USE OF DISCRIMINANT EQUATIONS TO CLASSIFY BIRCH IN THE NORTHEAST

by

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ABSTRACT.--Leaf, seed, and bract measurements from three species of birch (Betula alleghaniensis Britton, Betula papyrifera Marsh., and Betula populifolia Marsh.) were used to classify intraspecific crosses and their hybrids correctly. Seed and bract measures provide a sufficient basis for discriminating among superior trees that have been selected as potential breeding stock. The occurrence of introgression in both the parents and progeny can be established.

INTRODUCTION

Throughout the Northeast birch is one of the most important hardwood tree species groups. Three of the most abundant birch species are Betula alleghaniensis Britton (yellow birch), Betula papyrifera Marsh. (paper birch), and Betula populifolia Marsh. (gray birch). Frequently, it is necessary to identify and classify trees within a species for breeding programs and the selection of superior trees. A method is needed to assist forest geneticists in selecting trees that appear to be the desired species, and that do not have considerable variation from the parent population. The interchange of genetic material between species needs to be distinguishable with the use of physiological traits.

Reports of introgression of birch being common in nature include Betula alleghaniensis Britton X Betula papyrifera Marsh. (Barnes et al. 1974, Clausen 1966), B. alleghaniensis X Betula lenta L. (Sharik and Barnes 1971), B. papyrifera X Betula pumila L. (Clausen 1962), and B. alleghaniensis X B. pumila (Dancik and Barnes 1972). Because of their sympatry and great amount of polymorphism, frequent crossings between B. papyrifera and B. populifolia have long been suspected (Alam and Grant 1972, Dugle 1966).

Evaluation of introgression and hybridization has been based primarily on techniques which include descriptive statistics of morphological traits, and on various graphical displays. Methods include: hybrid indices (Anderson 1949, Clausen 1962, Dugle 1966, Sharik and Barnes 1971), biometrical studies (Alam and Grant 1972, Dancik and Barnes 1974, Jentys-Szaferowa 1937, Sharik and Barnes 1971), pictorial scatter diagrams (Anderson 1949, Clausen 1962, Dancik and Barnes 1974, Dugle 1966), and polygonal graphs (Clausen 1962, Guerriero et al. 1970). Recent studies have evaluated hybridization by canonical correlation analysis (Barnes et al. 1974, Dancik and Barnes 1974, Namkoong 1966) and principal component analysis (Dancik and Barnes 1974, Richens and Jeffers 1975, Sharik and Barnes 1971). Discriminant analysis has been used for studies of introgression since interpopulational differences are maximized (Dancik and Barnes 1974, Ledig et al. 1966, Namkoong 1963, Sharik and Barnes 1971, Smouse 1972).

The purpose of this study was to develop discriminant functions based on tree, leaf, and seed variables to identify individual species of trees and categorize them as intraspecific crosses or hybrids of yellow birch, paper birch, and/or gray birch.

METHODS

Trees described as B. alleghaniensis, B. papyrifera, and B. populifolia were selected at locations in Connecticut and Massachusetts. Progeny from controlled interspecific, intraspecific, and self-pollinated crosses were planted in 1946 at the Standing Stone Experimental Forest, Stone Valley, Pennsylvania.

Trees in the available crosses were sampled in 1969 for a total of 52 trees representing 23 crosses (Table 1). Fifteen of these were controlled-pollinated hybrids of yellow X paper birch and gray X paper birch. Hybrids between gray and yellow birch were not represented and are thought to be incompatible (Clausen 1966).

A total of 26 measurements and combination of measurements were made on branches, strobili, leaves, seeds, and bracts (Table 1, Fig. 1). The number of branch nodes joining the central branch were recorded for each branch (A). Because there are morphological differences between leaves from various parts of a tree's crown (Clausen and Kozlowski 1965, Jentys-Szaferowa 1937), 20 leaves were selected randomly from four locations on similar midcrown branches. Five

Table 1.--Mean values of shade leaf, seed, and bract variables for intraspecific and hybrid birch crosses.

Variable	Yellow	Yellow X Paper	Paper X Yellow	Paper	Paper X Gray	Gray X Paper	Gray
Crosses (number)	7	1	3	4	1	2	5
Trees (number)	14	2	7	11	1	5	12
Haploid (number)	42	42	42	42	28	28	14
A - branch nodes (number)	14.43	16.00	16.86	18.27	31.00	22.80	59.00
B - petiole length (mm)	9.36	9.00	18.43	21.82	23.00	23.60	21.92
C - blade length (mm)	83.71	74.00	77.86	76.36	58.00	71.20	57.08
D - blade width (1/4) (mm)	38.57	34.50	44.71	44.18	36.00	40.40	38.75
E - blade width (1/2) (mm)	45.21	37.00	46.29	47.18	30.00	35.00	26.42
F - blade width (3/4) (mm)	28.36	24.00	24.57	28.18	13.00	12.60	7.75
G - vein number	12.36	11.50	9.29	8.81	7.00	8.60	7.50
H - serration number/25.4 (mm)	13.14	13.50	13.42	12.36	14.00	13.80	14.92
I - petiole-leaf angle	99.50	88.50	92.29	82.54	84.00	66.00	77.00
J - strobilus length (mm)	24.07	22.91	37.16	43.72	52.32	42.20	22.72
K - strobilus width (mm)	12.07	10.57	8.08	7.95	7.01	5.93	5.79
L - seed length (mm)	3.51	3.11	2.13	1.93	1.71	1.73	1.58
M - seed width (mm)	1.60	1.61	1.30	1.30	1.01	1.14	.74
N - mean seedwing width (mm)	.94	.71	.96	1.18	1.11	1.00	.91
0 - samara width (mm)	3.48	3.03	3.22	3.66	3.23	3.14	2.56
P - mean stigma length (mm)	.73	.63	.67	.63	.45	.67	.45
Q - total bract length (mm)	8.67	9.31	6.05	6.02	5.08	4.26	3.37
R - middle lobe length (mm)	3.36	2.89	2.07	1.84	1.76	1.73	1.07
S - mean side lobe length (mm)	4.78	4.32	2.38	2.13	2.11	1.86	1.82
T - mean total side lobe length (mm)	7.97	8.46	4.87	4.72	3.54	3.26	2.87
U - mean lobe angle	38.93	34.50	72.29	68.18	87.00	80.20	80.75
B/C	.11	.12	.25	.29	.39	.33	.39
C/E	1.85	2.01	1.67	1.63	1.93	2.05	2.20
D/F	1.36	1.43	1.82	1.57	2.77	3.21	5.00
E/F	1.59	1.54	1.88	1.67	2.31	2.78	3.41
Q/s	.55	.46	.40	.36	.42	.44	.54

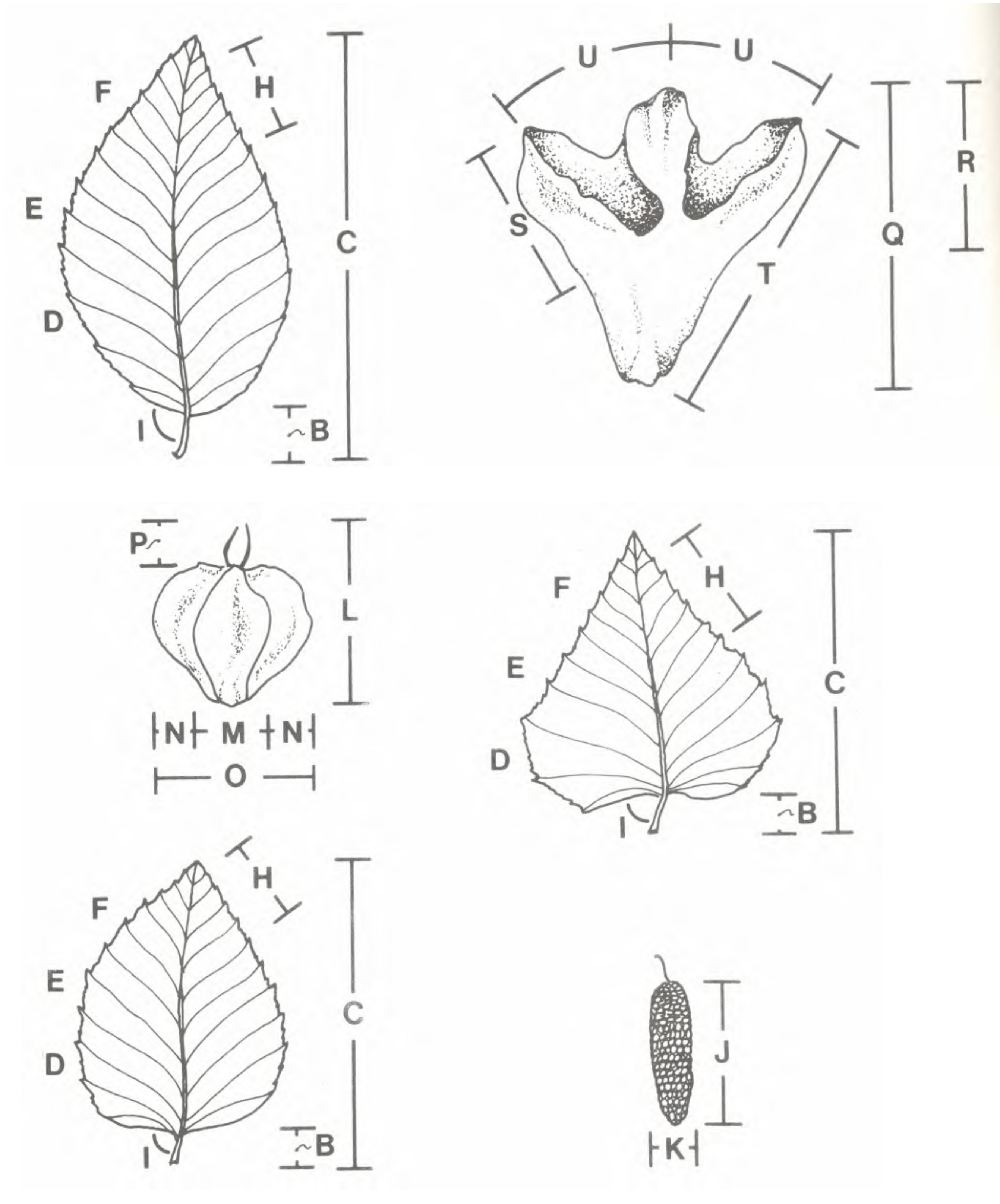


Figure i.---Measurements made on leaves, stroblius, seeds, and bracts.

sun leaves were taken from older shoots (spring, leaves) and five from elongating shoots (summer, leaves). Ten shade, leaves also were selected--five spring and five summer leaves. The shade leaves had the least amount of variation and were used in the discriminant equations. Measurements made on each leaf included: petiole length (B); blade length (C); blade width at 1/4 (D), at 1/2 (E), and 3/4 (F) of the length; number of veins per side of midvein (G); number of serrations per unit (H); and the average degree of base angle between the petiole and blade (I). Five female strobili were chosen at'random from each tree. The length (J) and width (K) of the green strobili were measured, and five bracts of five seeds were randomly selected from the middle of the of the catkin. Seed and bract measurements included, length (L) and width (M) of the seed, average width of the seedwings (N), total samara width (0), average length of stigma (P), length of total bract (Q), length of middle lobe (R), average length of side lobes taken from middle of bract (S), average length of side lobes taken from bract base (T), and average angle between bract lobes (U).

RESULTS AND DISCUSSION

A set of linear classification functions were computed from the measured variables of the form:

$$Z = \beta_0 + \beta_1 X_1 + \beta_2 X_2 + \beta_K X_K$$

where X_1, X_2 ... X_K are the measured variables, and β_1, β_2 ... β_K are the coefficients for the canonical variables. The first two canonical variables (Z_1 and Z_2) can be plotted to provide a separation of the three species groups. After testing, nonsignificant variables were eliminated from the equation and only significant combinations of variables were used for discrimination (Table 2).

The use of equations developed from measurements on leaves, seeds, bracts, and strobilus provides forest geneticists with flexibility in classifying trees (Table 3). Also, the addition of the second variable in the equation usually classified intraspecific crosses correctly. If the physical traits in Table 1 as described in Figure 1 are used, individual trees can be compared to the known intraspecific means for possible introgression or hybridization. Z-values for each tree can then be used as a means of species classification.

Table 2. Significant seed, leaf, strobilus, and bract variables and percentage trees correctly classified.

Variable	Yellow birch	Paper birch	Gray birch
B/C - petiole/blade length ratio	100	90.9	83.3
C/E - blade length/width ratio	100	100	100
L - seed length (mm)	100	81.8	91.7
J - strobilus length (mm)	100	100	100
L - seed length (mm)	100	81.8	91.7
M - seed width (mm)	100	100	100
L - seed length (mm)	100	81.8	91.7
Q/S - bract length ratio	100	90.9	100
M - seed width (mm)	100	100	100
L - seed length (mm)	100	81.8	91.7
J - strobilus length (mm)	100	100	100
Q/S - bract length ratio	100	100	100
S - length of side lobe (mm)	100	100	100
Q - total bract length (mm)	100	100	100
N - wing width (mm)	100	100	100
K - strobilus width (mm)	100	100	100
M - seed width (mm)	100	100	100

Table 3. Equations of leaf, seed, and bract variables used to discriminate among the three species of birch.

		1			Z_2		
Equation	Variable	Coefficient	Intercept	Canonical correlation	Coefficient	Intercept	Canonical correlation
1	Petiole/blade length ratio (B/C)	-26.924	8.872	0.96	-5.149	-7.936	0.71
	Blade length/width (C/E)	-1.082			4.870		
2	Seed length (L) (mm)	3.646	-4.303	.97	1.403	-9.184	.89
	Strobilus length *(J)* (mm)	-0.153			0.197		
3	Seed length (L) (mm)	2.945	-10.097	.96	-3.250	-2.332	.83
	Seed width (M) (mm)	2.423			8.257		
4	Seed length (L) (mm)	4.627	-16.497	.98	-0.774	-0.877	.94
	Total bract length ratio (Q/S)	18.713			-13.498		
	Seed width (M) (mm)	-3.109			7.585		
5	Seed length (L) (mm)	3.234	-7.336	.99	-1.786	3.646	.96
	Seed width (M) (mm)	-0.097			6.155		
	Strobilus length (J) (mm)	-0.120			0.073		
	Petiole/leaf length ratio (B/C)	-16.680			-10.876		
	Total bract length ratio (Q/S)	15.124			-12.926		

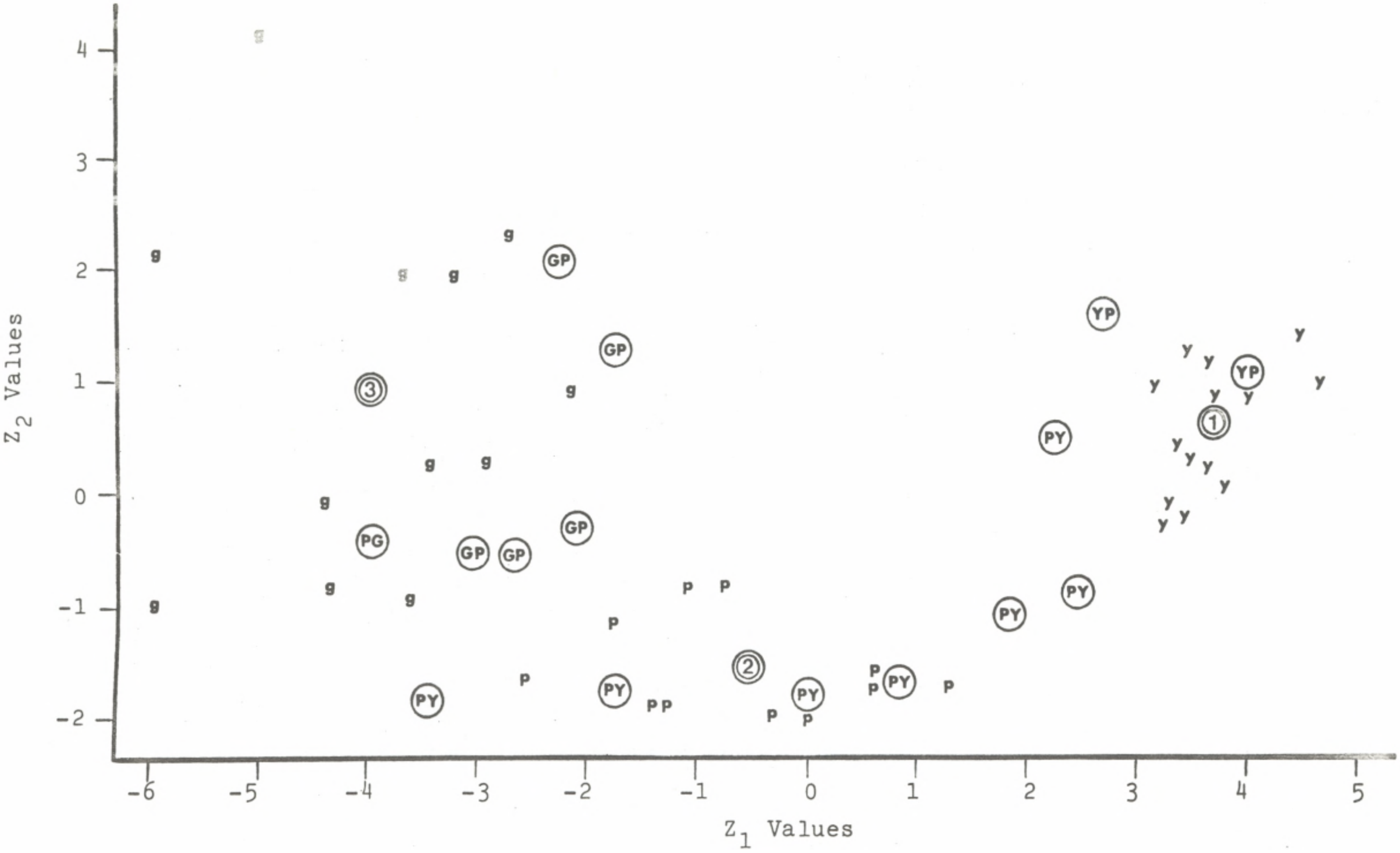
Leaf Variables

In general, spring leaves were found to be better discriminators than summer leaves and shade leaves better than sun leaves. There were small differences between spring/shade leaves and summer/shade as a result of very small elongation of shade branches. Thus, midcrown shade leaves were combined and used to develop discriminant functions for subsequent analysis.

The most effective leaf variable for discriminating among all three intraspecific crosses, and particularly for separating yellow birch from the other two species was the ratio of petiole length to blade length (B/C). The addition of the ratio of blade length to blade width (C/E) to the model was significant, and led to the correct classification of all three species. Others have found that the ratio of petiole length to blade length is an important variable for discriminating between different species of birch (Barnes et al. 1974, Dancik and Barnes 1975, Jentys-Szaferowa 1937, Sharik and Barnes 1971).

Although all three species were classified correctly, additional variables led to significant improvements in the discriminant functions. These measures on physical traits provided an additional basis upon which to discriminate between paper and gray birch.

However, of the list of leaf variables (Table 1), only the two most significant variables were used due to the high correlation between measures of leaf width, and because all three species when self-pollinated were classified correctly. The discriminant scores (Z-values) for each observation are plotted in Figure 2. Intraspecific crosses of yellow birch individuals were tightly clustered around their group mean, indicating low variability within this species on the discriminating variables. Intraspecific crosses of paper and gray birch trees were somewhat more variable than yellow birch. Yellow paper birch hybrids were classified as being most like the female parent in a majority of cases. The single paper X gray birch hybrid was classified as a gray birch. Of the five gray X paper birch hybrids, all were classified as gray birch, but were located between the means of the intraspecific crosses. Mean values for leaf characteristics of the hybrids were more like gray than paper when gray was the female parent (Fig. 2).



Intraspecific Crosses

1) Mean - yellow birch (Y)
2) Mean - paper birch (P)
3) Mean - gray birch (G)

Hybrids

yellow X paper birch (YP)
paper X yellow birch (PY)
paper X gray birch (PG)
gray X paper birch (GP)

Figure 2. Intraspecific and interspecific crosses of yellow, paper, and gray birch as classified by equation 1 using variables (B/C) and (C/E).

Seed, Bract, and Strobilus Variables

The classification of trees to a species group using leaf variables worked correctly for intraspecific crosses of yellow birch and of paper birch. However, one gray birch usually was misclassified. Therefore, a similar stepwise discriminant analysis of the data for seed, bract, and strobilus was used to determine if other variables could improve the classification of this species (Table 3).

Seed length (L) and length of the green strobilus (J) together were sufficient to classify all intraspecific crossed individuals correctly (Fig. 3). The intraspecific crosses were grouped closer to the overall species means for the three species. However, the hybrids were not located between the means of the intraspecific crosses, because yellow and gray birch have similar strobilus lengths, with paper birch having longer strobili. Since the hybrids were scattered, equation 2 discriminates better for intraspecific crosses than hybrids.

The plotting of Z-values in equation 3 indicated that the intraspecific crosses were classified correctly using seed length (L) and seed width (M) (Fig. 4). Yellow birch indicated more scatter of intraspecific crosses than paper or gray when compared with equation 2. However, the location of the hybrids is as expected with the majority of points located between the overall species means.

The addition of the ratio of bract length to side lobe length (Q/S) as a variable in equation 4 caused only slightly more variation in the intraspecific crosses of yellow and gray birch (Fig. 5). The plotting of the hybrids for the three species was closer to the expected hybrid mean between the species means. Paper and yellow birch crosses were plotted closer to the mean of the female parent, with several plotted as paper and gray birch crosses. However, the paper and gray birch hybrids were plotted as hybrids between the parental means.

All Variables Combined

Leaf, bract, and seed variables were combined to produce equation 5 (Fig. 6). The best variables for discriminating between intraspecific crosses were seed length (L), strobilus length (J), ratio of petiole length to leaf length (B/C), ratio of side lobe length to total bract length (Q/S), and seed width (M). The combination of leaf, seed, and bract variables used to

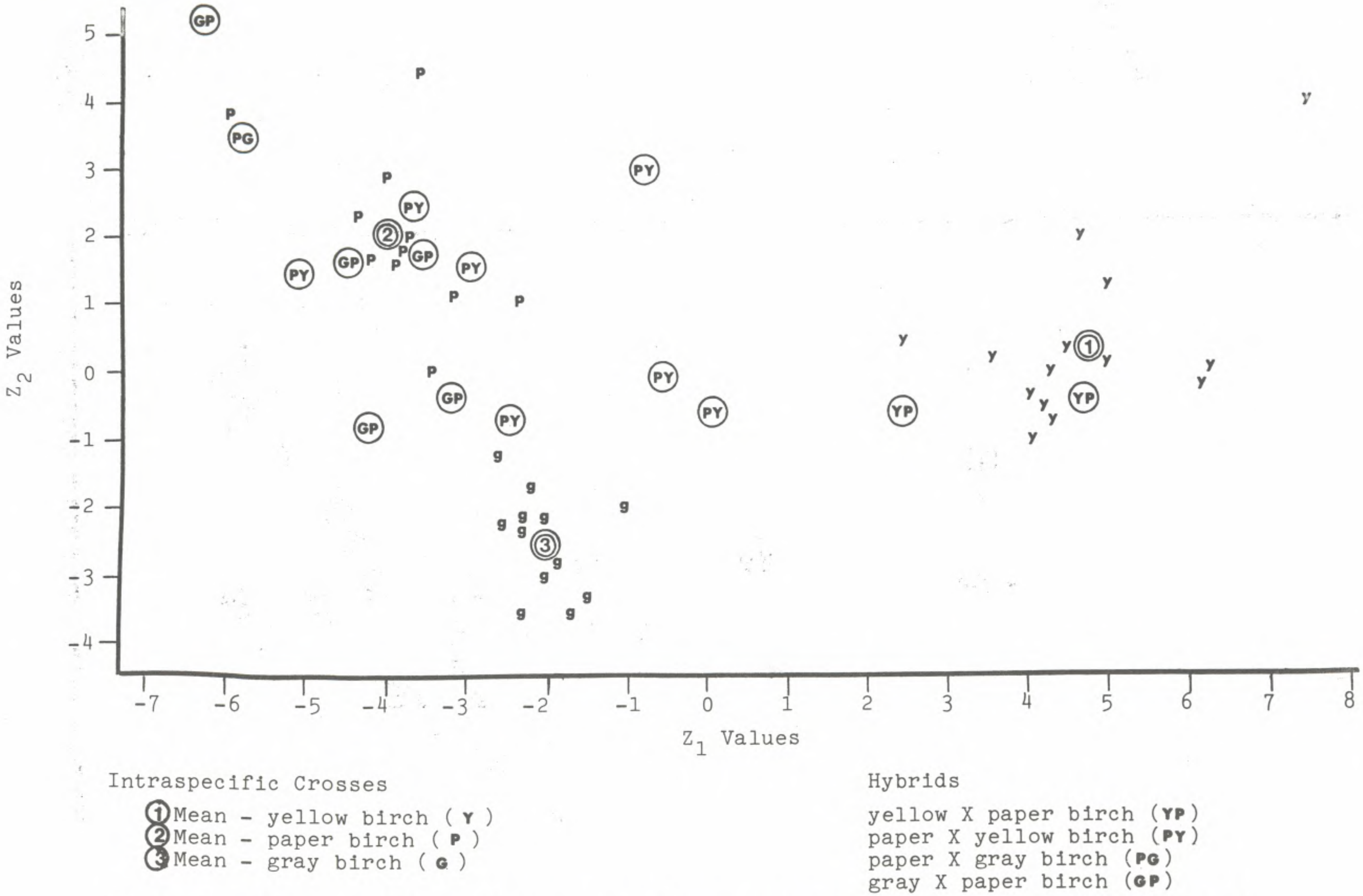


Figure 3. Intraspecific and interspecific crosses of yellow, paper, and gray birch as classified by equation 2 using variables (L) and (J).

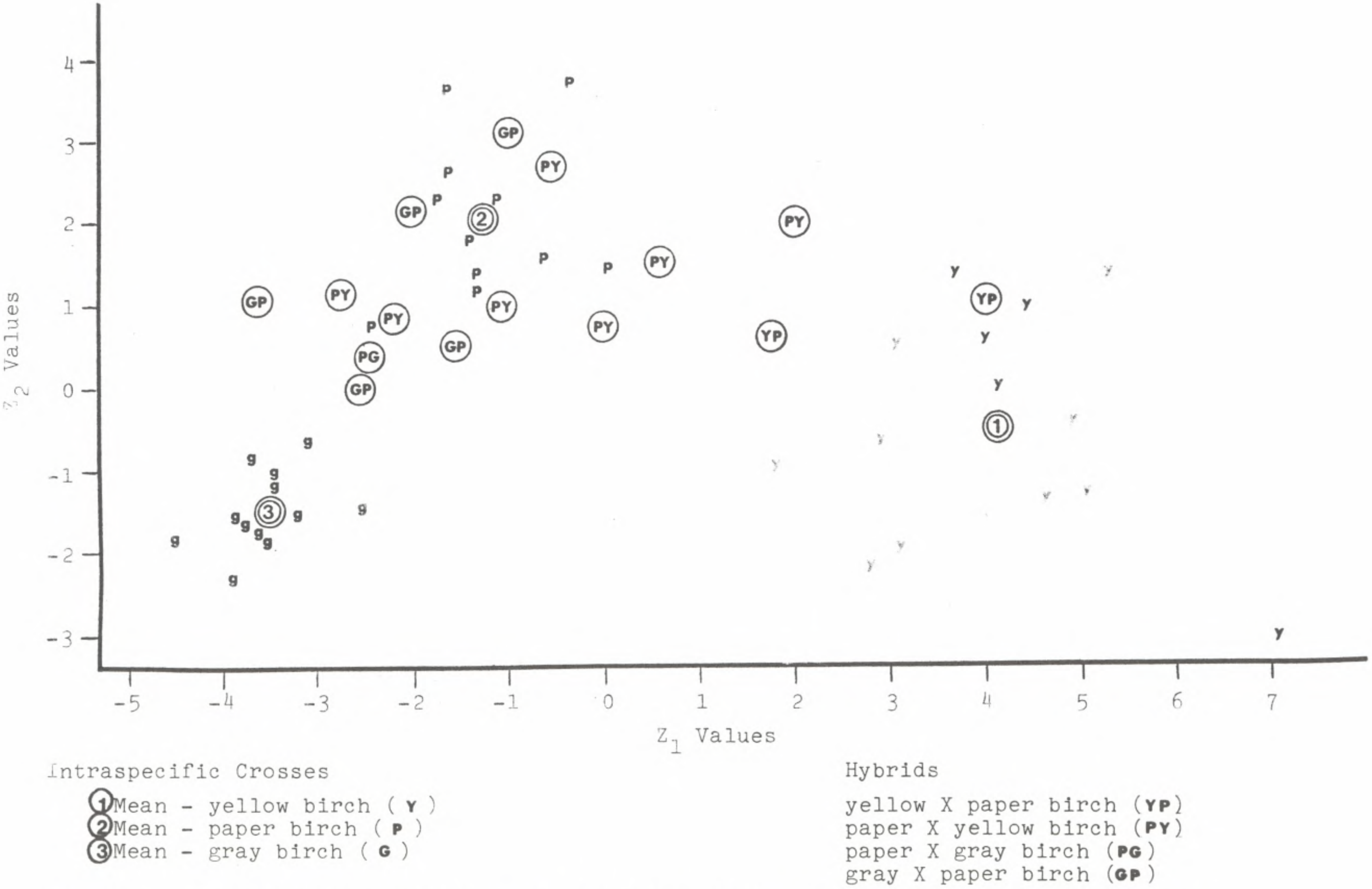


Figure 4. Intraspecific and interspecific crosses of yellow, paper, and gray birch as classified by equation 3 using variables (L) and (M).

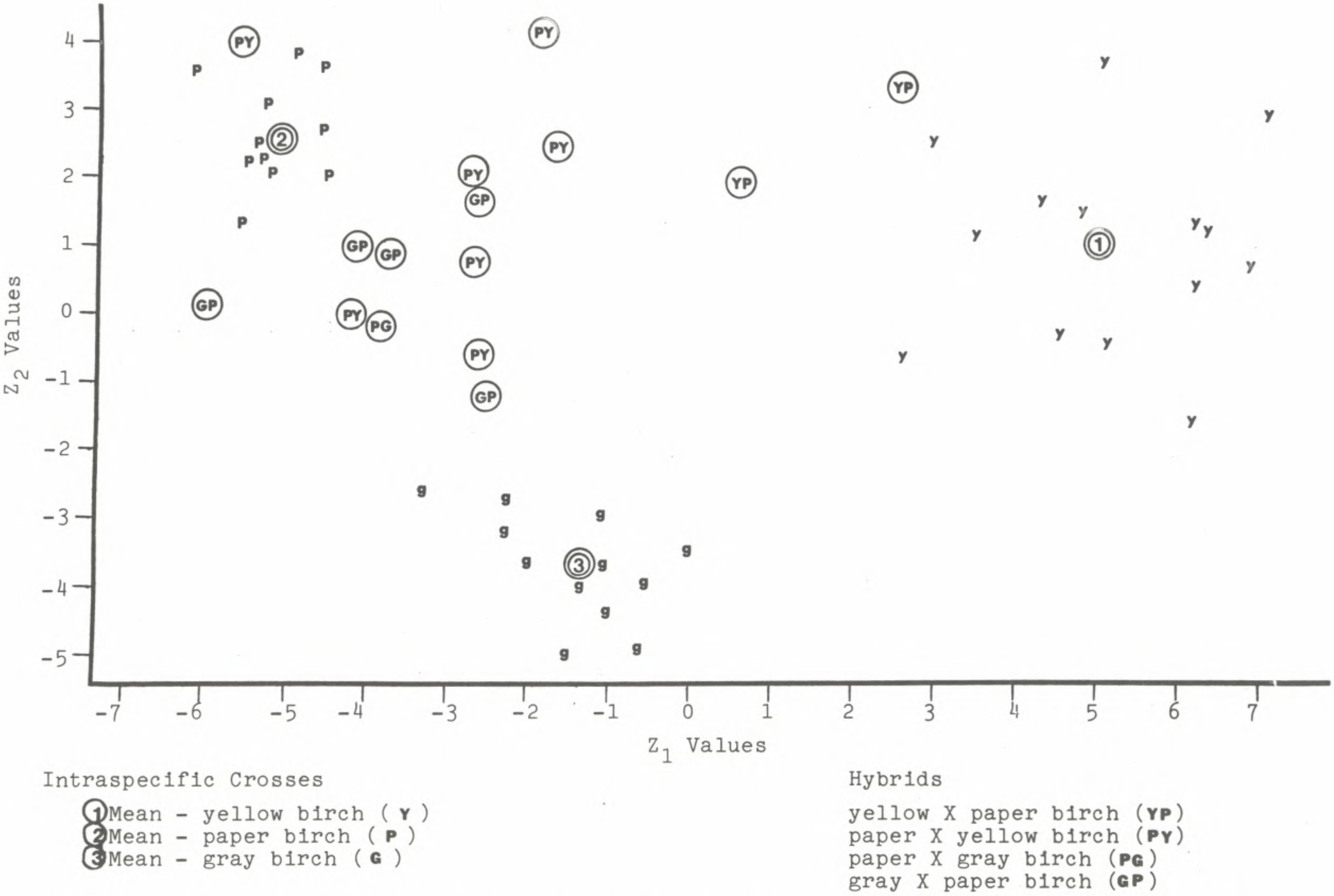


Figure 5. Intraspecific and interspecific crosses of yellow, paper, and gray as classified by equation 4 using variables (L), (Q/S), and (M).

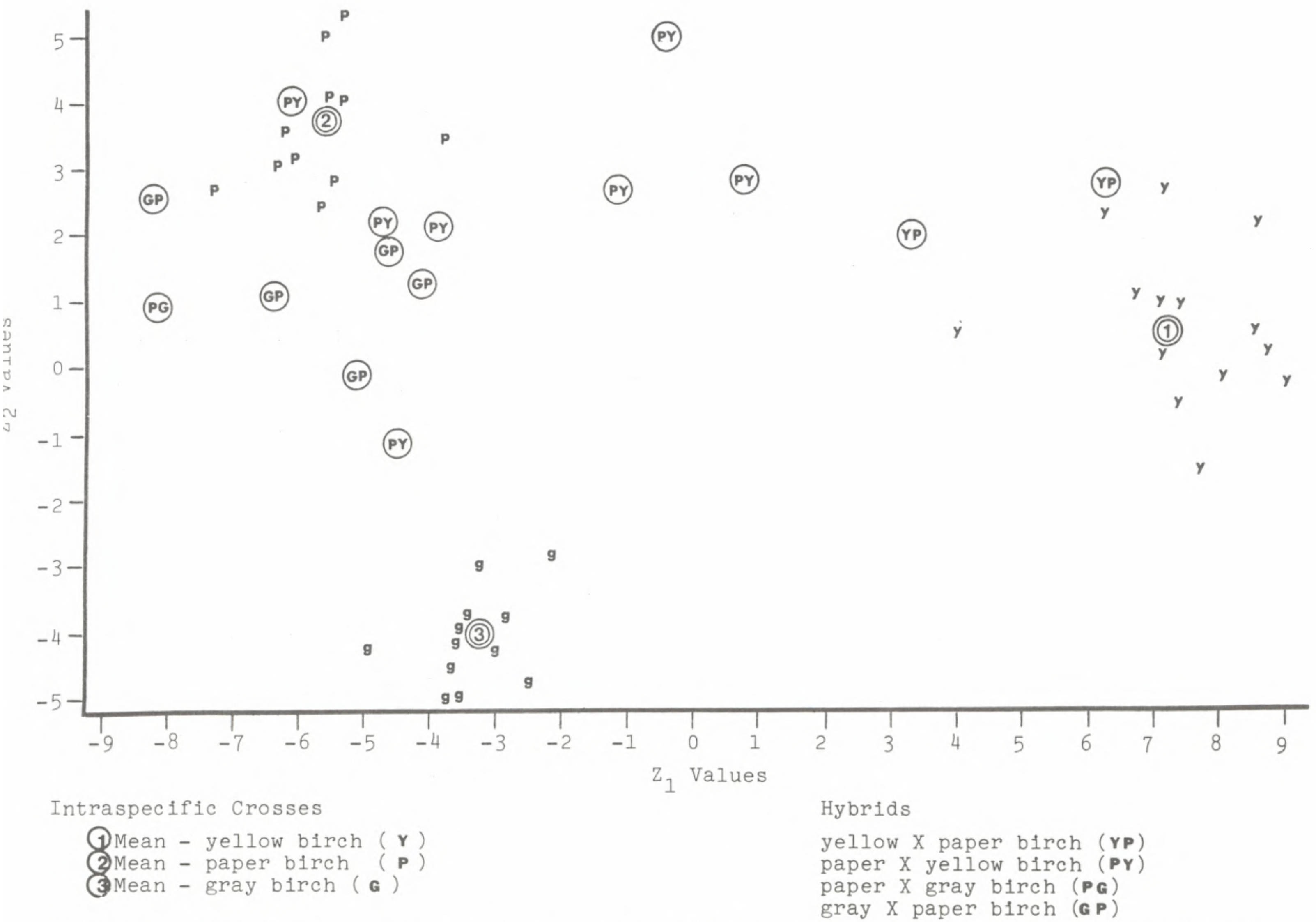


Figure 6. Intraspecific and interspecific crosses of yellow, paper, and gray as classified by equation 5 using variables (J), (L), (M), (B/C), and (Q/S).

derive equation 5 leads to the results found previously when each set of variables was analysed separately and all of the intraspecific crosses were classified correctly. Individuals of gray birch had slightly more variation. All yellow X paper birch crosses were classified as yellow birch, while paper X yellow were classified as paper birch, with one classified as gray birch. The gray X paper and paper X gray were classified as paper birch, apparently because strobilus length was included in the equation.

APPLICATION

Intraspecific crosses of yellow, paper, and gray birch were classified successfully when leaf, strobilus, seed, and bract variables were used. The average of five shade leaves from the midcrown proved more reliable for discrimination than sun leaves.

Discrimination between intraspecific species was possible with either leaf variables (ratio of petiole length to blade length, and ratio of blade length to blade width) or seed and bract variables (seed length, seed width, strobilus length, and ratio of bract side lobe to midlobe length). All trees were classified correctly with two variables for the five equations.

Paper birch crosses with yellow birch females were classified as yellow birch. Most yellow birch crosses with paper birch females were classified as paper birch, though one was classified as a gray birch, possibly because of the similarities in strobilus length.

Seed and bract variables seem to be more useful for discrimination between species and classification of hybrids than either leaf or seed and strobilus variables. When only leaf variables were used, gray X paper and paper X gray tended to be classified as gray. However, when seed and bract variables were used, the hybrids of paper and gray were intermediate between the intraspecific crosses. Including strobilus length as a variable led to variation in the classification of hybrids. The best combination of leaf, seed, and bract variables classified paper and gray crosses as paper.

Any of the discriminant equations can be used to classify individual trees with regard to intraspecific crosses. However, the use of the seed and bract equation is more reliable for the classification of hybrids. Indications of variation and possible introgression can be obtained with the use of the leaf variables or the equation with all variables combined.

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