PHENOTYPIC VARIATION IN SUGAR MAPLE IN RELATION TO ALTITUDE[1]

F. Thomas Ledig[2]

ABSTRACT

A nested sampling scheme was used to investigate phenotypic variation in sugar maple across an altitudinal transect. The transect extended from 570 ft. to 2,570 ft. above sea level in central New Hampshire. Forty of 42 leaf shape, size, and weight parameters, both measurements and ratios, varied significantly among trees within plots. It is tentatively concluded that at least some of the variation was genetic in origin because trees within plots were growing under very similar environments.

Twenty-three of 42 leaf characteristics were significantly correlated with elevation of the sample trees. The pattern of variation was not the same for all characteristics. Both negative and positive linear relationships with altitude existed and quadratic patterns with maxima at the intermediate elevations were common. The latter pattern for vigor traits (e.g. seasonal internode length) suggested a zone of optimal development in the altitudinal center of the transect. Some of the characteristics which varied in relation to the elevation of the sample tree may represent genetic adaptations; therefore, the force of natural selection must be stronger than that of gene migration across the transect.

INTRODUCTION

Transfer of seed across small horizontal distances but up or down a mountainside may result in failure of the stock to prosper or survive. In tree improvement it may be necessary to consider altitudinal variation when populations have become adapted to elevational zones.

There is little notion in forest trees of the relative importance of the opposing forces of selection and migration on altitudinal variation. Because of the relatively short distances involved and the influence of migration between adjoining populations, it is uncertain whether any appreciable genetic differentiation can be expected. However, some evolutionary biologists (e.g. Cook, 1961; Carson, 1959) feel that populations occupying marginal habitats, either geographically or with respect to some environmental factor such as altitude, may be centers of divergence. The objective of the present investigation was to determine whether migration was powerful enough to prevent adaptation to elevational zones or whether natural selection has resulted in differentiation.

---

[1] This is contribution No. 19 of the Hubbard Brook Ecosystem Study financed by National Science Foundation Grants GB 6742 and GB 6757 to F. H. Bormann and G. E. Likens. The author is Participating Scientist in this study. The assistance of Dr. R. S. Pierce of the USDA Forest Service is gratefully acknowledged.

[2] Assistant Professor of Forest Genetics, Yale University, School of Forestry, New Haven, Connecticut.

This report describes the initiation of a study of altitudinal variation in sugar maple (Acer saccharum Marsh). The phenotypic pattern of variation in leaf morphology was investigated as a first step in the study of adaptation. Leaf shape may be simply inherited, and the pattern of variation can be determined easily, providing information on the inheritance of more complex and economically important traits.

LITERATURE REVIEW

Previous studies of altitudinal variation in trees have been predominantly of western conifers, such as western white pine (Pinus monticola Dougl.), ponderosa pine (Pinus ponderosa Laws.), Jeffrey pine (Pinus jeffreyi Grev. and Balf.), and Douglas-fir LPseudotsuga menziesii (Mirb.) Franco]. These investigations have demonstrated variation in height and diameter growth, survival, stem taper, phenology, branch angle, and needle length (Barnes, 1967; Callaham and Liddicoet, 1961; Hermann and Lavender, 1968). In addition, other perennial plants have been divided into elevational ecotypes, particularly the Californian species investigated in the classic experiments of Clausen, Keck, and Hiesey (1940).

Most of the elevational populations investigated were separated by distances of a mile or more. Such distances might be sufficient to reduce gene migration, permitting differentiation to occur. Whether more local ecotypes can exist is a largely unexplored question. In cases of adaphic ecotypes, extremely narrow borders do separate very highly adapted and divergent populations of herbaceous species. Examples of the latter are the serpentine ecotypes in California (Kruckeberg, 1951) and lead-tolerant ecotypes in Great Britain (Wilkins, 1960); however, these populations may be partially maintained by vegetative reproduction, so that migration is not a force in preventing the development of adaptive gene complexes. In trees the best analyzed example of local ecotypes are the western white pine which occupy south--versus north--facing aspects of a single drainage (Squillace and Bingham, 1958). In this case, extremely intense selection must be required to maintain the differences which distinguished populations from the opposing slopes in the face of migration.

Leaf morphology provides convenient quantitatively measurable characteristics for a study of population differentiation in hardwoods. Leaf measurements and their ratios were used by Kellison (1967) to determine the relative importance of variation between physiographic regions, stands within regions, and trees within stands for tulip-poplar (Liriodendron tulipifera L.). He found differences among trees and among areas in the lateral sinus depth, the petiole length/leaf length ratio, leaf width/leaf length ratio, and other leaf measurement ratios. Based partially on phenotypic leaf characteristics, Kellison tentatively designated a deep peat ecotype occurring on the southern coastal plain of North Carolina, and in addition, found a cline of decreasing depth of leaf sinus with increasing length of the frost-free season of the source. In a study of phenotypic variation in sweetgum (Liquidambar styraciflua L.),Duncan (1959) tentatively concluded that there were genetic differences among trees in leaf shape. Subsequently, in a provenance test by Roberds[3] the leaf shape parameters measured by Duncan were shown to indeed vary genetically.

[3] Roberds, James Hall. 1965. Patterns of variation in several characteristics of sweetgum (Liquidambar styraciflua L.) in North Carolina. Unpublished M.S. thesis. North Carolina State University, Raleigh, North Carolina. 62 pp.

In conifers, various needle characteristics, such as number of serrations or number of stomatal rows, are accepted characteristics for diagnosis of natural variation (e.g. Critchfield, 1957; Mergen, 1963). Needle length generally decreases with an increase in altitude of the population (see review by Barnes, 1967).

METHODS

Study Area

The population of sugar maple investigated in this study extended from the banks of the Pemigewasset River at 570 ft. elevation to near the top of a ridge at 2,570 ft. elevation in the U. S. Forest Service's Hubbard Brook Experimental Forest, West Thornton, New Hampshire (Fig. 1). The length of the transect from the river to the ridge was only 3.25 miles and consisted entirely of southerly or southeasterly aspects.

The climate of the forest is humid continental. The high elevations are characterized by cooler summers, colder winters, deeper snow-pack, and shorter growing seasons than the low elevations. Annual precipitation is 123 cm. and does not appear to vary with elevation (personal communication, F. H. Bormann), despite the fact that the station at 2,570 ft. is occasionally enveloped in clouds. Personal observations over a limited period indicate that the clouds condense on the foliage of trees at the 2,570 ft. elevation. Details of the climate, soils, and cutting history of this watershed can be found in Likens et al. (1967) and in Bormann et al.[4], who present a vegetation analysis of a portion of this transect.

Sampling

A nested sampling scheme was used. At each of five elevations, spaced at 500-ft. intervals from 570 to 2,570 ft., two plots were located. Elevation was determined by topographic maps and by altimeter. On each of the plots, five trees were selected along the contour (as nearly as possible). The greatest elevational difference within an elevational station was 51 ft. and this was between trees in different plots. Generally, the difference was 30 ft. or less. Selected trees were dominants or codominants.

At both the highest (2,570 ft.) and the lowest (570 ft.) elevations, it was difficult to locate trees of sugar maple. The trees chosen at 2,570 ft. could be considered at the very margin of the altitudinal distribution of the species in this locality; there were none found further up the ridge. On the other hand, the lowest elevation was in the valley of the Pemigewasset River; there much of the land is under cultivation, so that trees are generally located only on the banks of water courses or in fence rows.

All trees were climbed, and two sample branch tips were chosen from the southern aspect above the midpoint of the crown and in full sunlight. Three leaves, as free of injury or disease as practical, were collected from each of the two sample branch tips. These leaves were usually the second pair of the

---

[4] Bormann, F. H., T. G. Siccama, G. E. Likens, and R. A. Whittaker. The Hubbard Brook Ecosystem Study: Dynamics of the tree vegetation. In preparation for Ecology.

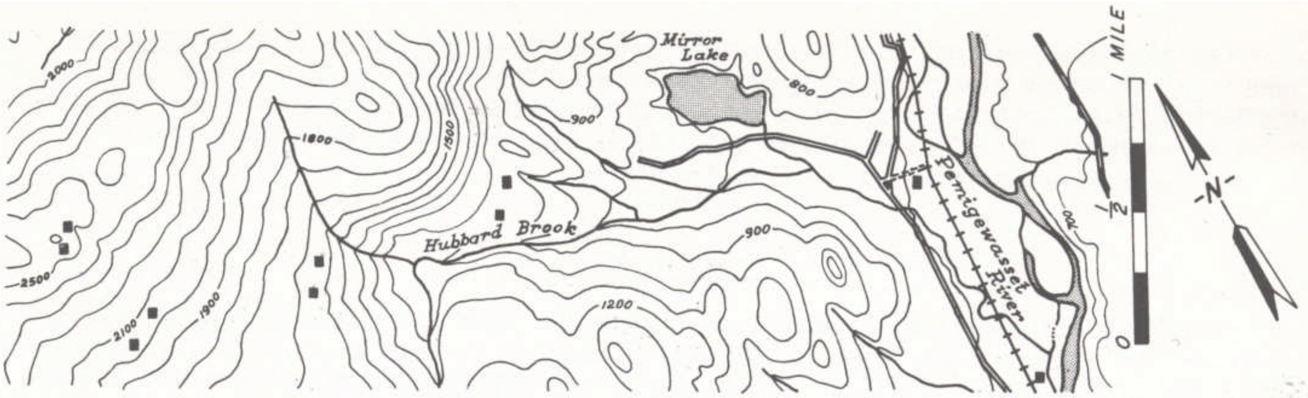


Fig. 1.--Contour map of the study area. Plot locations are shown by dots and mileage between elevational stations is indicated by scale.

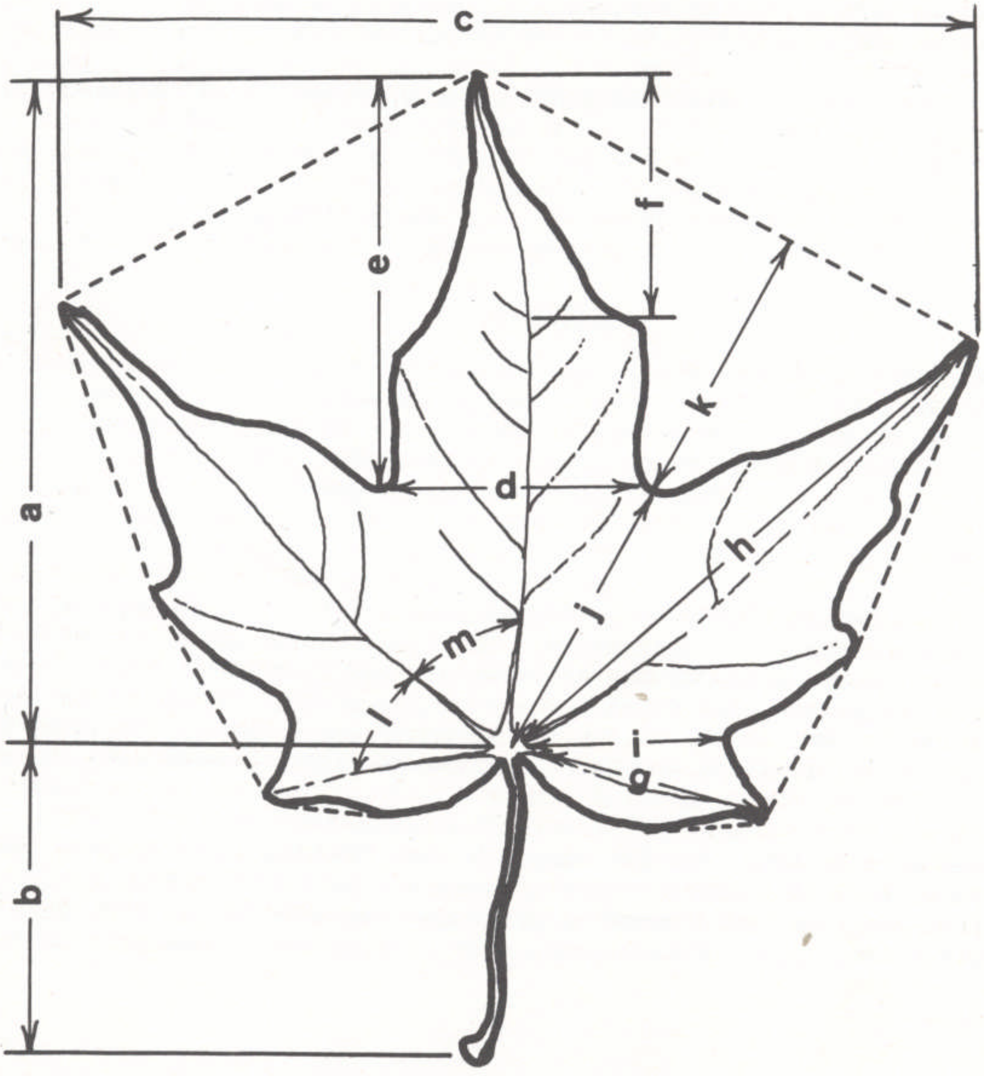


Fig. 2.--Diagram of a sugar maple leaf, showing basic measurements and circumscribed convex polygon: a = leaf length; b = petiole length; c = leaf width; d = inter-sinus width; e -length of apex; f= length of terminal extension; g = length of basal lobe; h= length of primary lobe; i =basal sinus radius; j =primary sinus radius; k = extension of primary sinus radius to perimeter of circumscribed convex polygon; 1=basal intervein angle; m= primary intervein angle.

season and one of the third pair, unless the presence of injuries made this impossible. Lengths of the last five seasonal internodes on the sample branch were recorded.

Leaf Measurement

Eighteen basic measurements were made for each of the six leaves per tree. Most of the measurements were made from leaf prints using a ruler graduated in millimeters (Fig. 2). Angles between major veins were measured to the nearest 5 with a protractor. Number of lobes (those with veins originating at the base of the leaf) and number of secondary lobes were counted. Area of the leaf and area of a circumscribed convex polygon were measured by planimeter. Air-dry weight of the pressed leaves was measured to the nearest milligram. From these measurements, an additional set of 24 variables was constructed as ratios (measurements and ratios are listed in Table 1).

RESULTS

Each one of the 42 measurements or ratios except the length of the leaf by the length of the primary lobe (a/h) and the primary sinus radius by the basal sinus radium (j/i) varied among trees within plots (Table 1) at (or nearly at, in the case of e/k) the one percent level of statistical significance. Only the two ratios, length of apex (polygon radius — sinus radius) and leaf air-dry weight by leaf area, varied between plots within elevational zones. Components of variance for trees, samples, and leaves were generally large, but for plots were often zero (Table 2).

Multiple measurements in space can be used to calculate repeatabilities (Falconer, 1960). Repeatability (R) is an upper limit to broad sense heritability and was calculated for the leaf parameters as:

$$R = \frac{\delta_t^2}{\delta_t^2 + \delta_s^2 + \delta_1^2}$$

where St is the among-tree component of variance, σ^2_s is the between sample component, and σ^2_1 is the among-leaves component. St contains the variance due to permanent environmental effects on the trees due to the microsites which they inhabit, plus total genetic variance. Estimates made for leaf measurements ranged from .02 to .61 but were, in general, moderately high (Table 2). Because the samples were restricted to a single position in the crown, variance due to localized environmental effects may have been eliminated from the denominator. In addition, year-to-year variation is not accounted for. Because a great deal of variation exists between sun and shade leaves and between years,[5] the elimination of these effects is likely to inflate the estimate of repeatability. The estimates, therefore, apply only to sun leaves taken from the southern aspect and above the level of the competing canopy, but they do suggest potentially important genetic effects on leaf shape.

---

[5] Gosz, J. R., G. E. Likens, J. S. Eaton, and F. H. Bormann. Leaching of materials from selected tree species in New Hampshire. In preparation for Plants and Soils.

Orthogonal contrasts were used to partition the variance among elevations into linear and quadratic components and a remainder. When this was done, variation among elevations was significant (5 percent or 1 percent level of probability) for 14 of the 42 variables. Regression analysis indicated statistically significant correlations with elevation for 23, or more than half, of the 42 variables (significant linear and quadratic regressions are in Table 3 and some of the relationships with respect to elevation are shown in Fig. 3). While many of the measurements were correlated (correlation matrices, Table 4 and Table 5), some varied relatively independently of others. A multiple regression was sought such that elevation of the source could be predicted based on a set of leaf variables. Of the many thousands of regressions computed, the best one utilized five variables and in some cases their squared terms; the coefficient of multiple determination was $R^2 = 0.68$. The equation was:

$$Y = 6886.5 - 2.5(x_1) + 148.5(x_2) - 1.0(x_2)^2 - 2405.5(x_3) - 19070.5(x_4) + 22397.0(x_4)^2 - 1879.0(x_5)$$

where Y = altitude of sample tree,

x_l = leaf weight,

x_2 = length of leaf apex (e),

x_s = primary sinus radius/basal sinus radius

(j/i), x_4 = leaf weight/leaf area,

x_s = primary intervein angle/basal intervein angle (m/l).

all variables made a contribution to the regression as indicated by t-tests with 42 degrees of freedom. The equation can be used to estimate the elevational origin of a tree in the Hubbard Brook Watershed based on leaf measurements. Of more interest, it indicated that there were complexes of characters which distinguished the elevational populations with a fair degree of assurance. This differentiation may be either genetic or environmental or both.

DISCUSSION

Trees within plots accounted for the major proportion of the variance for almost all characteristics measured. Trees within regions also accounted for the major portion of the variation in leaves of sweet gum in the investigation by Roberds [6]. The many differences among trees growing on the same site suggest that a great deal of genetic variation exists in leaf morphology. Because of the pleiotrophic nature of gene action, there is probably concomitant variation in other attributes, and some of these will be investigated in the future.

Adjacent individuals (i.e. those within plots) are more likely to be related than those further apart (i.e. in different plots). Harberd (1958) has warned that in genecology the unwitting selection of related individuals to represent a site will lead to an underestimate of the variance among trees and, therefore, tests of differences among habitats may prove falsely significant. Fortunately, the present

[6] Roberds, op. cit.

sample has been adequate to avoid this situation as shown by the comparatively high tree-to-tree variance in relation to the variance between plots. Therefore, in this study tests of differences among elevational zones were valid.

Because differences in leaf morphology among trees growing on the same sites could be tentatively interpreted as at least partly genetic, habitat- correlated variation in some of the same characteristics might be regarded as evidence of genetic adaptation. For example, the relationship of leaf dry weight per unit area to elevation was relatively strong (Fig. 3A). Leaf dry weight per unit area was relatively low at the high (2,570 ft.) elevation with an apparent maximum in the intermediate zone. Low dry weight per unit area is characteristic of shade leaves in sugar maple. However, all leaves collected were sun leaves. In addition, the sites at 2,570 ft. were characterized by a lower basal area or greater openness than the intermediate elevations on which sugar maple seemed to make its best development and where it was the major component of the stands. Thus, the trend in dry weight per unit leaf area would seem to reflect some real adaptive variation in the population. The situation in sugar maple might be analogous to that of the shade and sun ecotypes of goldenrod (Solidago virgaurea L.), described by Björkman and Holmgren (1963) and Holmgren (1968). The ecotypes from exposed habitats (i.e. alpine heaths) had a lower leaf dry weight per unit area than ecotypes from shaded habitats (i.e. forests). This distinction held regardless of whether the ecotypes were grown under conditions of high or low irradiation (Holmgren, 1968). The morphological distinction between the ecotypes of goldenrod had an effect on productivity; the exposed (or sun) ecotypes had lower mesophyll resistance to CO2 diffusion and were able to utilize higher light intensities for photosynthesis than plants from shaded habitats. It will be interesting to determine whether such adaptations have evolved in sugar maple, allowing high altitude populations to utilize the higher levels of irradiance which occur at high elevations. Likewise, in those species which have been investigated, shade ecotypes are adapted to growth within dense stands under forest canopies where ability to utilize low light intensities efficiently is important to survival.

The ratio (k + j)/j is also of interest. It is a measure of the relative depth of the sinus, and was significantly and positively correlated with altitude (r = 0.42). In tulip-poplar, depth of the top sinus was negatively correlated with length of the frost-free season across a geographic and physiographic gradient (Kellison, 1967). Thus, behavior of sugar maple was analogous to that of tulip-poplar, because the frost-free season decreases with altitude at Hubbard Brook.

For many other characteristics, a quadratic response to elevation was noted. Variables often took low values at both the highest and lowest elevations and higher values at intermediate elevations (Fig. 3B). Trees at the three intermediate elevations (1,070- 2,070 ft.) were generally similar and had maximum values for measurements supposedly reflecting vigor, (e.g. petiole length, leaf length). Seasonal internode lengths for the five years prior to 1968 were longest in the sample from 1,570-ft.

elevation (R- = 0.45 ft.), were slightly shorter at 1,070 ft. and 2,070 ft. (7 = 0.42 and 0.40 ft., respectively), and were shortest at 570 ft. and 2,570 ft. (7 = 0.35 and 0.20 ft., respectively). This suggests that there is an altitudinal zone of optimal development in the center of the elevational range in this locality. Such a zone was hypothesized by Squillace (1966) for slash pine in the center of its geographic range. This has practical significance because gains may be achieved by planting

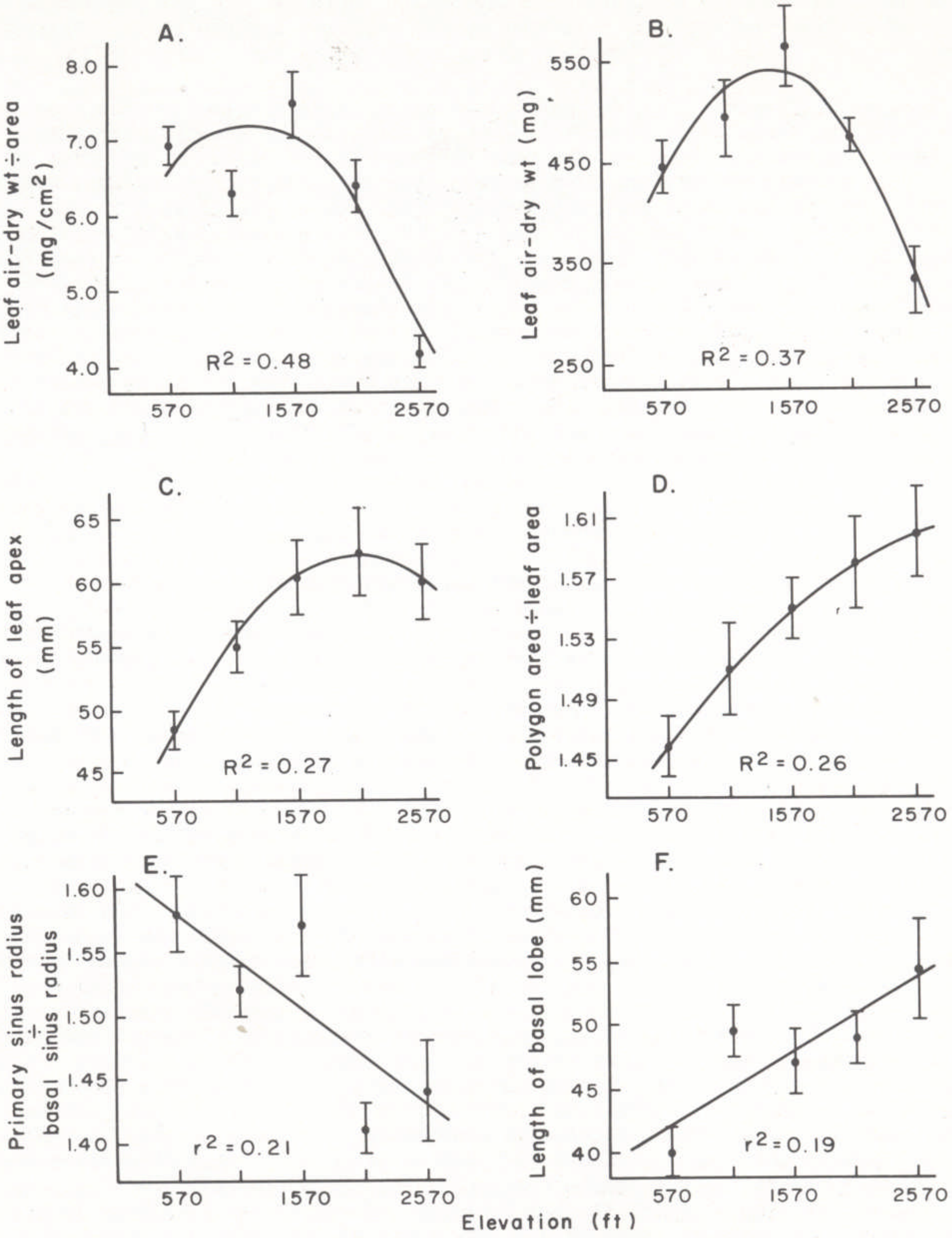


Fig. 3.--Relationship of six selected leaf characteristics of sugar maple to elevation of the tree at Hubbard Brook Experimental Forest, New Hampshire: A = leaf air-dry weight leaf area; B = leaf air-dry weight; C = length of leaf apex (e); D = area of circumscribed convex polygon % leaf area; E = primary sinus radius t basal sinus radius (jA); F = length of basal lobe (g), (n = 10, bars indicate ± s_x).

optimal races in place of non-optimal local races (Namkoong[7]); e.g. by planting progeny of trees from the 1,570 ft. population at lower and higher elevations.

Occasionally when quadratic patterns were evident in the relationship of a variable to elevation, there was an increase in value up to 2,070 ft. and then a break in slope (Fig. 3C). Bormann et al.8 note an extreme reduction in till depth at 2,400 ft. and consider 2,400-2,500 ft. as the lower limit of a zone sharply defined by extended periods of snow cover, frequent cloud envelopment, high and drying northwest winds, and reduced frost-free period. Thus, the populations at 2,070 ft. and at 2,570 ft. are occupying strikingly different environments. The sugar maple at 2,570 ft. were somewhat protected against gene flow from the central (elevational) population because of a greatly reduced density of individuals near the elevational extreme and differences between populations in flowering phenology. At 2,400 ft. the density of sugar maple begins to drop off drastically and it becomes an insignificant component of the stand. The 2,570-ft. population and, to a degree, the 570-ft. population represented marginal populations in the study area. Bormann et al. place an elevational limit of 2,500 ft. on sugar maple in this area and suggest that a similar situation prevails in northern Vermont and New York.

When the relationship between leaf parameters and elevation was linear, there was generally a decrease in value of the variable with increasing elevation (i.e. a negative correlation, see Fig. 3E). In the case of simple measurements (not ratios), this might reflect a general decline in site quality with increase in elevation or, conversely, an improvement in the environment as one proceeds downslope. Such differences may represent environmental modification of the phenotype. Occasional exceptions to the general trend for a decrease in value of the leaf parameters with elevation were also noted (e.g. Fig. 3F).

Distinctly different individuals could be selected over a 2,000-ft. range in elevation because of the important effect of altitude on climatic factors. According to Hopkins' Rule (Hopkins, 1918), 4 days lag in phenology is expected for every 400 ft. increase in elevation. During a phenology study on part of the transect used in this investigation, there was a 5-day delay in bud opening from 1,810 ft. to 2,580 ft. in 1966, while in 1967 the delay was 7 days from 1,818 ft. to 2,450 ft.[10]. Based on Hopkins'Rule there should be a 20-day lag in phenological events across the elevational transect reported here, and extrapolating the observations on this watershed, a 13- to 22-day delay. Thus, there is a substantial difference in the length of the growing season although the horizontal distance across the entire transect is only 3.25 miles. The differences in phenology are some indication of the seasonal difference in temperature regimes. Therefore, there was ample reason to expect that different individuals would be selected over the transect.

[7] Namkoong, Gene. 1969. The non-optimality of local races. In Proceedings of the Tenth Southern Conference on Forest Tree Improvement. Houston, Texas. In press.

[8] Bormann, op. cit.

[9] Bormann, ibid.

[10] Mahall, Bruce E., and F. Herbert Bormann. The herbaceous phenology of a northern hardwood forest over an altitudinal gradient. In preparation for Ecology.

On the other hand, Wright (1953) warned that a difference of 1,000 ft. in altitude would not result in the same degree of genetic differentiation as 250 miles in latitude because gene migration can be a potent force across the shorter distance of an elevational transect. In addition, it will be difficult for small marginal populations to become adapted because the total amount of recombination and of mutation will be so low (because of the small number of individuals) that the probability of favorable events being available for selection becomes very rare.

However, the present study suggests that populations separated by less than one-half mile (e.g. the 2,070-ft. vs the 2,570-ft. elevation) may be quite different and the observed variation is correlated with elevation. If the observed variation is genetic, then selection must be very intense or migration rates surprisingly low. The former explanation is favored although migration is probably minimal between the 2,070-ft. and 2,570-ft. elevations because of the very low density of the breeding population above 2,400 ft. and differences between populations in phenology.

In white fir, morphological needle characteristics did not vary as much across an altitudinal transect as they did across the equivalent latitudinal range, supporting Wright's (1953) hypothesis that 1,000 ft. in altitude is not equivalent to 250 miles in latitude (Hamrick[11]). Variation in growth response, however, was as great in relation to altitude as to latitude. Therefore, it might be that growth traits in the sugar maple investigated here would show an even greater response to altitude than that observed for leaf traits whose survival value is obscure.

SUMMARY AND CONCLUSIONS

Measurements and their ratios, expressing aspects of leaf shape, exhibit a surprising amount of variation among trees growing in close proximity on the same site. This suggests genetic variation. However, until experiments can be performed in controlled environments, this conclusion can be only tentative.

Habitat-correlated variation has been used by some genecologists as evidence of divergent selection resulting in adaptation and genetic differentiation. Correlations and multiple regressions were found between leaf-shape parameters and elevation in this study and may indicate the influence of natural selection. However, it could be argued with nearly equal likelihood that the correlations reflect environmental modifications in relation to some factors associated with altitude. The hypothesis that these correlations reflect population adaptation was favored because the characteristics which varied across the altitudinal transect also showed statistically significant differences from tree to tree on the same site.

If the differences among populations are genetic, then selection intensities must be high to maintain such differentiation in the face of migration between populations below 2,100 ft. Above this limit, migration rates may be low because of the greatly reduced density of sugar maple.

[11] Hamrick, James Lewis, III. 1966. Geographic variation in white fir. Unpublished M.S. thesis. University of California, Berkeley, California. 103 pp.

The pattern of variation suggests an optimal zone of development for sugar maple near 1,570-ft. elevation, the altitudinal center of the population in this locality.

LITERATURE CITED

Barnes, Burton V. 1967. Phenotypic variation associated with elevation in western white pine. Forest Sci. l3:357-364.

Bjorkman, 011e, and Paul Holmgren. 1963. Adaptability of the photosynthetic apparatus to light intensity in ecotypes from exposed and shaded habitats. Physiologia Plantarum 16:889-914.

Callaham, R. Z., and A. R. Liddicoet. 1961. Altitudinal variation at 20 years in ponderosa and jeffrey pines. Jour. Forestry 59:814-820.

Carson, H. L. 1959. The genetic characteristics of marginal populations of _Drosophila._ Cold Spring Harbor Symposium on Quantitative Biology 20: 276-286.

Clausen, Jens, David D. Keck, and William M. Hiesey. 1940. Experimental studies on the nature of species. I. Effect of varied environments on Western North American plants. Carnegie Inst. of Washington Publ. No. 520. Washington, D. C. 452 pp.

Cook, L. M. 1961. The edge effect in population genetics. Amer. Naturalist 95:295-307.

Critchfield, William B. 1957. Geographic variation in _Pinus contorta._ Maria Moors Cabot Found. Publ. No. 3. Harvard University, Cambridge, Massachusetts. 118 pp.

Duncan, Wilbur H. 1959. Leaf variation in _Liquidambar styraciflua_ L. Castanea 24:99-111.

Falconer, D. S. 1960. Introduction to quantitative genetics. Ronald Press, New York. 365 pp.

Harberd, D. J. 1958. A spurious significance in genecological trials. Nature 181:138.

Hermann, Richard K., and Denis P. Lavender. 1968. Early growth of Douglas-fir from various altitudes and aspects in southern Oregon. Silv. Genet. 17: 143-151.

Holmgren, Paul. 1968. Leaf factors affecting light-saturated photoshythesis in ecotypes of _Solidago virgaurea_ from exposed and shaded habitats. Physiologia Plantarum 21:676-698.

Hopkins, A. D. 1918. Periodical events and natural laws as guides to agricultural research and practice. USDA Monthly Weather Review, Supplement 9:1-42.

Kellison, Robert Clay. 1967. A geographic variation study of yellow-poplar _(Liriodendron tulipifera_ L.) within North Carolina. Tech. Report No. 33, School of Forestry, North Carolina State Univ., Raleigh, N. C. 41 pp.

Kruckeberg, Arthur R. 1951. Intraspecific variability in the response of certain native plant species to serpentine soil. Amer. Jour. Bot. 38:408-419.

Likens, G. E., F. H. Bormann, N. M. Johnson, and R. S. Pierce. 1967. The calcium, magnesium, potassium, and sodium budgets for a small forested ecosystem. Ecology 48:772-785.

Mergen, Francois. 1963. Ecotypic variation in _Pinus strobus_ L. Ecology 44: 716-727.

Squillace, A. E. 1966. Geographic variation in slash pine. Forest Sci. Monograph 10. Soc. Amer. Foresters, Washington, D. C. 56 pp.

Squillace, A. E., and R. T. Bingham. 1958. Localized ecotypic variation in western white pine. Forest Sci. 4:20-34.

Wilkins, D. A. 1960. Recognizing adaptive variants. In Proc. of the Linnean Soc. of London, 171 Session 1958-59, Part 1. pp. 122-126.

Wright, Jonathan W. 1953. Pollen-dispersion studies: some practical applications. Jour. Forestry 51:114-118.

Table 1. F-ratios and significance levels for 42 leaf characteristics and ratios analyzed by nested analysis of variance for elevations (pooled plus the linear and quadratic components), plots within elevations, trees within plots, samples within trees, and leaves within samples

Variable 1/	Elevations			Plots	Trees	Samples
	pooled	linear	quadratic			
Measurements						
Leaf Length (a)	1.82	3.72*	3.05*	0.92	3.81***	2.62***
Petiole length (b)	1.22	0.11	4.71**	0.31	2.21***	1.04
Leaf width (c)	1.73	5.71*	0.68	1.57	3.65***	2.01***
Intersinus width (d)	0.24	0.00	0.56	1.95	5.26***	1.35*
Length of leaf apex (e)	4.31***	12.49***	4.54**	0.47	3.92***	3.25***
Terminal extension (f)	0.97	3.56	0.05	1.14	3.36***	1.37*
Length of basal lobe (g)	2.14	6.39*	0.04	1.92	4.26***	2.29***
Length of primary lobe (h)	2.21	3.48	4.23*	1.13	2.97***	2.19***
Basal sinus radius (i)	0.89	1.28	0.00	1.43	4.93***	1.85***
Primary sinus radius (j)	0.84	0.32	0.06	1.36	5.08***	1.45**
Sinal chord (j+k)	2.19*	3.22*	4.31**	0.89	3.60***	2.57***
Basal intervein angle (l)	1.46	0.03	0.29	0.98	4.69***	0.66
Primary intervein angle (m)	1.74	0.00	3.64	1.15	5.51***	1.02
Air-dry leaf weight	7.28***	6.17**	21.14***	0.28	2.29***	2.11***
Number of lobes	2.12*	0.20	1.08	0.81	10.76***	0.85
Number of secondary lobes	1.71	3.41	1.94	1.13	4.84***	2.28***
Area of circumscribed convex polygon	1.33	3.97	0.84	1.75	3.64***	3.33***
Area of leaf	0.62	1.42	0.46	2.18*	4.05***	2.76***
Ratios						
c/d	4.76*	11.36**	5.34*	1.12	2.84***	1.78***
c/a	0.82	2.19	0.75	2.35*	3.88***	0.66
a/e	5.10***	15.79***	2.32	0.65	5.14***	2.53***
e/f	1.80	1.54	5.37*	1.79	2.30***	1.31*
a/h	1.94	0.00	3.32*	0.81	1.13	0.99
(k+j)/j	2.99	7.05**	3.40	1.76	3.13***	2.11***

(continued)

Table 1. (continued)

Variable	Elevations			Plots	Trees	Samples
	pooled	linear	quadratic			
<u>Ratios</u>						
j/i	5.51***	14.27***	0.04	0.48	1.44	1.88***
a/d	1.99	2.19	5.07*	1.78	4.60***	2.05***
h/g	2.85	6.92**	0.96	1.66	2.24***	2.15***
e/k	1.38	1.19	3.12	3.01**	2.07**	1.11
a/b	0.97	2.27	1.59	0.20	2.27***	1.07
i/e	2.71**	4.75**	3.01*	0.96	4.71***	2.54***
a/i	1.68	0.22	2.73	0.90	3.93***	1.72***
a/g	2.48	6.08*	0.24	1.67	2.62***	2.17***
g/f	1.69	2.13	0.53	1.05	2.48***	1.27
m/l	2.81	0.00	9.88**	1.16	1.96***	0.77
e/g	1.31	0.02	1.03	1.48	3.94***	1.99***
h/e	3.53**	10.26***	0.67	0.86	5.11***	1.35*
Polygon area/leaf area	2.73	10.50**	0.42	1.57	8.30***	1.16
Leaf weight/leaf area	5.63**	10.71**	7.48**	3.74**	2.14***	3.10***
No. of secondary lobes/a	0.67	1.30	0.34	0.49	4.83***	2.13***
No. of secondary lobes/e	0.42	0.36	0.00	0.66	4.49***	2.07***
No. of secondary lobes/h	0.83	1.33	0.08	0.53	4.52***	1.93***
m/polygon area	2.92	4.69*	5.24*	1.17	4.44***	1.60***

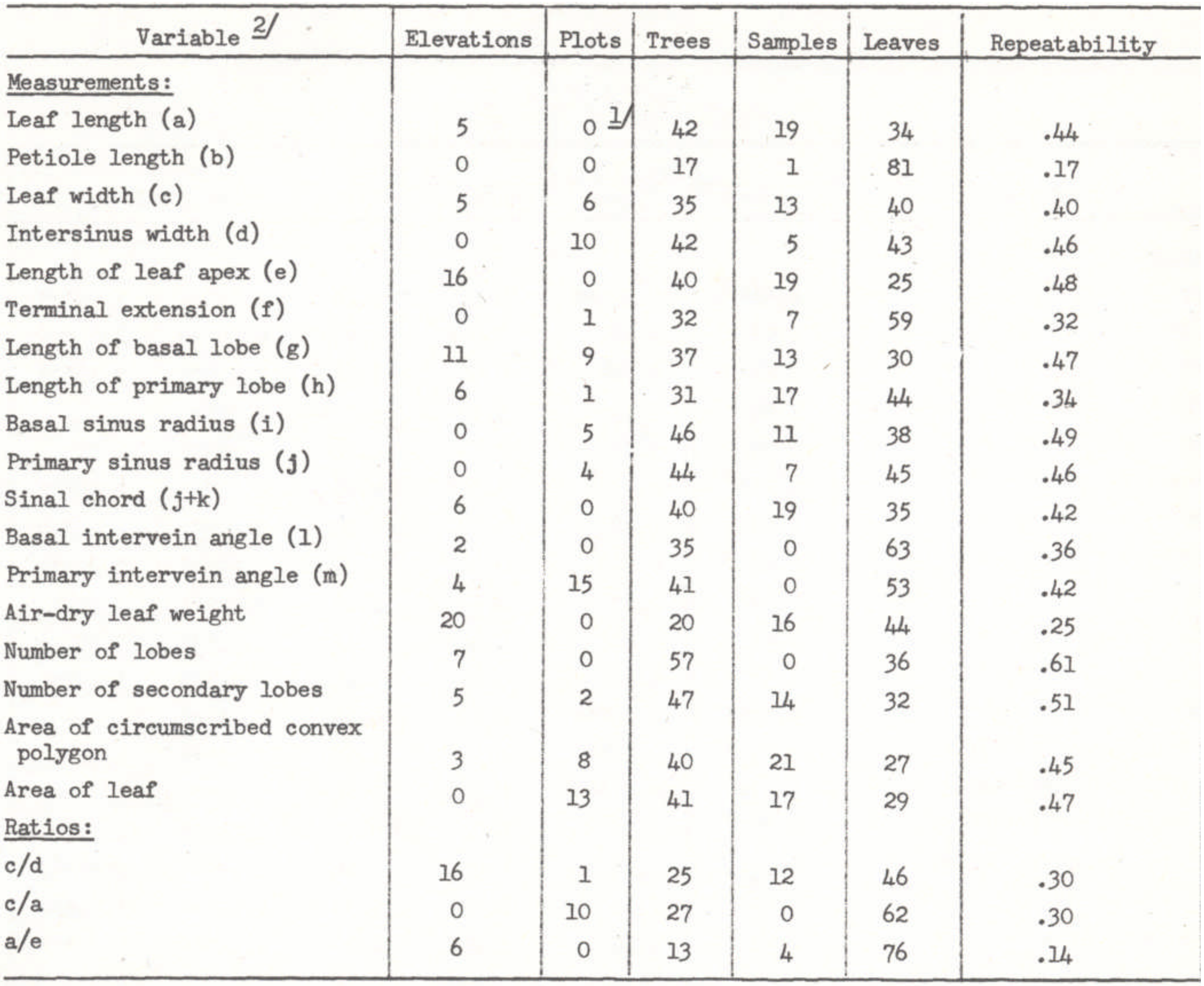
[1] Letters refer to Fig. 2

* 10 percent level of significance

** 5 percent level of significance

*** 1 percent level of significance

Table 2. Components of variance expressed as percent of the total variance for elevations, plots within elevations, trees within plots, samples within trees, and leaves within samples and repeatabilities for 42 leaf characteristics and ratios



Variable [2]	Elevations	Plots	Trees	Samples	Leaves	Repeatability
Measurements:						
Leaf length (a)	5	0 [1]	42	19	34	.44
Petiole length (b)	0	0	17	1	81	.17
Leaf width (c)	5	6	35	13	40	.40
Intersinus width (d)	0	10	42	5	43	.46
Length of leaf apex (e)	16	0	40	19	25	.48
Terminal extension (f)	0	1	32	7	59	.32
Length of basal lobe (g)	11	9	37	13	30	.47
Length of primary lobe (h)	6	1	31	17	44	.34
Basal sinus radius (i)	0	5	46	11	38	.49
Primary sinus radius (j)	0	4	44	7	45	.46
Sinal chord (j+k)	6	0	40	19	35	.42
Basal intervein angle (l)	2	0	35	0	63	.36
Primary intervein angle (m)	4	15	41	0	53	.42
Air-dry leaf weight	20	0	20	16	44	.25
Number of lobes	7	0	57	0	36	.61
Number of secondary lobes	5	2	47	14	32	.51
Area of circumscribed convex polygon	3	8	40	21	27	.45
Area of leaf	0	13	41	17	29	.47
Ratios:						
c/d	16	1	25	12	46	.30
c/a	0	10	27	0	62	.30
a/e	6	0	13	4	76	.14

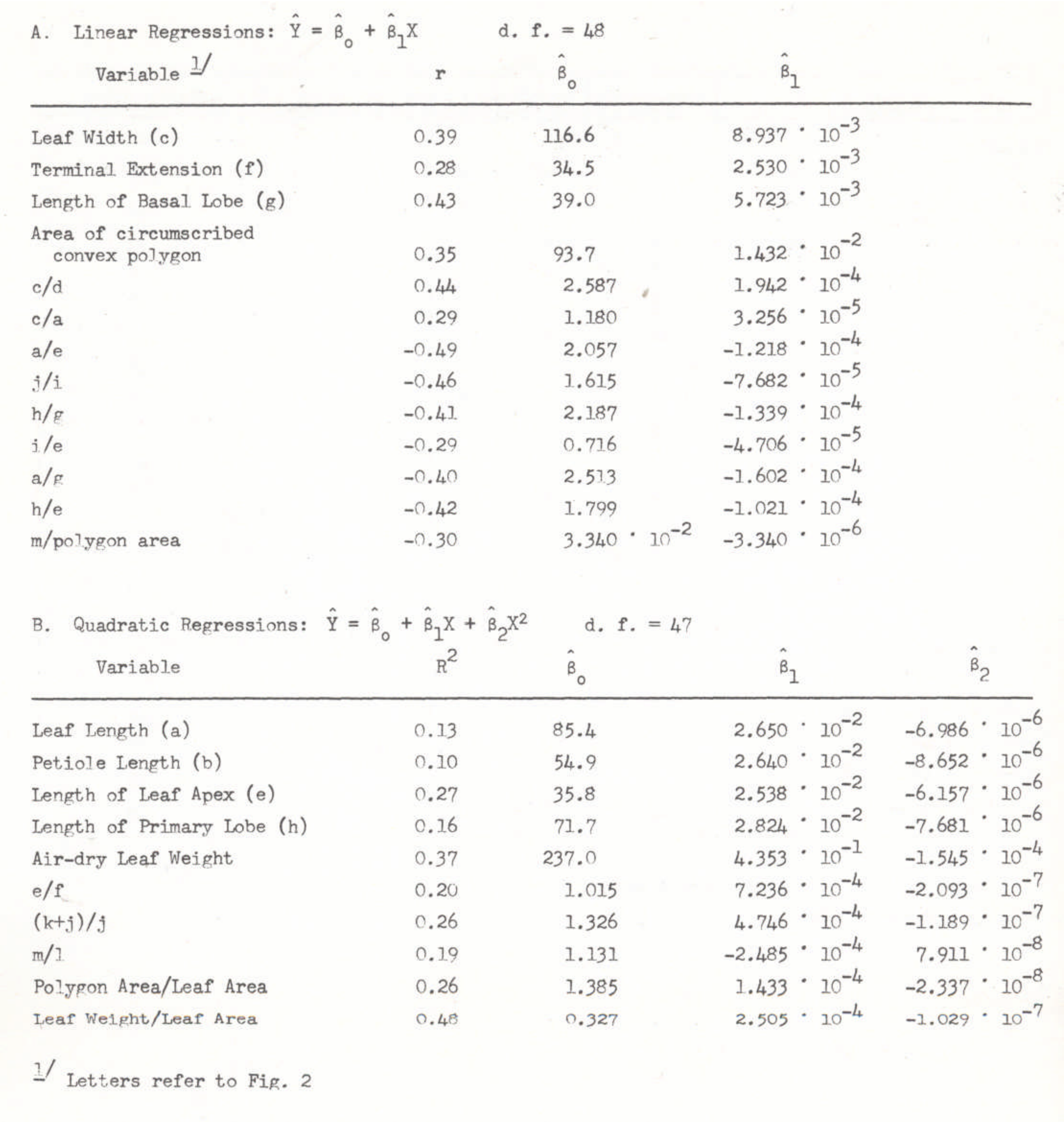
Table 2. (continued)

Variable	Elevations	Plots	Trees	Samples	Leaves	Repeatability
Ratios:						
e/f	5	0	18	7	65	.20
a/h	2	0	2	0	96	.02
(k+j)/j	14	6	28	14	37	.35
j/i	12	0	9	18	62	.10
a/d	8	7	43	11	31	.51
h/g	11	5	20	18	46	.24
e/k	3	11	14	2	70	.16
a/b	0	0	18	2	80	.18
i/e	10	0	46	15	29	.51
a/i	3	0	39	11	46	.40
a/g	10	5	25	17	43	.30
g/f	3	0	22	6	69	.22
m/l	90	0	1	0	9	.13
e/g	2	5	39	13	40	.42
h/e	12	0	40	5	43	.45
Polygon area/leaf area	14	6	46	2	32	.57
Leaf weight/leaf area	40	13	12	15	21	.26
Number of secondary lobes/a	0	0	61	0	39	.61
Number of secondary lobes/c	0	0	47	14	39	.47
Number of secondary lobes/h	0	0	46	13	41	.46
m/polygon area	12	2	38	8	41	.43

[1] Negative components were estimated as zero

[2] Letters refer to Fig. 2

Table 3. Linear and quadratic regressions for tree means of 23 leaf characteristics or ratios on altitude of the sample tree (significant at 5 percent or] percent level of probability)



A. Linear Regressions: $\hat{Y} = \hat{\beta}_o + \hat{\beta}_1 X$ d. f. = 48

Variable 1/	r	$\hat{\beta}_o$	$\hat{\beta}_1$
Leaf Width (c)	0.39	116.6	$8.937 \cdot 10^{-3}$
Terminal Extension (f)	0.28	34.5	$2.530 \cdot 10^{-3}$
Length of Basal Lobe (g)	0.43	39.0	$5.723 \cdot 10^{-3}$
Area of circumscribed convex polygon	0.35	93.7	$1.432 \cdot 10^{-2}$
c/d	0.44	2.587	$1.942 \cdot 10^{-4}$
c/a	0.29	1.180	$3.256 \cdot 10^{-5}$
a/e	-0.49	2.057	$-1.218 \cdot 10^{-4}$
j/i	-0.46	1.615	$-7.682 \cdot 10^{-5}$
h/g	-0.41	2.187	$-1.339 \cdot 10^{-4}$
i/e	-0.29	0.716	$-4.706 \cdot 10^{-5}$
a/g	-0.40	2.513	$-1.602 \cdot 10^{-4}$
h/e	-0.42	1.799	$-1.021 \cdot 10^{-4}$
m/polygon area	-0.30	$3.340 \cdot 10^{-2}$	$-3.340 \cdot 10^{-6}$

B. Quadratic Regressions: $\hat{Y} = \hat{\beta}_o + \hat{\beta}_1 X + \hat{\beta}_2 X^2$ d. f. = 47

Variable	R^2	$\hat{\beta}_o$	$\hat{\beta}_1$	$\hat{\beta}_2$
Leaf Length (a)	0.13	85.4	$2.650 \cdot 10^{-2}$	$-6.986 \cdot 10^{-6}$
Petiole Length (b)	0.10	54.9	$2.640 \cdot 10^{-2}$	$-8.652 \cdot 10^{-6}$
Length of Leaf Apex (e)	0.27	35.8	$2.538 \cdot 10^{-2}$	$-6.157 \cdot 10^{-6}$
Length of Primary Lobe (h)	0.16	71.7	$2.824 \cdot 10^{-2}$	$-7.681 \cdot 10^{-6}$
Air-dry Leaf Weight	0.37	237.0	$4.353 \cdot 10^{-1}$	$-1.545 \cdot 10^{-4}$
e/f	0.20	1.015	$7.236 \cdot 10^{-4}$	$-2.093 \cdot 10^{-7}$
(k+j)/j	0.26	1.326	$4.746 \cdot 10^{-4}$	$-1.189 \cdot 10^{-7}$
m/l	0.19	1.131	$-2.485 \cdot 10^{-4}$	$7.911 \cdot 10^{-8}$
Polygon Area/Leaf Area	0.26	1.385	$1.433 \cdot 10^{-4}$	$-2.337 \cdot 10^{-8}$
Leaf Weight/Leaf Area	0.48	0.327	$2.505 \cdot 10^{-4}$	$-1.029 \cdot 10^{-7}$

1/ Letters refer to Fig. 2

Table 4. Correlation coefficients between 18 leaf attributes in sugar maple[1]

Leaf Attributes[2]	Leaf Length (a)	Petiole Length (b)	Leaf Width (c)	Inter-sinus Width (d)	Length of Leaf Apex (e)	Terminal Extension (f)	Length of Basal Lobe (g)	Length of Primary Lobe (h)	Basal Sinus Radius (i)	Primary Sinus Radius (j)	Sinal Chord (j+k)	Basal In-tervein Angle (l)	Primary Intervein Angle (m)	Air-dry Leaf Weight	Number of Lobes	Number of Secondary Lobes	Polygon Area
Petiole Length(b)	0.48																
Leaf Width (c)	0.87	0.43															
Intersinus Width(d)	0.47	0.26	0.58														
Length of Leaf Apex(e)	0.85	0.29	0.80	0.18													
Terminal Extension(f)	0.65	0.24	0.79	0.50	0.73												
Length of Basal Lobe(g)	0.78	0.40	0.80	0.43	0.60	0.50											
Length of Primary Lobe(h)	0.97	0.50	0.85	0.41	0.80	0.55	0.82										
Basal Sinus Radius(i)	0.73	0.42	0.69	0.60	0.38	0.36	0.87	0.78									
Primary Sinus Radius(j)	0.63	0.47	0.50	0.70	0.16	0.19	0.61	0.65	0.83								
Sinal Chord (j+k)	0.98	0.52	0.83	0.41	0.80	0.54	0.79	0.98	0.76	0.67							
Basal Inter-vein Angle(l)	-0.20	-0.13	0.00	0.12	0.16	0.10	-0.35	-0.24	-0.43	-0.36	-0.26						
Primary Inter-vein Angle(m)	-0.29	-0.31	0.02	0.17	-0.04	0.27	-0.30	-0.37	-0.36	-0.44	-0.39	0.69					
Air-dry Leaf Weight	0.49	0.49	0.38	0.35	0.32	0.22	0.26	0.51	0.44	0.48	0.50	-0.01	-0.09				
Number of Lobes	0.24	0.04	0.29	0.22	0.08	0.18	0.51	0.24	0.43	0.30	0.23	-0.20	-0.11	-0.04			
Number of Secon-dary Lobes	0.49	0.33	0.50	0.15	0.48	0.41	0.55	0.46	0.32	0.20	0.47	-0.21	-0.14	0.13	0.25		
Polygon Area	0.95	0.47	0.96	0.55	0.83	0.72	0.86	0.93	0.76	0.58	0.92	-0.08	-0.11	0.41	0.34	0.53	
Leaf Area	0.91	0.49	0.92	0.67	0.70	0.67	0.85	0.90	0.84	0.70	0.89	-0.09	-0.10	0.49	0.39	0.51	0.97

---

[1] Correlations between tree means unadjusted for elevation or plot effects; coefficients are significant at $P \leq .05$ if larger than 0.28 and significant at $P \leq .01$ if larger than 0.36

[2] Letters refer to Figure 2.

Table 5. Correlation coefficients between 24 leaf shape ratios in sugar maple [1]

Leaf Measurement Ratios [2]	c/d	c/a	a/e	e/f	a/h	(k+j)/j	j/i	a/d	h/g	e/k	a/b	i/e	a/i	a/g	g/f	m/l	e/g	h/e	Polygon Area /Leaf Area	Leaf Weight /Leaf Area	No. of Secondary Lobes/a	No. of Secondary Lobes/e	No. of Secondary Lobes/h
c/a	0.15																						
a/e	-0.69	-0.18																					
e/f	0.43	-0.48	-0.35																				
a/h	-0.21	-0.11	-0.17	-0.22																			
(k+j)/j	0.83	0.01	-0.87	0.46	0.01																		
j/i	-0.47	-0.30	0.30	-0.02	0.29	-0.42																	
a/d	0.84	-0.41	-0.54	0.68	-0.15	0.77	-0.28																
h/g	-0.15	-0.32	-0.09	0.10	0.20	0.80	0.63	0.03															
e/k	-0.69	0.25	0.34	-0.58	0.31	-0.63	0.36	-0.78	0.07														
a/b	0.10	-0.03	-0.31	0.13	0.21	0.24	-0.22	0.11	-0.10	-0.05													
i/e	-0.57	0.02	0.92	-0.37	-0.32	-0.81	-0.05	-0.53	-0.36	0.32	-0.24												
a/i	0.40	-0.17	-0.68	0.35	0.37	0.63	0.40	0.46	0.58	-0.22	0.14	-0.90											
a/g	-0.19	-0.33	-0.13	0.04	0.41	0.08	0.65	0.00	0.97	0.14	-0.04	-0.42	0.64										
g/f	0.13	-0.21	0.31	0.54	-0.51	-0.13	-0.29	0.25	-0.59	-0.38	-0.03	0.45	-0.48	-0.67									
m/l	-0.03	0.26	-0.05	-0.37	0.19	-0.06	-0.32	-0.18	-0.21	0.17	0.17	0.09	-0.22	-0.14	-0.17								
e/g	0.25	-0.15	-0.64	0.23	0.38	0.55	0.33	0.31	0.81	-0.08	0.–4	-0.81	0.87	0.84	-0.67	-0.09							
h/e	-0.55	-0.10	0.96	-0.25	-0.45	-0.79	0.17	-0.45	-0.16	0.21	-0.33	0.93	-0.73	-0.25	0.44	-0.10	-0.70						
Polygon Area /Leaf Area	0.72	0.05	-0.78	0.40	0.01	0.78	-0.25	0.64	0.31	-0.55	0.27	-0.75	0.59	0.40	-0.09	-0.02	0.46	-0.70					
Leaf Weight /Leaf Area	-0.22	-0.35	0.09	0.18	-0.09	-0.11	0.31	-0.01	0.55	0.07	-0.34	-0.02	0.12	0.49	-0.18	-0.13	0.33	0.09	-0.20				
No. of Secondary Lobes/a	-0.14	0.11	0.34	-0.19	-0.07	-0.25	0.01	-0.19	-0.34	0.06	0.20	0.32	-0.25	-0.33	0.21	0.13	-0.43	0.32	-0.35	-0.20			
No. of Secondary Lobes/e	0.20	0.19	-0.15	-0.02	0.02	0.18	-0.14	0.08	-0.32	-0.10	-0.04	-0.13	0.08	-0.29	0.61	0.17	-0.13	-0.15	0.03	-0.26	0.87		
No. of Secondary Lobes/h	0.16	0.16	-0.18	-0.06	0.18	0.18	-0.09	0.06	-0.28	-0.05	0.00	-0.18	0.15	-0.21	-0.03	0.20	-0.06	-0.22	0.04	-0.27	0.84	0.99	
m/Polygon Area	-0.42	0.06	0.11	-0.27	0.17	-0.28	0.37	-0.43	0.46	0.43	-0.06	0.27	0.11	0.48	-0.46	0.15	0.30	0.04	-0.16	0.29	-0.01	-0.32	-0.01

---

[1] Correlations between tree means unadjusted for elevation or plot effects; coefficients are significant at $P \leq .05$ if larger than 0.28 and significant at $P \leq .01$ if larger than 0.36.

[2] Letters refer to Figure 2.

DISCUSSION

SOLOMON - Tom, one of the questions that always comes up, in taking measurements such as this, is the element of site. Do you feel you have a difference in site over the planted area?

LEDIG - Yes, we know of several differences across the elevational gradient, because Hubbard Brook is pretty well monitored. At 2,400 feet, for example, depth of the till decreases drastically. It is very shallow above 2,400 feet, and below that elevation it is very deep--a rather sharp break. Twenty-four hundred (2400)feet seems to be the breaking point for many environmental variables. Above this level there is a very deep snow pack. The snow pack remains late in the season. Clouds frequently envelop the ridge above 2,400 feet so that there is a lot of condensation on the leaves--a lot of drip. When we were working up there it felt like it was raining underneath the stand, but in the clearing it was merely foggy. Also, I have some data on what the differences in phenology might be across the gradient, based on some work that Bruce Mahall did there with the Hubbard Brook Project. This gives us some idea on the differences in temperature regime. Based on the two years in which he took data on bud burst, there might be from a 13- to a 22-day delay in bud break from the 570-foot elevation to the 2,570-foot elevation, this is across only 3 miles horizontal distance. So, the answer is yes, there are large environmental differences across the elevational gradient.

ANONYMOUS - Do you have environmental data across the entire gradient?

LEDIG - Not across the whole thing, unfortunately, because the Hubbard Brook Project only runs from the 2,100-foot elevation up in watershed 6 and most of my stands are below that watershed.

GABRIEL - Did you notice any evidence of so-called introgression of black maple and sugar maple in your study?

LEDIG - No, I didn't see any indication, but I didn't know too much about black maple until you pointed out some of the characteristics to me last year. I saw only one tree which had hairy leaves. You may have noticed that many leaf parameters vary across this elevational gradient and there are great differences in shape. The ratio of leaf polygon area to leaf area increases as you go up the hill. This means that leaves at higher elevations are more dissected; that is, their lobes are narrower relative to the sinus.

GABRIEL - Maybe you might be able to verify the theory of Dansereau and Desmarais regarding introgression of black and sugar maple by showing that the sinuses gradually get deeper and lobes get more obvious as you go higher up the slope into a cooler climate.

LEDIG - That's a good idea. I've been meaning to go back to their publication and look at their measurements again.

CONNOLA - Do you plan to collect seed and try studies in the nursery?

LEDIG - We are much more interested in collecting clones and we did try to clone each one of these trees. Because of various factors, we only have a couple which have pulled through. Sugar maple produces a tremendous number of seedlings, as you know. In this watershed, the ground is covered with seedlings. They survive a couple of years and then die off. I think that in a closed community, such as this, segregation is going to produce a lot of unfit types. Therefore, I feel that conclusions based on seedlings are not going to be the same as conclusions based on testing the parents as clones. So I prefer to use clones, but there is some seed on some of the trees this year and we might try to collect this.