

PRELIMINARY ESTIMATES OF THE GENETIC STRUCTURE OF TWO SYMPATRIC POPULATIONS OF BIRCHES AS DETERMINED BY RANDOM EFFECTS AND NATURAL SELECTION

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In applying the principles of quantitative genetics to natural populations of forest trees, some difficulties are encountered in tree breeding experiments because of the complicated structure of these populations. But, fortunately, there are some regularities in the pattern of genetic variation on the population level giving a base for the planning of experiments, the goal of which is to give an integrated picture of the genetic variance over the whole population. In order to establish an experiment of this type, we have to apply a particular sampling design, or better but much more expensive, mating design. The two experiments described below are based on samples of single tree progenies.

Two Japanese birch species were chosen as the test material: Betula japonica, a diploid species, which is probably the Japanese race of the European species B verrucosa, and B. maximowicziana, another diploid species sympatric with B japonica throughout most of its range. Each of these species belong to a separate section of the genus Betula, and their ecology is quite different, B. japonica is an opportunistic species, having an irregular reproduction and giving, therefore, the better chance for random differentiation on the population level. B. maximowicziana on the other hand is almost an equilibrium species, having a more regular reproduction (both terms are explained by McArthur in American Naturalist, 1960). One of the hypotheses to be tested in the two experiments is, that the degree of "subdivision" as a consequence of chance events should be higher in B. japonica. The populations of both species can be assumed to be natural populations

Open-pollinated seed were collected from trees in each of several subpopulations within different growth regions of the species by the Japanese Institute of Forestry at Meguro. This type of sampling design can be established easily and accepting some assumptions (to be discussed later), the data can be analyzed using a nested analysis of variance; the variance components of which have an interpretation in terms of quantitative genetics. If there is no indication of clinal variation within the growth regions, as is the case in our experiments, the above analysis of variance gives all information about subdivision within the growth regions .

The model underlying the analysis of variance can be written as follows:

$$y_{ijkl} = m + W_i + s_{ij} + b_{ijk} + e_{ijkl}$$

where y_{ijkl} stands for the plot mean of a particular character. m for the consistent mean of the experiment, w_i for the average effect of the growth region i , s_{ij} for the effect of the j -th subpopulation within growth region i , b_{ijk} for the effect of mother tree k in subpopulation s_{ij} , and e_{ijkl} for an error term, resulting from the field experiment (replicate \times progeny interaction of a randomized block design, the block effect is neglected for simplicity).

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The components of variance, expressed as a percent of the total genetic variance (sum of the three components for growth regions, subpopulations and half-sib families) in the two nursery experiments, are:

	<u>B. japonica</u>	<u>B. maximowicziana</u>
	<u>Percent</u>	<u>Percent</u>
$\hat{\sigma}_w^2$	78.5	59.8
$\hat{\sigma}_s^2$	11.3	12.0
$\hat{\sigma}_b^2$	10.2	28.4
	<u>100.0</u>	<u>100.2</u>

The species range of B. maximowicziana is the smaller of the two, and therefore, the smaller amount of variance among growth regions is not necessarily an indication of a lower intensity of natural selection for that species.

In both cases, the differences among growth regions in cessation of height growth is the most important source of variation. However, this is not so for all characters. If a character has no obvious selective advantage, or does not vary clinally, that component of variance should become considerably smaller. The other two components may show an amount of genetic variation comparable to the data for cessation of height growth.

As mentioned above, there is no clinal component detectable within growth regions. But the component of variance for among growth regions is to a great extent explained by two clines, as will be discussed later. This fact also emphasizes the necessity for wide distribution of populations for studies of clinal variation. On the other side, it stresses the concept of the "growth region", used by German foresters for about 40 years, which seems to be much more useful for practical purposes, than the "continuous concept" for seed certification and other regulations.

Before going further in the analysis of the data, some assumptions must be made in order to obtain a base for this analysis:

1. The genetic variance within any growth region is entirely additive.
2. The subpopulations in the same growth region are randomly derived from a common random mating population by means of inbreeding.
3. The degree of inbreeding is the same for all of the subpopulations within the same growth region,
4. The genetic variance and the degree of inbreeding is the same in all of the growth regions relative to the base population of any growth region.
5. There is random mating within subpopulations.

On the basis of these five assumptions, the expectations of the variance components for subpopulations (V_s) and half-sib families (V_b) can be given in terms of the additive genetic variance (the derivation is given in several textbooks):

$$E(V_s) = 2 F \sigma_A^2 \quad E(V_b) = (1-F) \sigma_A^2 / 4$$

From the above equations, it is possible to obtain estimates of σ^2 corrected for inbreeding, and the coefficient of inbreeding can also be used in this case as a measure of the degree of subdivision.

Using the analysis of variance given above, we obtain, for B. japonica and B. maximowicziana respectively

	$2\hat{F}\sigma_A^2$	0.1562	0.0441
	$\frac{1-\hat{F}}{4}\sigma_A^2$	0.1414	0.1065
and			
	$\hat{\sigma}_A^2$	0.6437	0.4481
	\hat{F}	0.121	0.049

It is of interest to note, that the estimates of the genetic variance within subpopulations uncorrected for inbreeding are 0.5656 and 0.4260 for the two species, which is too low by about 12 and 5 percent.

In progeny tests of trees selected over the entire growth region, as is done in most selection programs, the existing subdivision may cause considerable bias in the estimates of genetic variances, and consequently, in estimates of heritability. The amount of that bias depends on the distribution of the selected trees over different subpopulations and on the mating design chosen for the test. The most extreme bias is expected to occur in estimates based on open pollinated progeny if seed is collected from only one tree in each subpopulation, as is done in many cases. In such an experiment, the expectation of the genetic variance is not:

$$E(V_b) = \frac{1}{4}\sigma_A^2 \quad \text{but} \quad E(V_b) = (1-F)\sigma_A^2/4 + 2F\sigma_A^2$$

Using the data given above and assuming such an experiment, the bias resulting from subdivision would lead to an over-estimation of σ^2 as high as 85 percent for B. japonica and 35 percent for B. maximowicziana.

It should be possible to obtain estimates of the degree of subdivision from nursery experiments using the same or a similar technique as that described above. Clearly the degree of inbreeding may not be the same for all of the characters the forest tree breeder is interested in, but these estimates at least give some idea of the degree of subdivision within a population, and they might be a better approach than the estimates of subdivision obtained by experiments on pollen distribution.

Of the five assumptions listed above, some are probably not fulfilled. For example, a single or a few trees may be the predominant pollen sources. Also the assumption of completely additive genetic variance within any growth region may not hold. The amount of bias resulting from these deviations can be estimated only if the deviations are known. But it can be shown that only large deviations result in considerable bias.

Now, we may consider the effects of natural selection upon the two birch populations. For that purpose the regression of cessation of height growth to geographical latitude (X_1 , using degrees of latitude as unit) and altitude above sea level (X_2 , given in meters) is calculated for both species (c. f. Kriebel's 1957 experiments with sugar maple):

$$\begin{aligned} \underline{B. japonica}: & \quad Y = 21.6664 - 0.4227X_1 - 0.0009X_2 \\ \underline{B. maximowiczii}: & \quad Y = 22.2729 - 0.2991X_1 - 0.0010X_2 \end{aligned}$$

Here, it is necessary to introduce another assumption:

6. The population over the whole area we have sampled should have reached an equilibrium with the simultaneous forces of selection and migration.

Instead of assuming that the base population covers one growth region, a gradually changing population is introduced here with gradually changing gene frequencies parallel to the cline or clines. This assumption, along with the probability of gene exchange among subpopulations (migration), includes the assumption of selection, which has a stabilizing effect in that it acts in the conservation of the gradually changing composition (figure 1). The term "stabilizing selection" here is used in the sense given to it by K. Mather, not that of Schmalhausen. The latter defines it in a sense Waddington has given to his "canalizing selection".

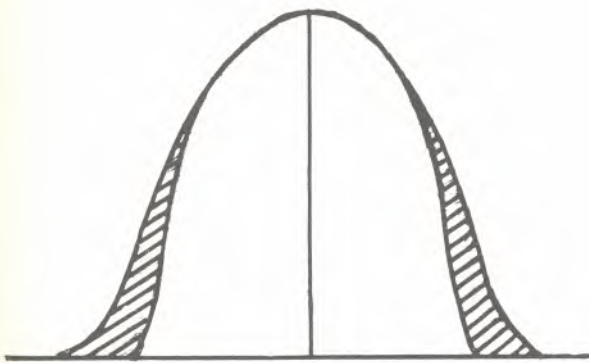


Figure 1. -- Stabilizing selection.
The individuals in the shaded areas will be eliminated.

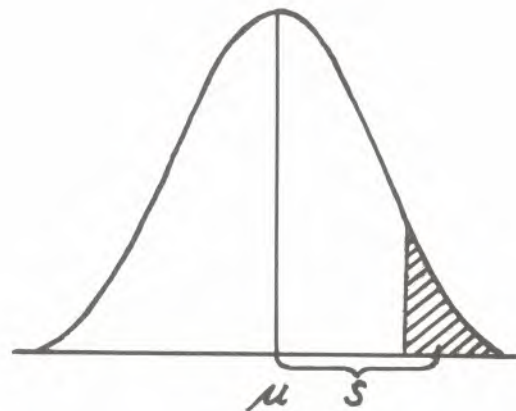


Figure 2. -- Model for selection.
S = selection differential
R = selection response = $i h \sigma_A$
 $i = S/\sigma_p$ $h^2 =$ heritability

By using the means of subpopulations, the sum of squares for "among subpopulations" can be split up into two components (assuming here only one cline that is linear in effect and neglecting the growth regions):

$$(Y - \bar{Y})^2 = (Y - \bar{Y}_x)^2 + b \text{Cov } X, Y$$

The expectation for the term $(Y - \bar{Y}_x)^2$ would be about the same as the sum of squares for "among subpopulations within growth regions" in the analysis of variance given above. The second term, $b \text{Cov } X, Y$, measures the proportion eliminated by the regression of the character Y on the "ecological variate" X.

Estimates of the effects or the intensity of natural selection can now be obtained using the conventional model for selection experiments and the cline (figure 2). Since the rate of migration and the effect of stabilizing selection per generation is unknown, the information obtained from the selection model is limited.

However, it seems possible and useful, for purposes of comparison, to obtain an estimate of the selection intensity necessary to change one of the populations, Y to another Y_{X+1} , in only one generation of selection. If the base populations, assumed to exist in the environments X and X+1, are considered, the difference between the two populations is the selection response to be achieved. R (c.f. figure 2) in this case is consequently an appropriate measure of the effectiveness of natural selection,

For the character "cessation of height growth" the following estimates of $6A$ were obtained:

$$B. \textit{japonica} - 0.802; B. \textit{maximowicziana} - 0.669$$

Since the heritability, as estimated from the experimental data, cannot be taken as an estimation of the heritability in the original population, we may only take into account the genetic variance (putting $h^2 = 1$).

By using only the regression of our character on latitude and in determining the difference between two populations one degree of latitude apart, R takes the values 0, 4227 and 0.2991 for the two species, as given by the regressions for the "cline". Now we obtain $i = 0.54$ for B. japonica and $i = 0.45$ for B. maximowicziana as estimates of the intensity of natural selection. The two estimates are not too much different.

Since $i = z/p$ (z = height of the ordinate at the point of truncation, p = percent of individuals beyond that point), we are in a position to determine the proportion of individuals which have to be selected in order to change one of the populations into the other in respect to the character in question by a single step of selection. These proportions are:

<u>B. japonica</u>	<u>B. maximowicziana</u>
<u>Percent</u>	<u>Percent</u>
66	73 (from X to X+1)
34	44 (" " " X+2)
13	22 (" " " X=3)

Apparently the difference between the two species is not large as far as intensity of selection is concerned. There was considerable difference, however, in the degree of subdivision, as expected for some reasons.

If only the differences between populations parallel to the cline have to be shown, Haldane's suggestion for evaluating rates of evolution can be followed. His "rate of evolution" gives the difference between the population means in terms of standard deviations. Here the genetic variance can be used. The results naturally are the same as given above in the analogous terms i . This is shown in figure 3.

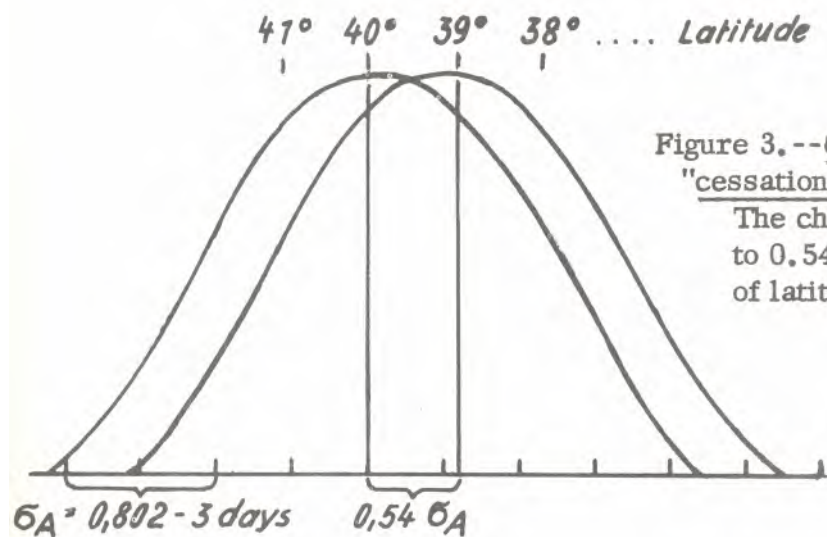


Figure 3. -- Clinal variation of the character "cessation of height growth" in *B. japonica*.
The change in population mean amounts to $0,54 \sigma_A$ for a distance of 1 degree of latitude between the two populations.

If we accept the estimate of the intensity of natural selection given above as somewhat realistic, this intensity seems to be not too high as long as a single character is concerned. But natural selection has to be understood as index selection. Then, assuming only a few characters each to be selected for with the same low intensity, the intensity of index selection apparently is very high. This would seem to point to the fact that natural selection indeed is the major factor of micro-evolution even in restricted areas, as the European "school" of forest genetics has considered in the past, but that genetic drift and related causes are also an important factor as far as single traits are concerned. The second view, if the writer is right, is a relatively new one in forest genetics and given for the first time by J. W. Wright.

This explanation of differences between "stands" within a limited area and in similar environments by accidents of sampling rather than by natural selection is of some importance for the concepts of tree breeders. But, at the present time, our view is a highly speculative one. More experiments are needed before the complicated structure of the forest tree populations is understood sufficiently.

ACKNOWLEDGMENTS

The experiments referred to in this paper have been established with the aid of the Deutsche Forschungsgemeinschaft. Areas for field experiments are submitted by the Niedersaechsische Landesforstverwaltung. The writer is further indebted to Dr. S. Wright and Drs. Kojima, Perry, and Zobel from the North Carolina State College at Raleigh for suggestions and discussions.

DISCUSSION

FUNK - Dr. Jaynes, am I correct in assuming that there was no treatment or you didn't mutilate the top of the stock plant during the marching or before the rooted in-arch was severed from the plant?

JAYNES - Only a little before I grafted. I did cut back the stock plants severely once the bud on the scion had broken.

FUNK - That was the following spring?

JAYNES - No, the same summer,. These are grafted in May, and the buds break dormancy quite rapidly. So it would be in June and July that I pruned back the stocks. This year I am also trying to girdle the stock just below the area where the scions are marched into it; that is, girdling the root of the stock at about ground level. I don't know what effect this will have.

FUNK - Perhaps forcing more auxins down into the scion?

JAYNES - Yes, The idea being that by girdling the roots of the stock perhaps this will encourage more auxin transport to the base of the scions and thus more rapid root formation,

FUNK - Had you done any fertilization of the stock plant?

JAYNES - No, this was in a field that several years ago was planted to corn and had been fertilized at that time, As you could see in the pictures they were mulched lightly with wood chips and there was no fertilization. I had also tried using indolebutyric acid on a few of these, but no apparent differences in rooting resulted.

WRIGHT - I have a question for Dr. Stern. Am I correct in assuming that your data are based on tests of seedling progenies of these birches in the nursery?

STERN - That's right.

WRIGHT - Do you think it was necessary to do such progeny testing to determine how selection will affect our population ?

STERN - I think it is quite useful in order to design our selection programs, to get a picture of the genetic structure of populations, and to get better estimates of heritabilities.

STAIRS - Dr. Valentine, you mentioned earlier that you plan to do heritability studies later on, I wonder if you have given any thought as to what you are going to consider your population since northern New York isn't necessarily a panmictic situation?

VALENTINE - For the initial experiments, to obtain estimates of heritability in this program, this population will be sampled. These estimates, as is the case for any heritability estimates, will be applicable only to the sample population, i.e. Populus tremuloides in the Adirondack area of northern New York State, and may not be representative for the species throughout its range. The project will, however, contribute useful information on the genetic control of these characteristics in this species. As in many studies the size of the sample population has been restricted by time and cost factors.

STAIRS - But you are considering the whole northern New York as a single population?

VALENTINE - Yes, this general area. It may be composed of subpopulations because certainly the occurrence of aspen is directly related to burns or abandoned areas. I suspect that the relationships within a given stand are much greater than between stands.

STAIRS - There is a publication on heritability in Populus tremuloides by van Buijtenen et al. in Tappi 1959. They reported a heritability figure of 17 percent for wood specific gravity. We did a limited study of this sort at Yale and obtained similar results for the same species. Given that both of these studies were conducted with clonal material, and thus differ from your approach, do such results discourage you in any way - especially in terms of obtaining more realistic and higher values ?

VALENTINE - No, I think the only way we will get a good estimate of heritability, as Dr. Stern said, is by progeny testing to obtain either parent-progeny correlations or sib correlations. One can get some notion of the heritability as Dr. van Buijtenen did in his Appleton work, but I think a study of this type has to be made to obtain a reliable estimate, Reliable estimates of heritability of various wood properties are necessary for the construction of a selection index in which wood characteristics are considered.

STAIRS - I suppose that in this instance you would use full-sib correlations rather than parent- progeny because of the possible difference in juvenile and mature woods.

¹ Editor's note The following comment was appended by Dr Valentine to his edited discussion remarks: It is assumed that the population sampled represents a part of a continuous population, with the mating individuals restricted to a limited distance or "neighborhood," so that individuals greatly separated by distance have little or no chance to mate This population model was referred to as "isolation by distance" by Wright (1943, after C. C. Li, 1955, Population Genetics, University of Chicago Press), According to this model, as neighborhood size increases, the population approaches that of a completely random mating population. The northern New York aspen population is probably composed of intermediate- sized neighborhoods, each with at least 100 individuals, Since the total, population is very large and since migration between adjoining neighborhoods would be expected to be very frequent adjacent areas should be quite similar and the heterogeneity would be gradual. The observed differences in area means support this interpretation."

VALENTINE - Parent-progeny correlations would be feasible if juvenile and mature wood characteristics exhibit a high correlation. Zobel has published results for southern pines which do show a high positive correlation. A project to determine these relationships in aspen is underway here.

GENYS - Dr, Valentine, you mentioned that you plan to make correlation analyses of specific wood gravity with soil qualities. I think that one of the best expressions of soil qualities could be made by determining the chemical composition of representative soil samples. The specific gravity of wood could be correlated with the concentration values of particular nutrients. I wonder if you have plans along this line in your study?

VALENTINE - This type of analysis is not planned in this study for several reasons.

First, the field selection phase is primarily to obtain a sample group for heritability determinations. Though sources of variation in the population are of interest, they are not critical for this experiment. The progeny from controlled crosses will be planted in field trials under relatively uniform conditions and it will be from these plantings that the heritabilities will be estimated.

It would be of interest, however, to include a detailed soil analysis so that these could be related to wood properties. Because of site variations within the areas sampled, a soil analysis would have to be made for each tree and this would be a rather tremendous task. I hope to devise some kind of index that would reflect soil types, drainage, etc., and use this to relate this aspect of the environment to the wood properties.

HOOKER - Dr. Valentine, you mentioned in your procedure that you did select initially 10 trees from each area, and then in your analysis I noticed that you used only 8. How did you decide to eliminate the other two trees ?

VALENTINE - Small areas of rot or discoloration are frequently present in aspen, even in trees otherwise free of rot. A number of sample trees had to be eliminated because this rot occurred in the particular part of the core to be analyzed. In two cases a portion of the core was lost so that these could not be used. The maximum number of usable samples in all 23 areas was eight. In those cases where there were 9 or 10 usable samples for an area, the extra one or two were eliminated, using a table of random numbers to obtain a non-biased uniform sample number of eight per area. This was done for ease in the statistical analysis of the data.