NATURAL VARIATION IN SPECIFIC GRAVITY IN POPULUS TREMULOIDES

IN NORTHERN NEW YORK

Fredrick A. Valentine[1]

A study of the genetic control of certain important wood characteristics, namely specific gravity, fiber length and chemical composition, has been initiated here at the College of Forestry in Syracuse, New York. The initial phase of this project concerns the natural variation in these wood characteristics.

Trembling aspen, Populus tremuloides Michx was selected for this study for the following reasons: (1) it is one of the most rapidly growing northern tree species and produces flowers at a relatively young age; (2) controlled crosses are easily made in the greenhouse by forcing flowering branches collected during late winter and early spring. The dioecious nature of this species allows large numbers of cross-pollinations to be easily effected; (3) abundant seed is produced approximately two to three weeks following pollination and, after cleaning, can be sown immediately; (4) this species is economically important and has an extensive range in North America.

The purpose of this paper is to present the results from a study of the variation in wood density in trembling aspen in Northern New York. The sample of approximately 200 randomly selected trees should provide a good estimate of the natural variation for this characteristic in the New York population. The preliminary analyses suggest a pattern in this variation which. no doubt, reflects environmental differences as well as genetic differences between what might be considered "breeding groups" in the total population.

MATERIALS AND METHODS

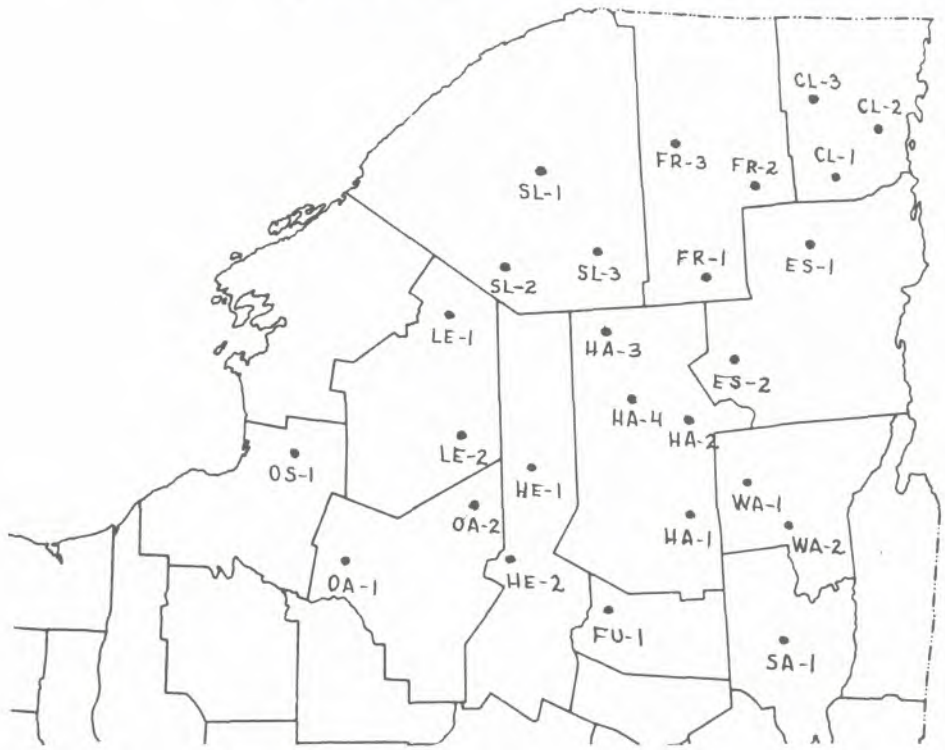
Twenty-six stands of trembling aspen located throughout the Adirondack Mountains and the immediately adjoining areas were selected for this study. The distribution of these stands is shown in the outline map of New York State (figure 1) , In each of these areas random procedures were followed in selecting ten trees, each tree meeting the following requirements: (1) minimum diameter at breast height seven inches; (2) less than a five degree lean in the lower part of the trunk; (3) a minimum distance of twenty- five yards as between any two trees in a stand; and (4) no external evidence of disease. If extensive heart rot was found when taking the wood sample, the tree was rejected.

An increment borer (4 mm diameter) was used to extract a radial core at breast height along the minimum diameter of the tree This diameter was selected to minimize the possibility of tension wood in the sample, An increment core, 11 mm in diameter, was also obtained from one of the ten trees in each stand This tree was selected at random. The cores were placed in tubes containing water (with one-half percent Dowicide G[2] added) and stored under refrigeration to prevent microbial action.

[1] Associate Professor of Forest Botany, State University College of Forestry at Syracuse University, Syracuse, New York.

[2] Sodium pentachlorophenolate and a small percentage of other chlorinated phenolates.

Figure 1.--Distribution of 26 stands of trembling aspen in northern New York State constituting the sample population in this study.



Laboratory Sampling of Increment Cores

Each of the 26 large diameter cores was examined at 20X magnification using a stereoscopic microscope and reflected light. The following information was determined for each core: number of annual rings, ring width, and location and extent of discoloration and/or rot. The ring width was measured using an eyepiece micrometer calibrated at the 20X magnification.

The cores were then sampled in two ways to determine the technique to be followed for the analysis of the 260 small cores. These methods are:

1. Wood samples at fixed radial distances from the pith. Segments 0.5 cm. in length were taken at 2 cm. intervals from a distance 1 cm. from the pith to the cambium, and specific gravity determinations were made.

2. Wood samples for common years of growth. The cores were then "reconstituted" by serially arranging the segments, and samples including three successive annual rings were taken at 10-year intervals, with the outermost sample representing the 1956-58 growth period, the next the 1946-48 growth period, etc., to the pith. The specific gravity of each of these samples was then determined.

The first method was to determine whether specific gravity changes radially with distance from the pith. The variation between wood samples from a given tree would be wholly environmental, that due to differences in growth conditions when the wood was produced and that due to differences in distance from the pith. The importance of the contribution to this variation due to distance from the pith can be determined by employing a covariance analysis with specific gravity (Y) the dependent variable and radial distance from the pith (X), the indepen dent variable By this method. the variation attributable to the linear regression of specific gravity on distance can be separated from the "within tree" or error variation, that is due to uncontrolled causes. The importance of the contribution of this source of variation to the differences between samples can be determined in a comparison with the error variation by using an F test. The variation between trees in this analysis would be due to differences in genotype and environmental factors such as site, exposure and period of growth.

The second method of sampling was employed so that the variations in specific gravity attributable to particular periods of growth would be common to all samples The variations between trees would be due to differences in site and other environmental factors, radial distance from the pith and genotype. The variation between samples within the tree could be adjusted for differences in distance from the pith by a covariance analysis. This would substantially reduce this variation if the relationship between radial position and specific gravity is of major importance. If however, the growth conditions during these particular years are of major importance, this adjustment would not be expected to affect this variation to any extent. The variation due to the linear regression of specific gravity on distance from the pith could be compared with the "error" variation in this analysis by an "F" test also.

Specific Gravity Determinations

Two methods were used to measure the specific gravity of the wood samples, the "volumetric method" and the "maximum moisture method" (Smith, 1954); Standard procedures were followed in determining the three sample weights necessary for the volumetric method.: the wet weight (m_m), the over-dry weight (m_o) and the weight of the sample in water (m_w).

The maximum moisture method is based on three values„ the wet weight of the sample (m_m), the oven-dry weight (m) and the cell wall density (G_{so}), a constant for a particular species (Stamm, 1929). T he formula to be used in this method is as follows:

$$G_f = \frac{1}{\dfrac{m_m - m_o}{m_o} + \dfrac{1}{G_{so}}}$$

The constant, G_{so}, for trembling aspen has been estimated by solving the above formula for G_{so}, and then substituting into the derived formula the wet weight., dry weight and the specific gravity (Gf) obtained by the volumetric method. The mean cell wall density (G_{so}) for wood samples 6 to 6 5 cm, from the pith was found to be 1.527, and for samples 8.5 to 9 cm. from the pith, 1.526. These values are based on 17 and 22 samples, respectively, all discoloration- and rot-free.

The specific gravity values for the 17 samples 6 to 6.5 cm from the pith and the 22 samples 8.5 to 9 cm. from the pith by the maximum moisture method, using these estimates of G_{so}, were compared with the values obtained by the volumetric method. The results in each case were statistically non-significant (for the 6-6.: 5 samples, t = 0: 176, d f = 16, P> 0; 5, and for the 8 5- 9 samples, t = 0.138, d. f. = 21, P> 0 5). It was concluded that the maximum moisture method gave a reliable determination for specific gravity and was adopted for the analysis of the 260 small diameter cores since it is a simpler and more rapid method.

Sampling of Small Diameter Cores

A wood sample 6 to 6.5 cm. from the pith was selected from each of the 260 cores for specific gravity determinations. The selection of a sample at a fixed distance from the pith to represent the tree was based on the results from the covariance analyses indicating that changes in specific gravity across the radius of the stem are closely related to distance from the pith.

Two tests were performed, one comparing samples at fixed distances from the pith in each of nine trees and the other comparing samples representing the same years of growth in nine trees. Six trees were common to the two tests., In the first of these tests, segments of the large diameter core 3,5 to 4 Cm,,, 6 to 6.5 cm., and 8.5 to 9 cm, from the pith were selected from each tree and the volumetric method was employed for specific gravity determinations. The mean specific gravity value and the regression coefficient for each tree are given in table 1. It is apparent that the regression coefficients are, on the whole, comparable, but a test to determine whether these are homogeneous was performed, In this test the deviations from the individual tree regressions are compared with the difference between the deviations from the average regression and the deviations from the tree regressions. The results are not significant (F = 0.2033, d. f. 8 and 9). This supports the hypothesis that the individual regression coefficients are homogeneous. The average regression coefficient, 0.0064, is therefore, the best estimate of the change in specific gravity associated with change in distance from the pith in these samples. The regression equation, expressing this relationship, is Y 0.380 + 0.0064 (X - 6. 25) and indicates that an increase in specific gravity of 0.0064 is associated with each centimeter increase in radial distance from the pith.

The variation in specific gravity within trees was partitioned into two components, that due to linear regression and that due to uncontrolled causes (error). The value for the mean square due to linear regression, 0.0046 with 1 d. f., when compared with the error variance 0.00027 with 17 d. f. by an F test is highly significant (F 17,037), It can be concluded, therefore, that a major portion of the variation in specific gravity across the radius in a tree is attributable to differences in distance from the pith.

The results from the second test using wood samples produced the same three-year periods (1926-1928, 1936-1938, and 1946-1948) in each of nine trees are also given in table 1. It should be noted that the regression coefficients for this group are also comparable. The test for homogeneity of regression coefficients is non-significant for this group also, although the value of F approaches significance at the 5% level (F = 2.9806, d. f. = 8 and 9, P>. 05 but < .10). This could indicate that the relationship between specific gravity and distance is not strictly linear, or that it is not linear throughout the radius of the stem. This latter interpretation would agree with the findings of Brown (1960) which indicate a non-linear relationship from the pith to a distance of approximately 3 cm. from the pith. The separation of the variation attributable to linear regression from the within tree variation resulted in a considerable reduction in the error variance (from 0.0062 to 0.0003). This reduction is significant at the 1% level (F = 20, d. f. = 1 and 17) which lends further support to the conclusion that changes in specific gravity are related to distance from the pith.

A comparison of the two regression coefficients for each of the six trees common to the two analyses shows many differences. These appear to be associated with range in distance of the portion of the core from which the samples were taken and the mean distance of the samples. In five of the six trees, the smaller regression coefficients are associated with the larger mean distance of the samples from the pith.

For tree CL 206, for example, a regression coefficient of 0.0114 for the samples at fixed distances from the pith is associated with a mean distance of 6.25 cm. and is less than the regression coefficient from the second sampling, 0.0148, in which the mean distance of the samples from the pith is 4.56 cm. The exception to this relationship is the comparison for tree CL 303. The apparent inverse relation between the regression coefficient and distance from the pith is what would be expected if changes in specific gravity with distance are not strictly linear, especially for the first few centimeters from the pith.

Table 1.--The relationship between changes in specific gravity (Y) of wood samples and changes in radial distance (X) from the pith at d.b.h. in each of twelve trees.[1]

Tree number	Samples 3.5-4, 6-6.5 8.5-9 cm. from the pith: Mean specific gravity ($\bar{y}$)	Regression Coeff. (b)	Samples of 1926-28, 1936-38, 1946-48 years of growth: Dist. from pith in cm. Mean	Range	Mean specific gravity ($\bar{y}$)	Regression Coeff. (b)
CL 206	0.418	0.0114	4.56	1.30-7.68	0.438	0.0148
CL 303	0.403	0.0108	5.28	2.25-8.00	0.413	0.0068
ES 202	-----	------	6.26	2.69-9.40	0.405	0.0065
HA 303	0.325	0.0030	9.12	6.70-11.42	0.376	0.0010
HA 407	-----	------	6.08	4.03-7.94	0.389	0.0048
HE-210	0.379	0.0048	----	--------	-----	------
LE 104	0.418	0.0176	6.45	5.14-7.81	0.415	0.0062
OA 207	-----	------	6.86	5.36-8.25	0.433	0.0070
SL 106	0.416	0.0036	----	--------	-----	------
SL 304	0.338	0.0050	5.06	2.22-7.62	0.349	0.0149
WA 106	0.373	0.0034	----	--------	-----	------
WA 209	0.354	0.0050	6.86	4.87-9.22	0.362	0.0007
Average	0.380	0.0064	6.28	--------	0.398	0.0074

[1]The regression coefficients and the mean specific gravity values for the nine trees in which samples were taken at fixed radial distances from the pith are given in the left part of the table. The results from the test using wood samples produced during specific years of growth in nine trees are given in the last four columns. Six trees are common to both tests.

The results from the covariance analyses supporting the conclusion that specific gravity changes with an increase in radial distance from the pith at d.b.h. can be summarized as follows: the reduction in the within tree variance due to the removal of the component attributable to the linear regression of specific gravity on distance is considerable. This source of variation is statistically highly significant in each analysis. This relationship within the tree is expressed by the regression coefficient. The test to determine whether the individual tree regression coefficients are homogeneous was statistically non-significant for each analysis. The best estimate of changes in specific gravity with changes in radial distance is, therefore , the average regression coefficient. This value is 0.0064 for the analysis of wood samples at fixed radial distances and 0.0074 for the second analysis using wood samples representing particular years of growth.

The wood sample selected from a radial core of a tree to represent a tree in comparisons with other trees should minimize variations attributable to uncontrolled causes. A sample taken at fixed distances from the pith would best accomplish this purpose.

The choice of the 6 to 6.5 cm.. sample was based on three observations made in the microscopic analysis of the 26 large diameter cores. These are, discoloration and rot were rarely found at this or greater distances from the pith, The specific gravity value for wood samples made up of the outermost annual rings, i.e., sapwood, was often less than that obtained for samples composed of earlier formed growth rings Since a minimum d.b.h. of 7 inches was one of the selection criteria for the sample trees, segments 8. 5 to 9 cm., from the pith would be expected to occur in the sapwood in trees whose d. b. h. approached this minimum. And, if the two radii of this diameter were not equal as is often the case, this segment would not be present. In three of the 26 trees the radius was less than 9 cm.

Each of the 260 small diameter cores was examined at a magnification of 20X. In many cases the pith was missed, but by positioning the core on a graph of concentric circles with many radii and matching the arc of the innermost complete rings with the circles and the wood rays with the radii, a reasonable estimate of the distance to the center of the pith could be made. For each core the number of complete annual rings was determined, the radius of the core was measured, and the mean width of complete annual rings was calculated. The segment of the core 6 to 6.5 cm, from the pith was located and the years of growth of the annual rings wholly or partially in this segment were recorded as well as the ring widths. The sample was than cut from the core and placed in distilled water in preparation for the specific gravity determination.

RESULTS

The mean specific gravity values obtained from the sample trees in each of the 23 areas analyzed to date are presented in table 2. These have been ranked according to value, from highest to lowest. In addition, the altitude, past history and mean ring width for each area are also given in table 2 In a preliminary statistical analysis to determine whether the specific gravity differences between areas are greater than the variation between trees within an area, only 8 of the 10 trees were used. This was necessitated because in two areas, CL-2 and ES-2, two of the samples contained decay in the 6 to 6.5 cm. segment of the core and could not be analyzed. In each of 8 other areas, only 9 samples could be used because of rot and discoloration, Eight trees, therefore, were selected by random procedures from each of the other 21 areas for this analysis. This gave a total of 184 specific gravity determinations in this test.

Table 2.--Summary of area characteristics for the 23 trembling aspen stands[1/]

Sample area	Mean specific gravity	Altitude in feet	Past history of site	Mean ring width in mm.
HE 2	0.432	1240	Abandoned farmland	2.99
LE 1	0.414	950	" "	2.19
SL 1	0.410	840	" "	2.69
OA 1	0.407	500	" "	3.79
LE 2	0.405	1260	" "	2.51
SL 3	0.403	1440	Old burn	2.24
HA 1	0.400	1820	" "	2.01
HA 2	0.399	1690	" "	1.81
CL 3	0.398	1640	Old burn or clear-cut	2.61
CL 1	0.397	1130	Old burn	2.61
FR 3	0.395	1080	Abandoned farmland (?)	2.60
CL 2	0.391	670	" "	3.59
HE 1	0.390	1800	Old burn	2.61
WA 1	0.390	1620	" "	2.22
ES 2	0.386	1770	" "	2.22
FU 1	0.386	1340	Abandoned farmland	2.25
WA 2	0.380	1640	Old burn	2.19
HA 4	0.378	2000	" "	2.28
FR 2	0.375	1690	" "	2.10
SL 2	0.375	1540	" "	2.66
FR 1	0.374	1600	" "	3.18
ES 1	0.369	2100	" "	2.89
HA 3	0.343	1780	" "	2.31

1/Mean specific gravity and ring width are based on 8 trees in each sample area.

The distributions of specific gravity values of the sample trees and the mean specific gravity values of areas approximate normal distributions with a range of 0.293 to 0.471 for the individual tree determinations and 0.343 to 0.432 for the area means.

The results of the analysis of variance of these data are shown in the following tabulation:

Source of variation	D.f.	Sums of squares	Mean square	Value of F
Between areas	22	0.05833	0.00265	2.65**
Within areas (error)	161	0.16070	0.00100	
Total	183	0.21903		

The difference between areas is highly significant (F=65, d.f. = 22, P < .01). The Student-Newman-Keul test was employed to determine which of the area means are statistically different. This test allows each area mean to be compared with each of the other area means. Sixteen of these comparisons proved to be significant at the 5% level. The eleven highest area means, 0.395 and higher, differ from that of area HA-3 which has the lowest mean specific gravity, 0<.343.

The other five significant comparisons involve area HE-2, which has the highest mean (0. 432), and the areas whose means are 0. 378 or lower (of course excluding HA-3 as this comparison was included among the other eleven).

The intraclass correlation coefficient, r_I , was determined to estimate the relative importance of the variation among the trees within areas due to genetic and/or environmental factors. This statistic compares the variance among the area means (σ^2_A) with the variance of randomly selected trees from the sample population (σ^2 +
The intraclass correlation coefficient of 0. 17 suggests that a relatively large amount of the variation is present among trees in the sample areas.

DISCUSSION

An attempt has been made to determine whether the variation in specific gravity between areas can be related to geographic location, altitude above sea level, rate of growth, and past history of the area. The five areas with the highest mean specific gravity values, 0.405 and larger, occur along the western periphery of the Adirondack Mountains (see figure 1). These areas are at an altitude of 1260 ft. or less and occur on abandoned farmlands. In contrast, the seven areas with the lowest means, 0.380 and lower, occur within the Adirondack Preserve with five of the areas in the general vicinity of the "high peaks" region. These seven are at elevations of 1540 feet or higher and occur in areas of old burns. A slight positive correlation between specific gravity and mean ring width appears to be present in a scattergram constructed from these measures. This has not been determined statistically, however, so that it is not known whether a real relationship exists between specific gravity and rate of growth in this population.

Until a complete statistical analysis is made, conclusions on the relationship between each of these factors and specific gravity are hazardous. A multiple correlation analysis is planned when the specific gravity determinations of the remaining samples are completed.

SUMMARY

This study demonstrates that a relatively large variation in specific gravity is present in the trembling aspen population in northern New York. The component of this total phenotypic variation attributable to differences between sample areas is statistically highly significant. This result indicates that real differences do exist within this population. A relatively low intraclass correlation suggests that a fairly large amount of the variation occurs within sample areas.

LITERATURE CITED

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