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Allometrical growth of the quantitative characters of plants I. Measurement of leaf size and shape

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Abstract. We developed a method to measure and track changes in the size and shape of individual leaves throughout the life of a plant, using photographs and a digital tablet. We studied the estimation of leaf shape by principle component analysis of a correlation matrix of leaf width — or under allometrical growth, between leaf length and leaf width — and found that a linear function of allometrical growth between leaf length and width gives a good description of leaf shape. We also studied a growth model of leaf area and obtained an exponential function which is a good expression of the growth of leaf area. From an analysis of principle component and linear function, there are two indicators of leaf shape — discriminator and index. Comparing the two indicators, the leaf-shape index gives a more reasonable expression of leaf shape. Our test subjects were two cultivated tobaccos with dissimilar shapes and sizes of leaves, and their four progeny.

Keywords: Allometrical growth; Leaf shape; Leaf-shape discriminator; Leaf-shape index; Leaf size; Principle component analysis; Tobacco.

Introduction

The growth rate of a biological body is determined by different organs. Different relative growth rates of organs produce different shapes. We often use allometrical growth to study the relationship among the various parts of an organism, a characteristic of an individual, or various organis of an individual during its growing period. (Huxley, 1924). This enables biologists to examine the growth gradients within an organism or a character, to understand its forms, functions, and biological niches, as well as the evolutionary processes which bring the organism to its present form.

The allometric formula proposed by Huxley (1924) is based on the assumption that the relative growth rates of two organs maintain a constant ratio throughout their growth periods. Although it measures only part of the growth of an organism and is approximate, it is simple and has proved to be a remarkably useful equation in many practical applications (Gould, 1966).

Recently, multivariate extensions of the simple allometry formula, such as multivariate growth and principle component analysis (Griffiths and Sandland, 1984, 1985; Sampson and Siegel, 1985a, b; Turner, 1978; Veitch, 1978) have become important subjects in this field. Most concern the extention of the simple allometrical equation to more than two dimensions, though some concern the effect of genetic factors and their mutual relationship. This study considers an allometrical model involving genetic parameters for studying the genetic mechanism of allometrical growth among various organs or characters of an individual. Future experiments will be conducted to test the reliability and applicability of the proposed method, using tobacco leaves as an example. In this report, we present the measurement of leaf sizes and shapes, as well as their relationships at different locations on the plant, for six generations.

Materials and Methods

Materials

Two different cultivated tobaccos with dissimilar leaf shapes and size were selected as test subjects: Turkish var. Samsum 15A (P_1, small elongated leaf), native fluecured tobacco var. TT5 (P_2, wide oval leaf), as well as their progeny (F_1, B_1, B_2, and F_2). Ten plants of P_1, P_2, and F_1, 50 plants of B_1 and B_2, and 100 plants of F_2 generations were planted. Photographs were taken every two or three days for three different layers of leaves — the lower, middle, and top. We measured five to six times during the growth of each leaf.

Methods

The width, length, and area of a photographic image of a leaf were measured with a digital tablet and the DGL software package on a HP 1000 computer. We studied the simple correlations between leaf length and width.

The relationship between leaf length (u_j) and width (w_{ij}) at the ith width (i=1, 2, . . . 8) for the jth growth time (j=1, 2, . . . 5 or 6) is assumed as (Huxley, 1924; Hopkins, 1960; Sprent, 1972):

$$w_{ij} = \alpha_i u_j^{\beta i} \qquad (1)$$

where αi and βi are parameters that will be estimated by the log linear regression equation:

$$\ln w_{ij} = \ln \alpha_i + \beta_i \ln u_j \qquad (2)$$

Note that β_i is the ith allometrical coefficient between leaf length (u_j) and the ith width (w_{ij}) at various growth times. The absolute growth rate of leaf width is:

$$\frac{dw_{ij}}{dt_j} = \beta_i w_{ij} \left(\frac{du_j}{dt_j}\right)\left(\frac{1}{u_j}\right) \qquad (3)$$

If Z_j is the sum of two widths (i and i'th), thus

$$Z_j = w_{ij} + w_{i'j} \qquad (4)$$

then

$$\frac{dZ_j}{dt_j} = \frac{dw_{ij}}{dt_j} + \frac{dw_{i'j}}{dt_j} = (\beta_i w_{ij} + \beta_{i'} w_{i'j})\left(\frac{du_j}{dt_j}\right)\left(\frac{1}{u_j}\right) \qquad (5)$$

On the other hand, we can also express $\beta_{i''}$ for the allometrical coefficient between leaf length u_j and the sum of two widths Z_j by the form of

$$Z_j = \alpha_{i''} u_j^{\beta i''} \qquad (6)$$

then;

$$\frac{dZ_j}{dt_j} = \beta_{i''} Z_j \left(\frac{du_j}{dt_j}\right)\left(\frac{1}{u_j}\right) \qquad (7)$$

therefore, from equations (5) and (7), we obtained

$$\beta_{i''} Z_j = (\beta_i w_{ij} + \beta_{i'} w_{i'j}) \qquad (8)$$

hence

$$\beta_{i''} = \frac{\beta_i w_{ij} + \beta_{i'} w_{i'j}}{Z_j} = \frac{\beta_i w_{ij} + \beta_{i'} w_{i'j}}{w_{ij} + w_{i'j}} \qquad (9)$$

If Z_j also represents the sum of all widths, then we have

$$Z_j = w_{1j} + w_{2j} + \ldots + w_{ij} + \ldots + w_{nj} = \Sigma w_{ij} \qquad (10)$$

and

$$\beta_z = \frac{\Sigma \beta_i w_{ij}}{\Sigma w_{ij}} \qquad (11)$$

where the summation of w_{ij} represents the leaf size, and β_z is the leaf shape index.

One method used to estimate parameters of leaf shape (shape discriminator) is principle component analysis, which uses a correlation matrix of leaf widths to find the size and shape component (Griffiths and Sanland, 1985; Sampson and Siegel, 1985a,b).

Results and Discussion

Leaf Size

The areas of ten leaves were measured with a leaf-area meter and a digital tablet, and regression analysis was used to check the accuracy of the digital tablet measurments. Figure 1 shows the relationship between

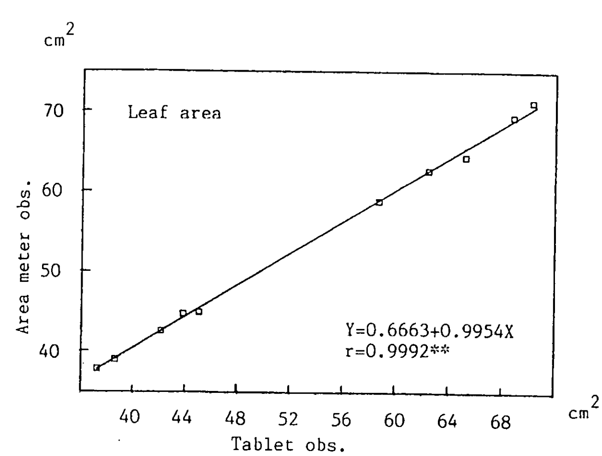

Fig. 1. The reltionship between measurements with **X**) a digital tablet, and **Y**) an area meter.

the readings from the digital tablet and leaf area meter; a high correlation (r = 0.9992**) was obtained. This shows that the digital tablet measures the actual area of a leaf. Similar results for the leaf width and length were obtained using the same method.

Because the growth period of a leaf is short, a lower leaf and a top leaf were measureed six times, and a middle leaf five times. In this case, four-parameter Richards' growth formulae (Richards, 1969) are not applicable to fitting the growth of leaf size. The exponential function:

$$A_j = \exp(b_0 + b_1 t_j + b_2 t_j^2)$$

can be used for fitting, where A_j is the leaf area at the jth observation time. The estimated value of the leaf size during the growth period is shown in Figure 2. The leaf area of P_2 was larger than that of P_1 for three different leaves, and the leaf areas of F_1 and F_2 hybrids were between those of P_1 and P_2 parents. B_1 and B_2 generations had similar size leaves as did P_1 and P_2 parents.

The leaf sizes we measured from photographs with a digital tablet were very close to those we measured with a leaf area meter . We measured the leaf size over the whole growth period for the same leaf, and thus we obtained the growth function for a single leaf. Application of this method, however, has some restrictions. A leaf should have a smooth blade and pseudo-landmarks must be made at the time of the first measurement. If the

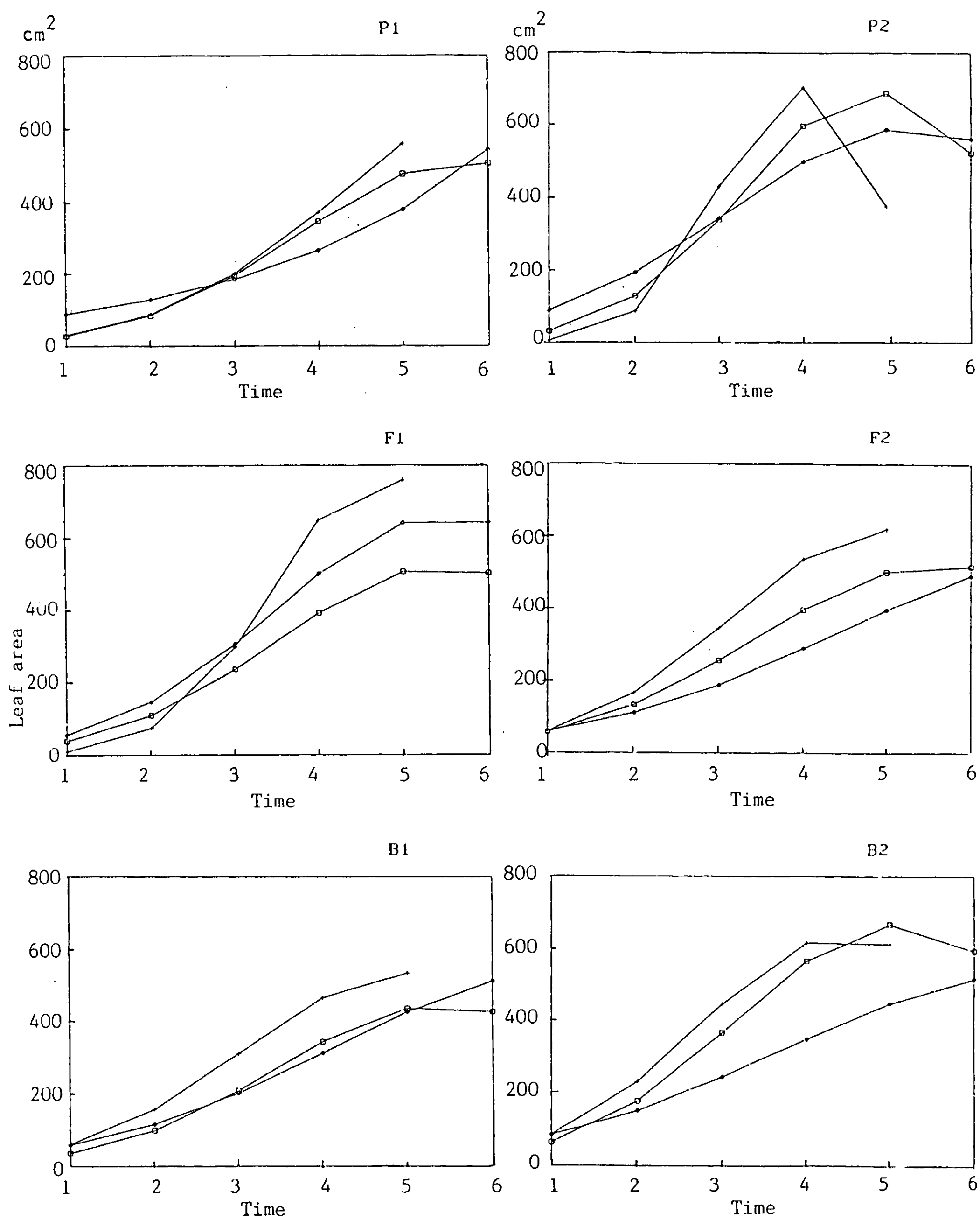

Fig. 2. The changes in leaf area for six generations. □:lower leaf, +: middle leaf, ◊:top leaf.

shape of the leaf blade is irregular or unsymmetrical, the method may incur some measurement error.

The Correlation Between Leaf Length and Width

The correlations between leaf length and width at six measuring times are shown in Figure 3. It shows that the leaf width grew almost synchronously with the length throughout the growth period, and therefore the leaf shape remained similar. Nevertheless, in the later growth stage of the top leaves of P_1 parents, the length grew faster than the width. Similar results were observed during the later growth stage of lower leaves of P_2 parents; middle to later growth stages of top, middle, and lower leaves of F_1; later growth stage of top leaves of B_1; early growth stage of lower leaves of B_2 and middle growth stage of lower leaves of the F_2 generation.

The growth of leaf length and width for most leaves was syntonic, and positive correlations were obtained between two parts. For some lower and top leaves, however, the length grew faster than the width, and hence the leaf shape became elongate, while in the later growth period, the middle leaves became wide oval because the width grew faster than or equaled the length. From this correlation analysis we show that the leaf shape is mainly determined by the growth rate of different parts of a leaf during different growth periods.

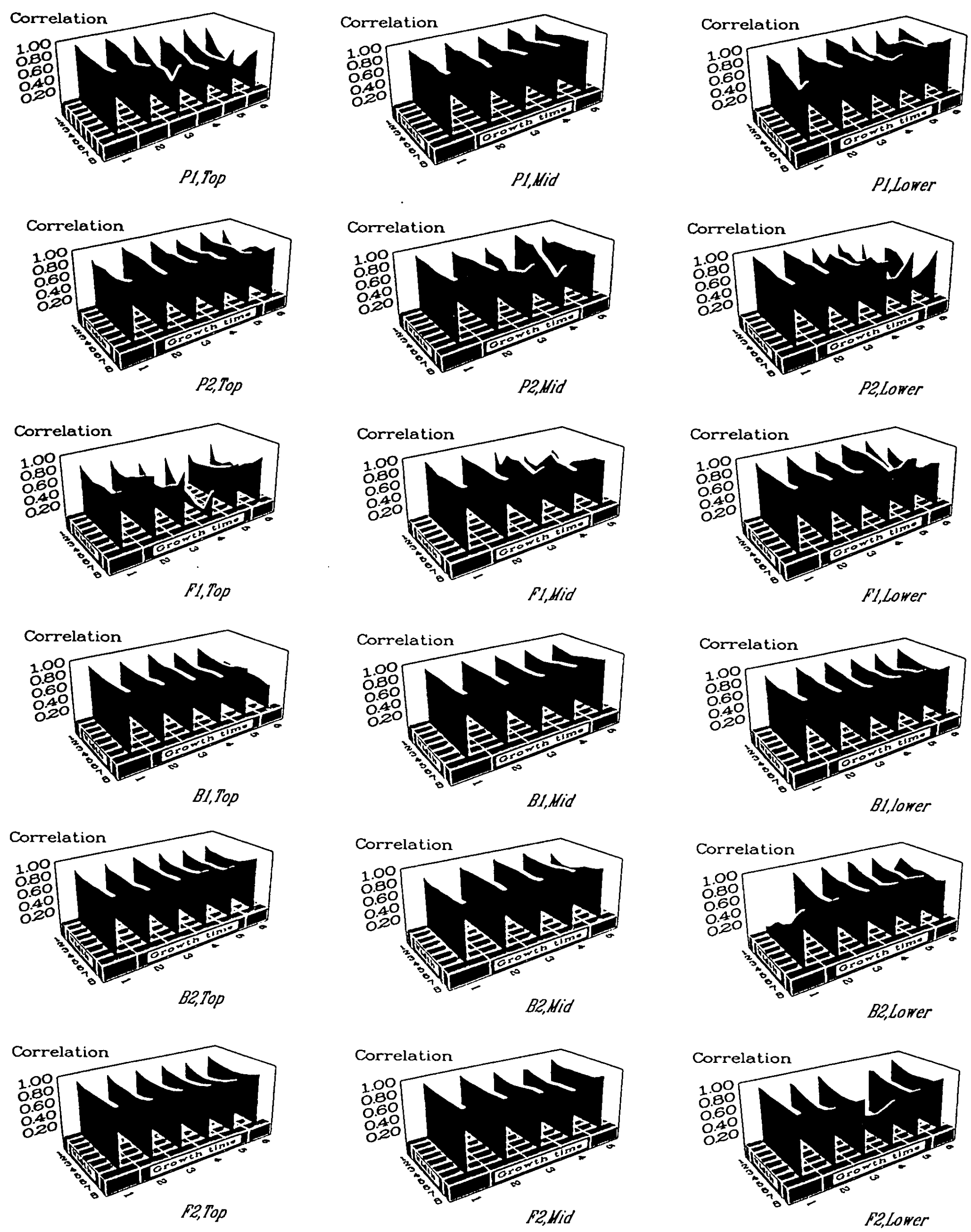

Fig. 3. The correlation between leaf length and width at three leaf positions during various growth stages for six generations.

The Correlations Among Various Leaf Widths

If a leaf grows at a constant rate during different growth stages, its shape will remain the same, but if the rate changes, its shape changes. Figure 4 shows the correlations between the first width and the subsequent widths. The leaf shape changed during the later growth stage of the top leaves of P_1 parents; the same phenomena were found in the middle and lower leaves of P_2 parents, all leaves of F_1, the top leaves of B_1, and the lower leaves of B_2 and F_2 generations.

A high correlation coefficient was found between the growth rates of width at two nearby points on a leaf, and a low coefficient was observed between points farther apart, but the correlation between the rates at the top and base part of some leaves was very high even though far apart.

During the initial growth period, the growth of top and base parts is faster than that of the middle part in the top and middle leaves, but in the middle and later growth periods this tendency reverses. The growth rates of different parts of lower leaves were similar during the entire period and showed symmetrical growth. The P_1 parents had a lot of small leaves, and so the coefficient changed little during growth. The P_2 parents had larger, but fewer, leaves, so the coefficient changed during growth. Little change of leaf shape in different leaves and generations was observed, therefore we decided to study the allometrical growth of different leaf widths.

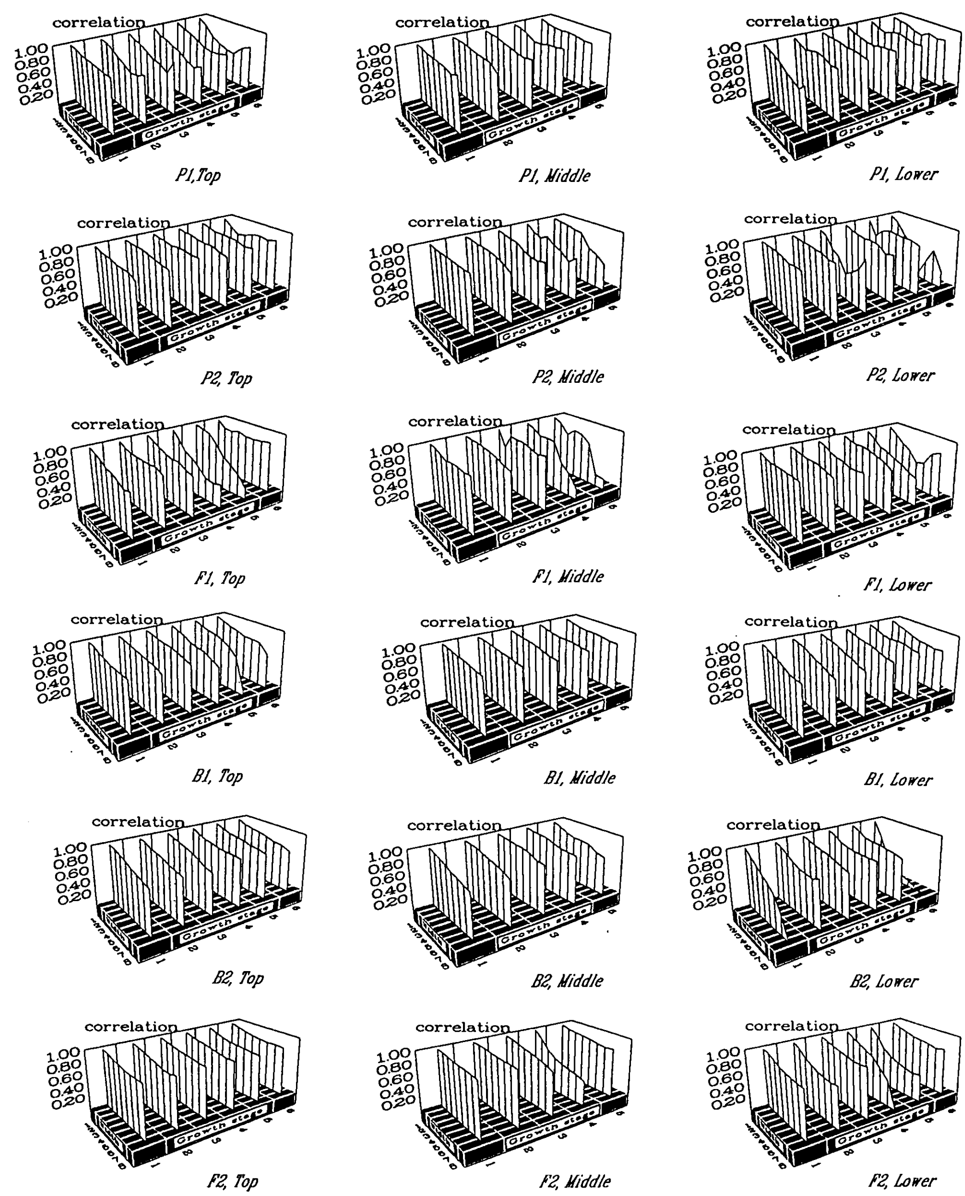

Fig. 4. The correlation between the first and other leaf widths at various growth stages for six generations.

Principle Component Analysis of Width Correlation

A principle component analysis of a correlation matrix of leaf width was performed; Table 1 shows the result for the P_1 parents. Values of the first component are located between + 0.3 and + 0.5; the second component values have a larger range between -0.8 and + 0.7. The first component may be regarded as the size factor and the second as the shape factor. The estimated discriminator of the second component for various growth stages of different leaves in six generations is shown in

Table 1. The coefficients of the 1st and 2nd principle component of P_1 in various growth stages (X_1 – X_8 indicate the location of leaf width)

Stage	1		2		3		4		5		6	
Comp.	1	2	1	2	1	2	1	2	1	2	1	2
Top leaf												
X_1	0.30	0.19	0.31	0.62	0.33	-0.67	0.23	0.40	0.28	0.17	0.28	0.56
X_2	0.33	0.28	0.33	0.42	0.38	-0.33	0.30	0.45	0.36	0.31	0.35	0.36
X_3	0.37	0.41	0.36	0.24	0.39	-0.12	0.36	0.36	0.36	0.38	0.39	0.02
X_4	0.37	0.37	0.37	0.02	0.39	0.10	0.40	0.21	0.36	0.28	0.46	-0.23
X_5	0.35	0.03	0.37	-0.22	0.41	0.32	0.43	-0.12	0.34	0.05	0.45	-0.32
X_6	0.39	-0.47	0.40	-0.44	0.33	0.32	0.43	-0.45	0.35	-0.07	0.35	-0.29
X_7	0.36	-0.55	0.36	-0.35	0.29	0.46	0.33	-0.47	0.31	-0.11	0.30	-0.06
X_8	0.35	-0.21	0.31	-0.14	0.30	-0.06	0.28	-0.13	0.45	-0.80	0.15	0.56
e.v.	0.06	0.00	0.06	0.00	0.04	0.00	0.03	0.00	0.04	0.00	0.02	0.00
cuml.	0.94	0.97	0.91	0.98	0.91	0.98	0.88	0.96	0.89	0.93	0.83	0.96
var.	0.0679		0.0624		0.0458		0.0336		0.0424		0.0263	
Middle leaf												
X_1	0.34	0.32	0.34	0.25	0.29	0.34	0.31	0.68	0.33	0.34		
X_2	0.34	0.48	0.33	0.29	0.32	0.36	0.33	0.44	0.32	0.37		
X_3	0.33	0.37	0.32	0.35	0.36	0.36	0.35	0.16	0.33	0.31		
X_4	0.39	0.17	0.32	0.13	0.37	0.18	0.37	-0.33	0.36	0.26		
X_5	0.38	-0.23	0.41	0.10	0.42	0.10	0.35	-0.34	0.39	0.08		
X6	0.38	-0.36	0.37	0.04	0.35	-0.33	0.35	-0.21	0.40	-0.27		
X7	0.34	-0.53	0.39	-0.30	0.34	-0.49	0.37	-0.20	0.37	-0.50		
X8	0.32	-0.17	0.35	-0.79	0.36	-0.48	0.39	-0.04	0.32	-0.50		
e.v.	0.08	0.00	0.04	0.00	0.05	0.00	0.05	0.00	0.04	0.00		
cuml.	0.95	0.98	0.92	0.97	0.93	0.98	0.95	0.99	0.93	0.97		
var.	0.0889		0.0410		0.0551		0.0539		0.0454			
Lower leaf												
X1	0.37	-0.49	0.30	0.11	0.32	-0.38	0.33	-0.04	0.37	-0.01	0.35	-0.39
X2	0.39	-0.40	0.31	0.24	0.35	-0.49	0.29	0.30	0.34	-0.39	0.41	-0.40
X3	0.41	-0.22	0.35	0.40	0.38	-0.32	0.31	0.50	0.33	-0.51	0.37	-0.41
X4	0.39	-0.02	0.39	0.37	0.39	-0.17	0.37	0.44	0.35	-0.30	0.35	0.16
X5	0.34	0.28	0.38	0.19	0.34	0.34	0.36	0.11	0.41	0.06	0.34	0.56
X6	0.31	0.39	0.39	-0.26	0.36	0.35	0.36	-0.21	0.39	0.25	0.36	0.37
X7	0.31	0.52	0.37	-0.59	0.32	0.42	0.40	-0.41	0.31	0.49	0.33	0.22
X8	0.29	0.21	0.32	-0.43	0.37	0.27	0.38	-0.49	0.32	0.43	0.31	0.01
e.v.	0.04	0.00	0.06	0.00	0.05	0.00	0.03	0.00	0.03	0.00	0.02	0.00
cuml.	0.87	0.95	0.94	0.99	0.96	0.98	0.91	0.95	0.90	0.97	0.91	0.95
var.	0.0511		0.0595		0.0571		0.0278		0.0285		0.0233	

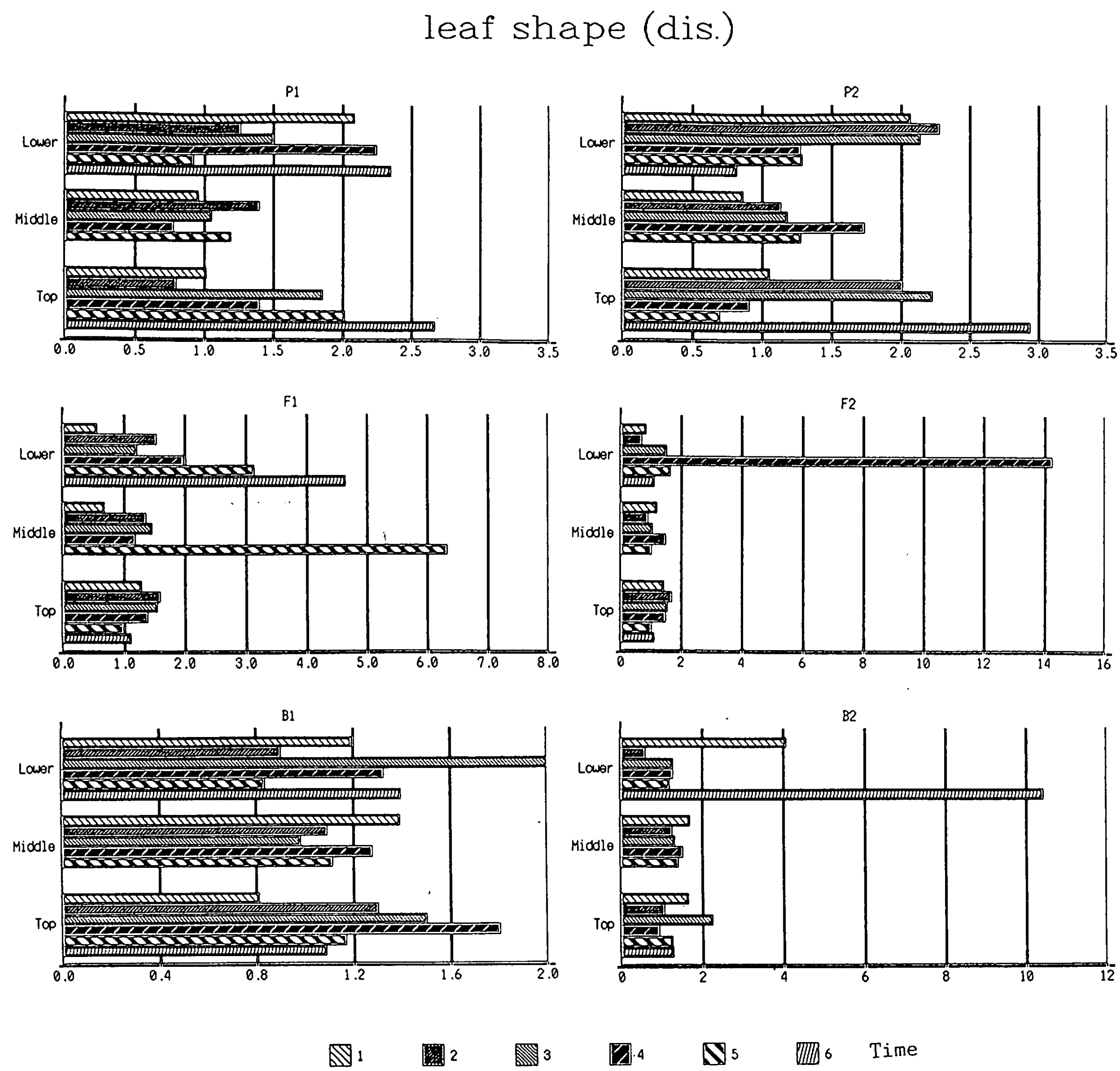

Fig. 5. Leaf-shape discriminator (horizontal axis) at various growth stages for six generations.

Figure 5. The leaf-shape discriminator values vary during different growth stages in some leaves, especially in the lower leaf of F_1, F_2 and B_2, and in the middle leaves of F_1. Note that P_1, P_2 and F_1 are homogeneous populations, hence the shape of a leaf does not change during growth, but the discriminator values obtained in this study do show change over time, and thus the discriminator is not a good parameter to represent leaf shape.

Estimated Allometrical Growth Coefficient

The allometrical coefficients of leaf length versus width as estimated by equation (2) are shown in Figure 6 for different leaves of six generations. The coeficients vary for different locations on a leaf, different locations of the leaf on the plant, and different generations. We obtained small allometrical coefficients for P_1, F_1, B_1, B_2 and F_2, but large coefficients for P_2 parents. We obtained similar results for leaf shape — wide oval leaves on P_2 parents and small elongate leaves on P_1 parents. A large length increases the width of the base part of the top leaves of P_2, F_1 and B_2; that of the middle leaves of P_1, P_2, B_1 and B_2; and that of the lower leaves of P_2 and F_2. Similar phenomena were found in the middle part of the top leaves of P_1, P_2, B_2 and F_2, and in the upper part of the lower leaves of P_1 and the middle leaves of F_2. Owing to the different leaf shapes in different leaves and generations, different allometrical coefficients were observed in different parts of leaves.

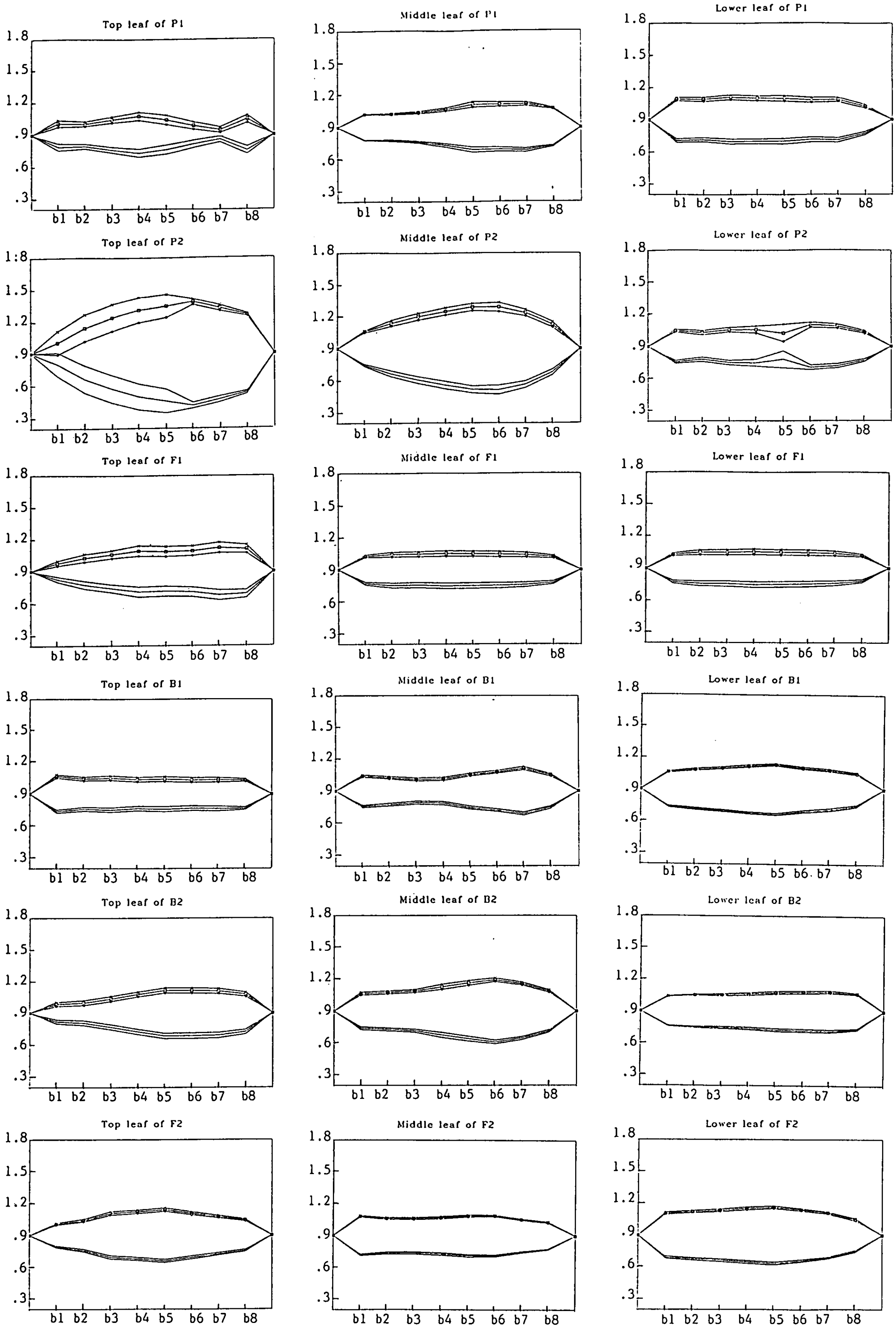

Fig. 6. Allometrical growth coefficients of leaf length and each leaf width (horizontal axis) for six generations. x=upper bound, ◊=lower bound

Estimation of Leaf-Shape Index

The estimated values of leaf-shape index (β_z) determined by equation (11), for the three leaf locations, in various stages of growth, for six generations, are shown in Figure 7. The leaf-shape indices for various growth stages are very similar; this means that the leaf shapes remained relatively constant during growth. The wide oval leaves, such as the top leaves of P_2 parents, have a large index, and the smaller, elongated leaves, like the top leaves of P_1 parents, have a small index. The shapes of the lower and middle leaves of P_1 parents are similarily elongated, though the top leaves are somewhat longer

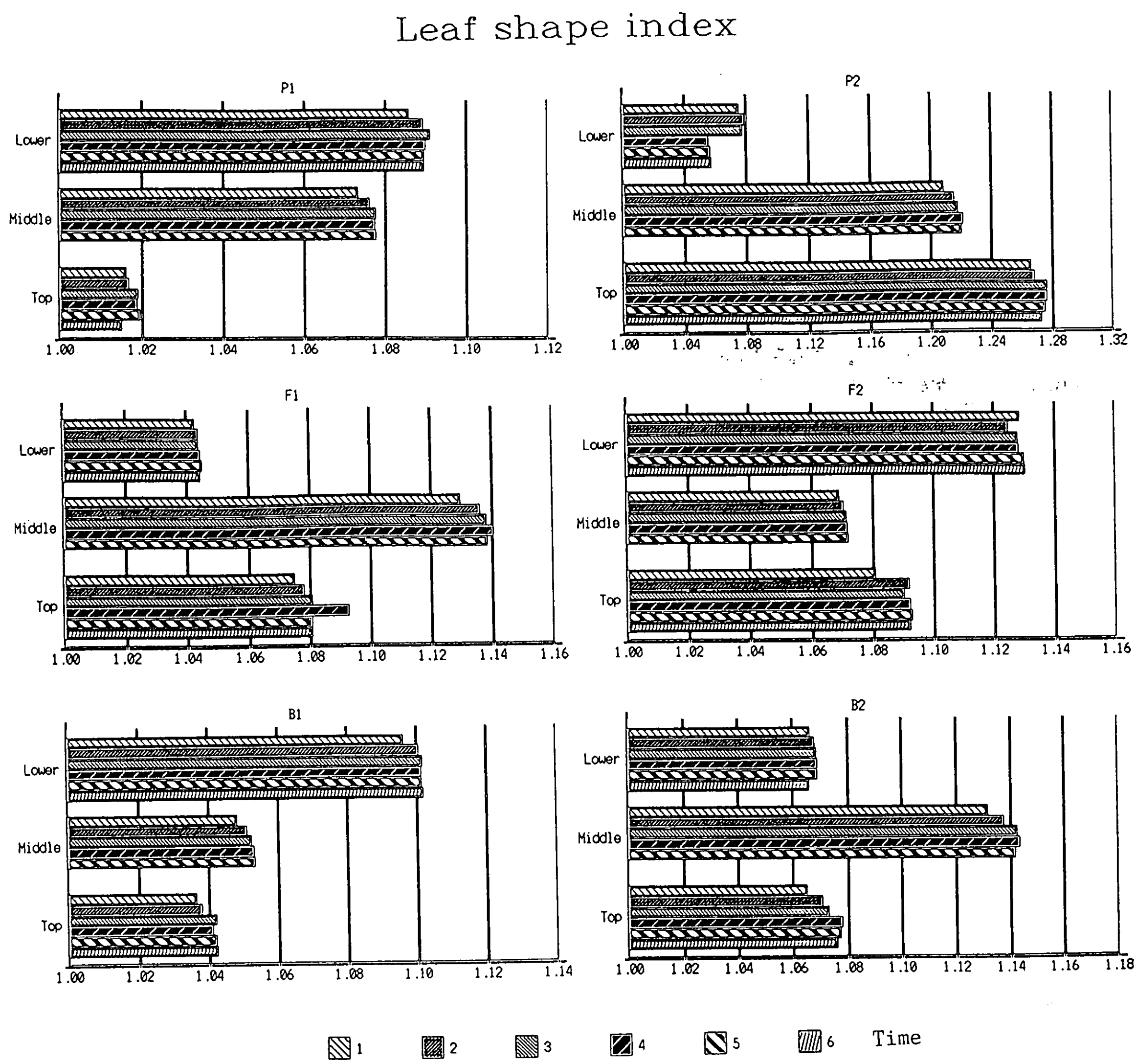

Fig. 7. Leaf-shape index (horizontal axis) at various growth stages for six generations

still. The leaves of P_2 parents are all wide and oval, the top leaves are the largest and the lower leaves are the smallest. The middle and top leaves of F_1 are intermediate in shape between those of the P_1 and P_2 parents, but the lower leaves are less elongated than those of P_1 parents. The top leaves of the F_2 generation are like those of F_1 — intermediate in shape between those of the P_1 and P_2 parents. The lower leaves are oval and the middle leaves are like those of P_1 parents. The leaves of the B_1 generation are similar to those of the P_1 parents. The lower leaves of the B_2 generation are similar to those of P_2 parents, while the middle and top leaves are the same as those of F_1 in the same locations.

We used two methods to estimate the parameters of leaf shape — leaf-shape discriminator (Fig. 5) and leaf-shape index (Fig. 7). The results show that the parameter of leaf-shape index gives a more reasonable representation of the leaf shape, because the estimators are similar during different growth periods, as shown in Fig. 7. The estimation with leaf-shape discriminator (Fig. 5) varies at different growth stages in same leaf, and will not be studied further.

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Literature Cited

Gould, S. J. 1966. Allometry and size in ontogeny and phylogeny. Biological Review **41:** 587–640.

Griffiths, D. A. and R. L. Sandland. 1984. Fitting generalized allometric model to multivariate growth data. Biometrics **40:** 139–150.

Griffiths, D. A. and R. L. Sandland. 1985. Fitting allometric models to multivariate growth data. Proc. International Biometrics Conference, pp. 229–234.

Hopkins, J. W. 1966. Some considerations in multivariate allometry. Biometrics **22:** 747–760.

Huxley, J. S. 1924. Constant differential growth ratios and their significance. Nature, **114:** 895–896.

Richards, F. G. 1969. The quantitative analysis of growth. *In* F. C. Steward (ed.), 'Plant Physiology, VA', Academic Press, N. Y., pp. 3–76.

Sampson, P. D. and A. F. Siegel. 1985a. The measure of size independent of shape for multivariate lognormal populations. Jour. Amer. Stat. Asso. **80:** 910–914.

Sampson, P. D. and A. F. Siegel. 1985b. The measure of size independent of shape for multivariate lognormal populations: Definition and application. Proc. International Biometrics Conference, pp. 235–244.

Sprent, P. 1972. The mathematics of size and shape. Biometrics, **28:** 23–37.

Turner, M. E. 1978. Allometry and multivariate growth. Growth, **42:** 434–450.

Veitch, L. G. 1978. Size, shape and allometry in *Uca*; A multivariate approach. Math. Scientist **3:** 35–45.

植物數量性狀間相對生長之遺傳研究

I.形狀與大小之測定

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本研究利用葉形全異的二栽培種菸草及其四種雜交後裔爲材料，建立在不剪葉片的前提下，利用照相法與數位板測定土葉、中葉及天葉各生長期的葉寬及葉長，推測葉面積及其生長模型，並利用葉寬相關矩陣及主成份分析法或葉長與葉寬的相對生長函數等建立葉形的估計方法。由照相法與數位板測定葉寬與葉長後所推測的葉面積大小與利用葉面積儀測定者相等，兩者間之相關係數達 0.9992 ，葉面積之生長函數呈指數函數，而利用葉長與葉寬間的相對生長函數所推測之葉形指數較利用葉寬相關矩陣與主成份分析所得葉形判別值者較能代表葉形的介值，似可利用前者進行葉形的遺傳分析。

關鍵詞：相對生長；葉形；葉形判別值；葉形指數；葉大小；菸草；主成份分析。