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PROGENY TESTS IN A PINUS SILVESTRIS (L.) SEED ORCHARD IN FINLAND

P. M. A. TIGERSTEDT

HELSINKI 1969

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PREFACE

Genetic tests of forest trees are invariably both time-consuming and long-termed. Almost two years elapse before the necessary control pollinated seed is at your disposal and elaborate nursery facilities are imperative for maximum reliability. Another couple of years pass by before the first estimates of genetic variability are at hand.

I am greatly indebted to the CENTRAL BOARD OF FORESTRY, TAPIO, who kindly offered me assistance in raising progeny at their plant nursery and also put their Scots pine seed orchard at my disposal.

Likewise, I want to express my gratitude to THE FOUNDATION OF RESEARCH OF NATURAL RESOURCES IN FINLAND for their valuable economic support, which enabled me to carry out the necessary data processing.

Professor RISTO SARVAS, Head of the Department of Silviculture at the Finnish Forest Research Institute has kindly given me advice on topics concerning early plant development and pollination.

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I also wish to extend my thanks to a number of unnamed persons who have helped me during this investigation.

Helsinki, September 1969

P. M. A. Tigerstedt

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1. BASIC GENETICS OF QUANTITATIVE CHARACTERS

It is well known that optimization of breeding can only follow from a clear understanding of population and quantitative genetics. The genetics of quantitative characters is of course only a part of the knowledge necessary, other investigations must scrutinize population delimitation and problems of population biology; strategy of species.

We are working with a living control system which can only be optimized to meet our needs if we know the components of this system; the strategy of its genes, its individuals and its populations.

The phenotype comprises two main factors: the genotype and the environmental influence on the genotype. In provenance research and in testing certified seedlots, field trials primarily aim at a testing technique that makes it possible to distinguish between these two components of variance.

The quantitative genetics of characters can only be analyzed by meticulous methods were the genotype is further fractioned into different components of gene action. This is where different cross desings must be applied.

Statistically the phenotype is expressed by the total variance of the individuals (V_p). This total variance comprises the genotypic variance (V_g), the environmental variance (V_e), and the interaction of the genotype and environment (V_{ge}). The interaction is often non-linear, i.e. a change in the environment affects different genotypes in different ways (Comstock and Moll 1963). In all:

$$V_p = V_g + V_e + V_{ge}$$

The interactions can often be very disturbing factors in field trials: as soon as there are environmental differences when a trial has been replicated, these interactions appear as noticeable sources of error. The field trials have to be planned so that the V_{ge} -sources of variance can be distinguished. This is possible if the trials are repeated with respect to place, and if possible, to time as well. The size of the field trial is on the other hand dependent of the aim of the investigation; what traits are under investigation? are we dealing with a provenance test or with a test of genetic variability? In this connection it is important to notice that very small trial plots (1—10 individuals) usually increase the proportionate part of the V_{ge}. This is one of the most difficult questions pertaining to progeny trials with forest trees. Provenance trials and corresponding breed trials that primarily aim at comparing different breeds or origins for parctical purposes ought to be established on sufficiently big plots so that the inner interaction of the plots resembles that of a normal cultivation. Genetic trials, again, are established for quite a different purpose. Here the aim is to divide the genetic variance into its components that depend on different kinds of gene action. In this case we do not aim at any result applicable to practice, but to a trial that gives us valuable information of the genetic structure of different traits. Our largest unit in this trial is a delimited population whereas a provenience trial may comprise a whole species. It must be expected that V_{ge} interactions are considerably smaller in trials within a population than they are if the trials include several different provenances or breeds. For this reason the number of individuals in the plots need not be high if genetic variance within a population is concerned.

Åll field trials naturally require that the environmental influence is distinguishable from the genetic influence. There are many different designs that fulfil this requirement and the most simple is used in this investigation: the randomized block design.

Proper breeding aims at actively improving the stock or provenance. The best breeding method can be applied only when the genetic structure of the population (or species) under work is known. This structure is best expressed as genetic variation that theoretically divides in the following way (Kempthorne 1957, Kidwell and Kempthorne 1966 etc.):

$$V_g = V_a + V_d + V_{aa} + V_{ad} + V_{dd} + + V_{aaa} + V_{aad} + \ldots \text{etc.}$$

where

V_a = component of variance that depends on the mean influence of the genes; *additive gene effects*,

V_d = component of variance that depends on the interaction of allelic genes; *dominant gene effects*,

V_{aa}, V_{aaa} etc. = components of variance that depend on the interaction of genes with additive influence belonging to two or several non-alleles; *epistatic gene effects*,

V_{ad}, V_{aad} etc. = components of variance that depend on the interaction of the additive and dominant gene effect; *epistatic gene effects*.

Of these components V_a and V_d are the most important to breeding. Epistatic gene effects may cause a lot of variability particularly in respect to fitness traits (Tigerstedt 1969), but the utilization of epistasis in breeding is almost impossible except in cases where genotypes can be vegetatively propagated. Epistatic effects are very difficult to distinguish statistically and they are usually included in the main genetic components or in the error variance (see e.g. Kempthorne 1957 p. 423).

In practical breeding based on mass selection the additive gene effect is considered the most important. This component enables the making of evaluations of the genetic gain $\triangle G$ which may be expressed as the per cent increase of the yield. The additive variance is usually expressed as the quotient V_a/V_p which is the expression for »heritability in the narrow sense» (Lush 1945). Thus the genetic and the environmental variance influence the quotient of heritability. It is apparent that among these components the genetic part is constant, but the environmental variance changes as conditions change. It is particularly difficult to define the influence this environmental variability has on the interaction V_{ge}. In an ideal field trial, the environment is not variable and heritability expresses the ratio V_a/V_g. This quotient would enable one to express the quantity of genetic gain depending on mass selection without an error component. However, a situation like that is quite unrealistic since there are no uniform growth sites in nature and the heritability will in trials have relative values which change from one trial to another. Heritability decreases if the environment where the field tests take place is more heterogeneous — this is a problem of the heritability concept which was initially defined for animal breeding purposes!

When deciding on a certain breeding design it is important to lay particular stress on the structure of the genetic variance. If the trait in question has a high degree of additive genetic variance, mass selection is probably the most effective breeding design, and if the main part of the genetic variance is a dominant variance, it pays to use individual cross breeding. The best genetic gain can often be obtained by combining both breeding designs (reciprocal recurrent selection), but such designs require much time and are perhaps best applicable to animal breeding. Naturally there are still a number of practical matters owing to the biology of flowering which have to be kept in mind when choosing a design. Only a breeding based on mass selection is used as regards *Pinus silvestris*, for instance, since artificial crossing is quite expensive and with respect to practical production of seeds, simply impossible. When improving *Alnus* or *Betula*, again, it is quite possible to use controlled cross breeding where individual crosses are particularly made use of.

2. THE STUDY OF GENETIC STRUCTURE

The genetic variance in a population is further divided into components that depend on the actions of the genes. This division is carried out by a special analysis based on the covariance of half-sib and full-sib families. Genetically this is a degree of similarity owing to relatedness, statistically it is a degree of intra-class correlation. Analogously it is possible to use regressions between parents and off-spring for this purpose but this is probably practicable only in animal breeding or when breeding plants with short generations. To deduce mathematically the components of variance (σ^2_a and σ^2_d etc.) obtained in this way is tedious and will not be dealt with in this context. It is to be observed, however, that the symbols of variance V and σ^2 are not identical. As described above the phenotypic variance can be divided into components that depend on different factors. This division is theoretical and the components are symbolized by V. The phenotypic variance can also be experimentally divided into components. It is possible to isolate these components by observing the similarities of relatives in full- or half-sib families, for instance. They are estimated in the very phenotypes and differ in this respect from the theoretic or causal components. This experimental division of variance is based on Gauss normal distribution and symbolized by σ^2.

The analyses require that the individuals belonging to the crosses fulfil several conditions, all of which in some way secure the normal distribution of the traits. The most important of these are presented as follows (COCKERHAM 1963): 1) individuals incorporated in the cross design form a random sample of the tested population. 2) the genetics of traits is based on a normal diploid meiotic division, 3) similarity of relatives must not be due to the environmental correlation, or in other words, no reciprocal cross differences must appear, 4) there is no inbreeding depression (or the coefficient of inbreeding has to be known), 5) there is no linkage of genes. It is apparent that a series of crosses carried out in seed orchards, for instance, does not fulfil these conditions in every aspect. We should really know the population genetics of the species before we could begin to analyse in this way. We should know the physical quantities connected with pollination, such as flying distance and the distribution of pollen around the pollen-source. The dispersion capacity of the seed ought to be known, too. From a theoretical aspect, again, it ought to be stressed that a linkage of genes always appears merely owing to the linear structure of chromosomes. One has to assume, however, that such a linkage is balanced and it then has no effect on the mathematical interpretation. A different linkage appears in polymorphic populations, however, and this can lead to an erroneous mathematical interpretation. The covariance of related offspring comprises genetic components according to the following table:

	V_a	V_d	V_{aa}	V_{ad}	V_{dd}
Half-sib, $Cov_{(HS)}$	1/4		1/16		
Full-sib, $Cov_{(FS)}$	1/2	1/4	1/4	1/8	1/16

Interactions have little influence on the covariance and they are usually included in the main components V_a and V_d. Accordingly the additive genetic variance (V_a) will also include 1/4 part of the V_{aa} interaction and, analogously, the dominant variance will include some epistatic interaction. For this reason the estimated values for the main components will perhaps be somewhat too high.

21. Polycross analysis

For practical reasons Scots pine breeding in Finland is entirely based on mass selection. Selection of superior trees is carried out in the forest. These trees represent favourable phenotypes with respect to growth and quality. They are propagated vegetatively by the help of grafting. The grafts are planted in seed orchards and nature is left to take care of the crossing between different clones. The method is very plain and simple. However, it seems to be the only large scale breeding method applicable to Scots pine since controlled cross breeding for practical purpose is too expensive.

Since we are here dealing with a breeding method based on mass selection our main interest lies with the additive genetic variance. All seeds collected from the same tree must naturally be related; they must be half-sibs or, if pollinated only by one or a few fathers, comprise groups of full-sib families. It will be impossible to judge the exact composition of the pollen source and furthermore its character may change from year to year depending on weather condition and profusion of flowering.

In the polycross we wish to obtain a progeny which consists of half-sib groups. The best way of assuring this is to remove the male flowers from the seed orchard in question before flowering begins. The pollination must now take place with the »general» pollen of that area. Its consitution is of course unknown but one may assume that it consists of a pollen mixture of surrounding pine forests. The reliability of the analysis essentially depends on how intense the anthesis of local Scots pine forests has been. The more richly the pines have flowered, the more completely has the pollen from different trees and stands been mixed. We must expect, however, that different years give different results; poor flowering should envetually give a high percentage of full-sib progeny, less polyzygotic proembryos per ovule and therefore less opportunity for genetic competition on the embryonic stage. (SARVAS 1962). It is therefore preferable to use seed harvests of good flowering-years for the analysis.

In a seed orchard in south Finland (Oitti) the seed harvest of the 1964 flowering was gathered according to clones. There were 25 clones in the orchard and a sufficient amount of seeds were gathered from 23 clones. The cones were kiln-dried and the seedwings were removed by scrubbing the seeds in cloth-bags. Seed weight was put at the level of 5.9 grams by a countercurrent sorter. All empty seeds were removed in this way. Investigations have showed that the weight of the seed influences the initial development of the seedlings until their 4th or 5th year (HADDERS 1967, SQUILLACE *et al.* 1967). This variability of seed weight may to some extent be a genotypic feature of the embryo and in this case adjustments of seed weight may cause certain selection of genotypes. Conifer seed endosperm being haploid however, it obviously is a maternal effect to a very great extent and so should be eliminated for clear genetic analysis. The seeds were sowed using a special sowing form in plastic basins filled with peat. This sowing form ensured that the seeds were sowed at an even spacing of 4 × 4 cm. A basin contained 104 seeds maximally and this unit constituted one plot. A randomized block desingn was used and each lot was replicated 3 times. A plot gave on the average about 80 seedlings. All trial seedlings ($3 \times 23 \times 80 = 5\,520$) were measured in centimeter height classes after the first and the second growth periods in 1967 and 1968. The data were transferred to punch-cards and the analysis was programmed and operated on an electronic computer at Helsinki University. During the first year of growth the plants grew in a plastic greenhouse and were artificially irrigated and fertilized to obtain a maximal growth. During the second growth period the plants still grew in the same basins but they were now exposed to normal weather conditions. Plants averaged about 10 cm after the first growth period and about 20 cm after two periods. In the spring of 1969 the trial was transferred to a field nearby and planted in a randomized block design. No measuremets are planned within the next 5 years in the field because of planting shocks arising at transplantation (TIGERSTEDT 1966).

The polycross analysis is presented below.

1) analysis of plot means

Source of variation	*d.f.*	*Sum of squares*	*Mean squares*
Replicates	$r-1$	$\sum_k \frac{Z^2_{.k}}{g} - \frac{Z^2_{..}}{rg}$	
Between mothers	$g-1$	$\sum_i \frac{Z^2_{i.}}{r} - \frac{Z^2_{..}}{rg}$	S
Error	$(r-1)(g-1)$	Difference	I
Total variation	$rg-1$	$\sum_{ik} z^2_{ik} - \frac{Z^2_{..}}{rg}$	

2) analysis of individual plant data

Source of variation	*d.f.*	*Sum of squares*	*Mean squares*
Between plots	gr − 1	$\sum_{ik} \frac{Y^2_{ik.}}{n_{ik}} - \frac{Y^2_{...}}{N..}$	
Within plots	N.. − gr	Difference	E
Total variation	N.. − 1	$\sum_{ikl} y^2_{ikl} - \frac{Y^2_{...}}{N..}$	

3) interpretation

Parameter	*Estimate*	*Genetic component*
$Cov_{(HS)}$	(S − I)/r	1/4 V_a
σ^2_e	$I - n_h E$	
$\sigma^2_f + \sigma^2_g$	$E + Cov_{(HS)}$	

where replicates = r, mothers = g, yikl = the height of the l : th plant in the mother group i and replicate k, z_{ik} = mean value of plot, n_{ik} = number of plants in the plot, n_h = reciprocal of the harmonic mean of the plants per plot.

The number of plants per plot naturally varies to some extent, since there are germinative differences in the seed quantities. This variation would cause great difficulties if the estimations were made according to an orthogonal analysis. For this reason it is best to have two different analyses of variation that are not directly connected with each other (KEMPTHORNE 1957 p. 459). These are: 1) a genetic analyses based on the means of plots where the possible differences of exactness of the means are not noted, 2) an analysis within plots and between plots.

In interpreting the analysis of variance the expected mean squares (or sums of squares) have to be deduced according to the genetic model. This fact is not dealt with in this context, but the genetic implication of the individuals of the trial ought to be kept in mind: 1) all individuals of the same plot are presumably half-sibs, 2) individuals from the same mother in different replicates are half-sibs.

The error or environmental variance is thought to consist of two factors: 1) an individual factor (f) that is independent of other individuals and the expected value of which is zero and variance is σ^2_f, 2) a common factor (e) which is the same for all individuals of the same plot but independent of other plots, the expected value of which is zero and the variance σ^2_e. An apparent consequence of this is that the expected mean square within plots is $\sigma^2_f + (\sigma^2_g - Cov_{(HS)})$.

It is possible to deduce the expected mean squares of the plot means in an analogous way.

In the following the results of the first and second year measurements are presented and the mean squares are genetically interpreted.

Source of variation	*d.f.*	*M.S. 1 year*	*M.S. 2 years*	*Symbol*
Replicates	2	2.89	10.68	
Between mothers	22	4.99*	24.78*	S
Error	44	2.69	14.18	I
Within plots (n_h = 0.01)		3.93	17.48	E
interpretation:		1 year	2 years	
$Cov_{(HS)}$		0,76	3.53	
σ^2_a		3.06	14.13	
σ^2_e		2.64	13.93	
$\sigma^2_f + \sigma^2_g$		4.69	21.02	

In this context the meaning of the concept of heritability should be brought to mind. According to LUSH (1945), heritability is the proportion of the additive genetic variance in the phenotypic variance: $h^2 = V_a/V_p$. This can also be interpreted as the proportion of the additive genetic variance in the sum of the genetic and environmental variance: $h^2 = V_a/(V_g + V_e)$. In our model, however, the environmental variance is divided in two parts owing to different environmental correlations. It is a matter of interpretation whether we use both factors or either in order to define the heritability (KEMPTHORNE 1957 p. 464). It is, consequently, most uncertain to use the quotient of heritability in plant breeding without exactly defining in what circumstances it has been estimated. The greater the environmental variability the smaller the heritability! Here the plot to plot error variance is rather large and therefore it was considered necessary to include both sources of environmental variance in the denominator. Hence heritability is here computed as:

$$h^2 = \frac{\sigma^2_a}{\sigma^2_f + \sigma^2_e + \sigma^2_g}$$

This gives a fairly high heritability for height growth for both years which must depend on the rather uniform conditions in the seed beds located in the plastic greenhouse:

1 year	*2 years*
3.06/7.33 = 0.42	14.13/34.95 = 0.40

22. Diallel analysis

Our next object is to examine the structures of the diallel. In this context there is no need to deal with the details of their statistical background. All parameters, estimates and genetic components appearing in this design can easily be derived in the manner described above in connection with the polycross. All genetic and statistical premises enumerated above are important in this case, too, perhaps even more important than in the polycross, since it is possible to make a much more detailed genetic examination.

The general modified diallel cross, used here, is derived from the classical diallel where all individuals (or pure lines) are crossed reciprocally including the self-pollinations (JINKS 1954, HAYMAN 1954). Originally the diallel was particularly used as a cross desing of »pure lines» presupposing that the coefficients of inbreeding of the lines were known. When the diallel was applied to naturally cross-breeding plants the coefficient of inbreeding was supposed to be zero. This condition is extremely important and still requires much consideration, since anemophilous or entomophilous pollination of natural populations might possibly form inner groups of relatives in the population.

The classic diallel is of course a most exact mating design but it is often difficult to make use of it in practice because of its laboriousness. Its modification (Fig. 1) where crosses are made in one direction (not reciprocally) and selfings are left out, still gives almost the same amout of information but requires considerably less work. Its handicap is that it does not indicate whether there are reciprocal maternal effects. It is very easy to analyse the modified diallel, since all individuals in the design take part in the same number of combinations (KEMPTHORNE 1957 p. 465 *etc.*). A draw-back, on the other hand, is the fact that all trees are paternal trees as well, and for this reason one needs to collect pollen from all cross partners of the diallel.

		♀ A	B	C	D
	A		x	x	x
♂	B			x	x
	C				x
	D				

Fig. 1. The general modified diallel cross.

In 1964 crosses were carried out according to the general modified diallel at the above-mentioned seed orchard. The cross-seed was sown in connection with the polycross trial presented above, and within the same randomized block design. However, some of the combinations resulted in so few seeds that it was insufficient for the full 104 seeds per plot. Nevertheless a seed spacing of 4 × 4 centimeter was kept to but some of the plastic basins were only half-filled. There was not sufficient seed for countercurrent sorting and all seeds were sown regardless of their size and weight. This will have a disadvantageous effect on the reliability of the trial, especially if the measurements are made at an early stage (HADDERS *loc. cit.*).

The structure of the analysis of variance has been laid out by KEMPTHORNE (1957) and later by HINKELMANN and STERN (1960):

1) analysis of plot means

Source of variation	*d.f.*	*Sum of squares*	*Mean squares*
Replicates	r − 1	$\sum_{k} \frac{Z^2_{..k}}{n} - \frac{Z^2_{...}}{rn}$	
♀♀ ; ♂♂	p − 1	$\sum \frac{Q^2_i}{r(p-2)} - \frac{2(Z^2_{...})}{rp\frac{(p-2)}{2}}$	S

Source of variation	*d.f.*	*Sum of squares*	*Mean squares*
♀♀ × ♂♂	$p\,\frac{(p-3)}{2}$	$\sum_{ij} \frac{Z^2_{ij.}}{r} - \sum_{i} \frac{Q^2_i}{r\,(p-2)} + \frac{Z^2}{r\,(p-1)\,\frac{(p-2)}{2}}$	R
Crosses × repl.	$(n-1)\,(r-1)$	Difference	I
Total variation	$rn-1$	$\sum_{ijk} z^2_{ijk} - \frac{Z^2_{...}}{rn}$	

2) analysis of individual plant data

Source of variation	*d.f.*	*Sum of squares*	*Mean squares*
Between plots	$rn-1$	$\sum_{ijk} \frac{Y^2_{ijk.}}{n_{ijk}} = \frac{Y^2_{....}}{N_{...}}$	
Within plots	$N_{...}-rn$	Difference	E
Total variation	$N_{...}-1$	$\sum_{ijkl} y^2_{ijkl} - \frac{Y^2_{....}}{N_{...}}$	

3) interpretation

Parameter	*Estimate*	*Genetic component*
$Cov_{(HS)}$	$\frac{S-R}{r\,(p-2)}$	$1/4\ V_a$
$Cov_{(FS)}-2Cov_{(HS)}$	$\frac{R-I}{r}$	$1/4\ V_d$
σ^2_e	$I-n_hE$	
$\sigma^2_f+\sigma^2_g$	$E+Cov_{(FS)}$	

where replicates = r, ♀♀ and ♂♂ = p, yijkl = the height of the l:th plant in a cross where the parents are i and j in replicate k, z_{ijk} = plot mean, $Q_i = z_{i..} + z_{.j.}$, n_{ijk} = the number of plants per plot, n = number of crosses = $\frac{p\,(p-1)}{2}$, n_h = reciprocal of the harmonic mean of the plants per plot.

All statistical facts mentioned in connection with the polycross are also valid for the diallel. It should be kept in mind that the expected mean square between plots is now $\sigma^2_f + (\sigma^2_g - Cov_{(FS)})$. The height measurements after one and after two growth periods have been subject to analysis and the results are given below.

Obviously this very small diallel gives very unreliable estimates of heritability. The author has no explanation to the sudden change in heritability after the second growth period. It only shows that heritability estimates have to be done over a number of years and that a sufficient amount of parent trees should be included Generally heritability increases with increasing age of the plants (Stonecypher 1966).

Source of variation	*d.f.*	*M.S. 1. year*	*M.S. 2. years*	*Symbol*
Replicates	2	6.03	26.75	
♀♀ ; ♂♂	3	4.90	5.70	S
♀♀ × ♂♂	2	2.98	17.73	R
Crosses × repl.	10	2.20	13.77	I
Within plots		4.56 (n_h=0.04)	22.02 (n_h=0.09)	E

interpretation:	*1 year*	*2 years*
$Cov_{(HS)}$	0.32	negative
$Cov_{(FS)}-2Cov_{(HS)}$	0.26	1.32
σ^2_a	1.28	negative
σ^2_d	1.04	5.28
σ^2_e	2.01	11.79
$\sigma^2_f+\sigma^2_g$	5.46	23.34
$h^2 = \frac{\sigma^2_a}{\sigma^2_f+\sigma^2_e+\sigma^2_g}$	0.17	negative

23. Factorial desing

The most suitable cross design in each case mainly depends on the reproductive manner of the species in question. Members of the genus *Populus* are for instance dioecious and for this reason they are not applicable to the diallel. In such case a suitable factorial design has to be used. Usually only a few (4—10) males are used in factorial designs and these are crossed with all females; but it is of course just as possible to use only a few many-flowered mothers each of which is crossed with a greater number of fathers; the direction of the cross only depends on technical matters. In plant breeding the factorial matings-design of the type AB has been named »design II» by its originators (Comstock and Robinson 1952). It has been used extensively in the North Carolina State-Industry Program for testing seed orchards of *Pinus taeda* (L.) as well as in several other tree breeding programs. (Zobel and Kellison 1963).

A genetic test on the Scots pine can easily be carried out with the help of a factorial design (Fig. 2) owing to the fact that large amounts of desired pollen can be collected from only a few trees used paternally. Usually clones in orchards may serve as pollen producers but very large quatities of pollen of desired genotype can be collected directly from the plus-trees. Under proper storage this pollen can be kept viable for a number of years. Hence it is easy to repeat unsuccessful crosses in subsequent years. A full design often takes years to cross as some crosses almost always will fail to produce seed.

In some respect this design is not quite as good as the diallel since, by using only a few males, strong male x female interactions are brought about. However, it is perhaps possible to lessen this error by selecting the male partners so that they represent the population tested both geographically and phenotypically. This is a delicate question however; by selecting representative fathers we are breaking against the demand of random sampling. A sample of four fathers can easily become very biased if it is randomly picked; it may e.g. only represent a certain stand of the numerous stands involved.

	♀ A_1	A_2	A_3	A_4	.	.	.	A_d
	B_1 x	x	x	x	.	.	.	x
	B_2 x	x	x	x	.	.	.	x
♂	B_3 x	x	x	x	.	.	.	x
	. .	.	.	.	.	.	.	.
	. .	.	.	.	.	.	.	.
	B_s x	x	x	x	.	.	.	x

Fig. 2. Factorial matings-design of type AB

An AB cross design was carried out in the same seed orchard parallel with the polycross and the diallel. It was also incorporated in the same randomized block design with the other two. Four males and 21 females were included in the cross, that is, all the clones in the seed orchard were used. Some of the crosses did not succeed, however, and the final material to be studied thus consisted of only 4 males and 15 females. A reduction was carried out in order to keep the trial orthogonal; if more than one of the four males failed to produce seed in a certain female combination the whole group was left out from the analysis. If only one of the four male combinations was missing, this was compensated by the mean of the male and female in the other successful crosses. The main part of the crosses were carried out in June 1964 but the design was completed during the following year. The analysis and its interpretation is given below.

1) analysis of plot means

Source of variation	*d.f.*	*Sum of squares*	*Mean squares*
Replicates	$r-1$	$\sum_{k} \frac{Z^2_{..k}}{sd} - \frac{Z^2_{...}}{rsd}$	

Source of variation	*d.f.*	*Sum of squares*	*Mean squares*
Paternal plants	s − 1	$\sum_{i} \frac{Z^2_{i..}}{rd} - \frac{Z^2_{...}}{rsd}$	S
Maternal plants	d − 1	$\sum_{j} \frac{Z^2_{.j.}}{rs} - \frac{Z^2_{...}}{rsd}$	D
Males × Females	(s − 1) (d − 1)	$\sum_{ij} \frac{Z^2_{ij.}}{r} - \sum_{i} \frac{Z^2_{i..}}{rd} - \sum_{j} \frac{Z^2_{.j.}}{rs} + \frac{Z^2_{...}}{rsd}$	SD
Error[1])	(sd − 1) (r − 1)	Difference	I
Total variation	rsd − 1	$\sum_{ijk} z^2_{ijk} - \frac{Z^2_{...}}{rsd}$	

[1]) male-female combinations x replicates interaction.

2) analysis of individual plant data

Source of variation	*d.f.*	*Sum of squares*	*Mean squares*
Between plots	sdr − 1	$\sum_{ijk} \frac{Y^2_{ijk.}}{n_{ijk}} - \frac{Y^2_{....}}{N...}$	
Within plots	N... − sdr	Difference	E
Total variance	N... − 1	$\sum_{ijkl} y^2_{ijkl} - \frac{Y^2_{....}}{N...}$	

3) interpretation

Parameter	*Estimate*	*Genetic component*
$Cov_{(HS)} = \sigma^2_S$	$\frac{S-(SD)}{rd}$	1/4 V_a
$Cov_{(HS)} = \sigma^2_D$	$\frac{D-(SD)}{rs}$	1/4 V_a
$Cov_{(FS)} - 2Cov_{(HS)}$	$\frac{(SD)-I}{r}$	1/4 V_d
σ^2_e	$I - n_h E$	
$\sigma^2_f + \sigma^2_g$	$E + Cov_{(FS)}$	

where replicates = r, males = s, females = d, y_{ijkl} = height of the l:th plant in the cross $male_i$ x $female_{ij}$ in the replicate k, z_{ijk} = plot mean, n_{ijk} = the number of plants per plot, n_h = reciprocal of the harmonic mean of the plants per plot.

The differences between the diallel and the factorial design are in the structure of the expected mean squares. There are half- and full-sib relationships in both analyses. Therefore in the factorial design, too, the expected mean squares within plots are $\sigma^2_f + (\sigma^2_g - Cov_{(FS)})$.

The results for height growth of the first and the first and second vegetation period are presented below.

Source of variation	*d.f.*	*M.S. 1 year*	*M.S. 2 years*	*Symbol*
Replicates	2	25.71	119.36	
Males	3	13.11	36.64	S
Females	14	3.87	34.06	D
Males × Females	42	5.27	35.17	SD
Error	118	5.72	32.23	I
Within plots		4.48 (n_h = 0.03)	19.48 (n_h = 0.04)	E

Interpretation:	*1 year*	*2 years*
$Cov_{(HS)} = \sigma^2_S$	0.17	0.03
$Cov_{(HS)} = \sigma^2_D$	neg.	neg.
$Cov_{(FS)} - 2Cov_{(HS)}$	neg.	0.98
$\sigma^2_a = 4\,\sigma^2_S$	0.68	0.12
σ^2_d	neg.	3.92
σ^2_e	5.59	31.53
$\sigma^2_f + \sigma^2_g$	4.82	19.54
$h^2 = \frac{\sigma^2_a}{\sigma^2_f + \sigma^2_e + \sigma^2_g}$	0.07	0.002

3. DISCUSSION

On the whole heritability estimates from tree plants only a few years old are apt to be highly unrealible (see e.g. STONECYPHER 1966 or ILLY 1967). It seems reasonable to conclude that progeny testing of seed orchards of *Pinus silvestris* must be based on field trials that cover at least 1/4 of the mature age of this species, or in other words around 15—20 years. At that age Scots pine trees will have reached a height that makes it difficult to measure them using an ordinary stick. Of course other traits must be considered also and some of the wood properties can be measured at the end of the 20 year trial. Can we then make any early estimates of genetic gain when our tests show such a very pronounced heterogeneity at this early stage? The only possibility seems to be to have large enough trials so that our estimates are statistically as reliable as possible. In this Scots pine investigation there are six different estimates on heritability of early height growth, two from each cross design. It is commonsense that we must try to calculate an average heritability that must be the most reliable single estimate. This average must of course be corrected as soon as we can obtain better estimates from older field trials.
The different heirtability estimates are given in tabular form below. (Table 1).

Table 1. Heritability estimates of height growth in *Pinus silvestris*.

Polycross	*heritability*	*Standard error*
1 year	0.42	0.28
2 years	0.40	0.30
Diallel		
1 year	0.17	0.22
2 years	0.00	(0.17)
Factorial		
1 year	0.07	0.07
2 years	0.002	0.03
Mean	0.18	0.18

The heritability was above defined by the help of the division of genetic variance. The fact has been established that the value of the heritability substantially depends on the trial circumstances. In field trials the trial individuals are so organized that the plot either consists of half- or of full-sibs. This clearly influences the heritability since genetically homogeneous plots do not appear in nature or in artificial plantations. To avoid this trouble a recent method in plant breeding aims at defining the heritability on the basis of selection differential. According to this the heritability is that part of the selection differential which is supposed to be inherited when selection is based on a defined comparative unit — the standart deviation (HANSON 1963).

The genetic gain $\triangle G$ can be estimated by the formula

$$\triangle G = h^2 S$$

where S is the selection differential and h^2 is the heritability. The selection differential implies the difference of the means of the entire population and the specially selected population. In other words $\triangle G$ implies the repeatability of the mass selection in the next generation. The selection differential can also be expressed by the standard deviation of the non-selected population and it is then called intensity of selection (i). In this case $i = S/\sigma_p^2$ and the genetic gain $\triangle G = i\sigma_p h^2$ (FALCONER 1964 p. 192 etc.). This formula can be used when the selection differential is not known since it can be estimated when the proportion of the selected population is known (for table of i see e.g. BECKER 1967).

Many agricultural plants are annual and the harvest marks the end of their life cycle. A heritability estimate will thus comprise the total production. However, forest trees are in this respect exceptional since the growth periods follow each other and the harvest is gradual. Concerning forest trees one is compelled to study the yield characteristics for many years but yet one wants to have some early estimates for the making of predictions. Several trials have shown that reliable results concerning the genetics of growth traits can only be had when 1) the physiologic differences of the seed no more influences growth (HADDERS *loc. cit*) (may it be a maternal effect or a genetic interaction between embryo and endosperm or a purely nutritional conditional) and 2) the differences depending on transplanting of the seedlings have been levelled (TIGERSTEDT 1966).

All progeny trials that do not follow the development until mature age (time of harvesting) are »early tests», i.e. the results of the tests are extrapolated till the end of the life cycle. Many investigations point at the fact that the main part of the information can be had 5—10 years after sowing (NANSON 1967). Considering Finlands boreal climate and slow plant growth a proposed test period of 15—20 years seems plausible.

The heritability values calculated here are certainly quite unreliable. This can also be judged from their standard errors given in table 1. It is a fact, however, that the results from these first two periods of growth are the most reliable one can get within the next five years! The plants were transplanted in the spring of 1969 and this treatment will blur heritability estimates for at least the next 5 years (TIGERSTEDT *loc. cit.*). The mean of six heritability estimates was 0.18 which seems a little high when height growth is concerned. The heritability of *Pinus monticola* (Lamb.), for instance, has been found to be 7 % as calculated on the basis of fourth year height growth (SQUILLACE *et al. loc. cit.*).

ILLY (1967) concluded that there is very little correlation of height growth of six year old plants to their 30 year old parents in *Pinus pinaster* (Ait.) but there is, however, a very strong correlation of wood density. Generally it seems reasonable to expect that traits of great fitness value, like juvenile growth rate, will be very poorly correlated to mature age but traits of lesser fitness value will correlate better between juvenile and mature age. Branching habits, wood densities, cellulose contents and other wood properties appear to be better correlated. It is well known that the more a trait contributes to fitness the less additive genetic variance it possesses that is the lower is its heritability (FALCONER 1964 p. 167). Careful classification of different traits on a fitness basis together with investigations considering the strength of natural selection in tree populations may reveal a number of economically valuable traits which readily respond to mass-selection. It appears that heritability estimates of the kind presented in this investigation are valuable but, owing to the very long life of trees, they have to be substantially completed by investigations aimed at the classification of characters in relation to fitness and by other investigations that aim at a better understanding of population structure and population strategy (TIGERSTEDT 1967).

One of the most difficult questions of forest tree breeding is to decide on the characters which ought to be recognized. It is apparent that growth is a central trait, but in addition it is important to estimate the bearing of wood quality. The more numerous the traits simultaneously considered the smaller will be the genetic gain in each separately as long as they are not tightly linked. Linkage of different traits is another question that is very important for efficient tree breeding; here is another field for further genetic investigation.

The best practical result through mass selection will perhaps be attained by using some kind of selection index which includes a group of economically important traits. The final genetic gain ought to be expressed by an economic quantity. The most difficult problem in forest tree breeding is to be found in the making of these estimations since the lag between selection and its impact is so long that economic premisses may change meanwhile.

4. SUMMARY

Heritability of first and of first and second year height growth of *Pinus silvestris* (L.) plants was studied using three different mating designs.

Plants grew in a plastic greenhouse during the first growth period. During the second growth period they were exposed to open air conditions.

Heritability values varied from zero to 42 %. An average heritability based on six separate calculations (two from each mating design) was computed and its reliability was discussed. This heritability value was 18 %.

Different ways of improving heritability estimates was discussed and the need of more investigations on trait quality in relation to fitness and on the effect of natural selection and population strategy was emphasized.

5. REFERENCES

Bekker, W. A. 1967. Manual of Procedures in Quantitative Genetics. Wash. State Univ., Pullman, Washington.

Cockerham, C. C. 1963. Estimation of genetic variances. In Statistical Genetics and Plant Breeding (eds: Hanson, W. D. and H. F. Robinson. Nat. Acad. Sci., Wash. D. C., 53–93.

Comstock, R. E. and H. F. Robinson 1952. Estimation of average dominance of genes. In Heterosis (ed: J. W. Gowen). Iowa State College Press, 494–516.

–»– and R. H. Moll 1963. Genotype-Environment Interactions. In Statistical Genetics and Plant Breeding (eds: Hanson, W. D. and H. F. Robinson). Nat. Acad. Sci., Wash. D. C., 164–196.

Falconer, D. S. 1964. Introduction to Quantitative Genetics. (Reprinted with amendments). Oliver and Boyd, Edinburg and London.

Hadders, G. 1967. Arftlig variation i plantutvecklingen hos tall (Pinus silvestris L.). Akademisk avhandling. Akademisk maskinskrift, Uppsala.

Hanson, W. D. 1963. Heritability. In Statistical Genetics and Plant Breeding (eds: Hanson, W. D. and H. F. Robinson). Nat. Acad. Sci., Wash. D. C., 125–140.

Hayman, B. I. 1954. The theory and analysis of diallel crosses. Genetics 39, 789–809.

Hinkelmann, K. und K. Stern 1960. Kreuzungspläne zur Selektionszüchtung bei Waldbäumen. Silvae Genetica 9, 121–133.

Illy, G. 1967. Recherches sur l'amélioration génétique du pin maritime. Extrait des Annales des Sciences Forestières, Tome XXIII – Fascicule 4 – 1966.

Jinks, J. L. 1954. The analysis of heritable variation in diallel cross of Nicotiana rustica varieties. Genetics 39, 767–788.

Kempthorne, O. 1957. An Introduction to Genetic Statistics. Wiley and Sons Inc., New York.

Kidwell, J. F. and O. Kemthorne 1966. An experimental test of genetic theory. Der Züchter 36, 163–167.

Lush, J. L. 1945. Animal Breeding Plans (3rd edn.). Iowa State College Press, Ames.

Nanson, A. 1967. Contribution à l'etude de la valeur des tests précoces. II. Experience internationale sur l'origine des graines de pin sylvestre (1906). Administration des Eaux et Forêts. Travaux – Série E, No 2.

Sarvas, R. 1962. Investigations on the flowering and seed crop of Pinus silvestris. Comm. Inst. forest fenn. 53.

Squillace, A. E., Bingham, R. T., Namkoong, G. and H. F. Robinson 1967. Heritability of juvenile growth rate and expected gain from selection in Western White pine. Silvae Genetica 16, 1–6.

Stonecypher, R. W. 1966. Estimates of genetic and environmental variances and covariances in a natural population of Loblolly pine (Pinus taeda L.). Ph. D. thesis of North Carolina State University.

Tigerstedt, P. M. A. 1966. Entwicklung der genetischen Varianzen des Höhenwachstums in einem Feldversuch mit Betula verrucosa. Silvae Genetica 15, 136–137.

–»– 1967. Quantitative genetics in provenance research of forest trees. IUFRO, München, papers III, 395–412.

–»– 1969. Experiments on selection for developmental rate in Drosophila melanogaster. Ann. Acad. Sci. fenn. A. IV. 148.

Zobel, B. and R. Kellison 1963. Progeny testing seed orchards in the cooperative industry – North Carolina State tree improvement program. Duplicate.

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