | 322 | 21.33 | 1.80 | yes |
| U32 | light green | elliptic | acute | obtuse | 168 | 321 | 24.91 | 1.84 | yes |
| U42 | light green | elliptic | acute | obtuse | 177 | 282 | 22.96 | 1.81 | yes |
| U62 | light green | elliptic | acute | obtuse | 146 | 298 | 22.63 | 1.79 | yes |
| U82 | light green | elliptic | acute | obtuse | 165 | 307 | 21.66 | 1.77 | yes |
| *D. intortum* | | | | | | | | | |
| I13 | green | ovate | acute | truncate | 257 | 1098 | 21.88 | 1.62 | yes |
| I23 | green | ovate | acute | truncate | 185 | 405 | 18.44 | 1.59 | yes |
| I33 | green | ovate | acuminate | truncate | 111 | 97 | 32.01 | 1.49 | no |
| I43 | green | ovate | acuminate | truncate | 65 | 220 | 21.87 | 1.58 | no |
| I53 | green | ovate | acuminate | truncate | 170 | 370 | 26.45 | 1.56 | no |
| I63 | green | ovate | acuminate | truncate | 88 | 438 | 21.77 | 1.55 | no |

UNIVERSITY OF HAWAII
COLLEGE OF TROPICAL AGRICULTURE
HAWAII AGRICULTURAL EXPERIMENT STATION
HONOLULU, HAWAII

---