PROCEEDINGS

OF THE

TWENTY-FIFTH MEETING

OF THE

CANADIAN TREE IMPROVEMENT ASSOCIATION

Part 2

Held in
Victoria, British Columbia
August 28 - September 1, 1995

Editor:
J. Lavereau

COMPTES RENDUS

DE LA

VINGT- CINQUIÉME CONFÉRENCE

DE

L'ASSOCIATION CANADIENNE POUR L'AMÉLIORATION DES ARBRES

2 e PARTIE

Victoria, en Colombie-Britannique
du 28 août au 1 septembre 1995

Rédactrice:
J. Lavereau

TABLE OF CONTENTS/TABLE DES MATIERES

THE CONFERENCE

Page

WELCOMING REMARKS

EXPERIENCE FROM THEORETICAL AND EMPIRICAL WORK IN QUANTITATIVE GENETICS

APPLIED CONSIDERATIONS FOR BREEDING POPULATION STRUCTURE AND SELECTION CRITERIA

THE USE OF MOLECULAR MARKERS IN QUANTITATIVE GENETICS AND BREEDING

NORTH AMERICAN QUANTITATIVE FOREST GENETICS

Estimating Inbreeding Depression

Phenotypic and Marker Approaches to Inbreeding Depression

Among-Population Variation and Experimental Inbred Populations

MOLECULAR GENETICS OF DISEASE

GENERAL TOPICS OF TREE IMPROVEMENT

GENERAL BIOCHEMICAL FOREST GENETICS

QUANTITATIVE GENETICS OF DISEASE

GENERAL MOLECULAR GENETICS

GENERAL TOPICS IN FOREST GENETICS

BREEDING STRATEGIES

WORKSHOP: GENETICS OF WOOD QUALITY

WORKSHOP: TREE SEED WORKING GROUP

SYNTHESIS, CONCLUSION AND OUTSTANDING PROBLEMS IN ADVANCED GENERATION BREEDING IN FOREST TREES

POSTER: ABSTRACTS

PARTICIPANTS OF THE 25TH CTIA/ACAA MEETING

WELCOMING REMARKS

THEME

Evolution and Tree Breeding:
Advances in Quantitative and Molecular Genetics for Population Improvement.

BACKGROUND

Research developments in genetic theory, molecular genetics and applied breeding in animal crop species have been substantial over the last two decades. A vast amount of information has accumulated particularly in the area of genetic response to selection over many generations and understanding the basis of these responses.

Research and breeding efforts directed toward forest trees is relatively new, compared to those efforts directed toward animals and crop species. Many tree breeding programs around the world are just starting to move into the second and third generation of breeding; however, breeding strategies chosen today will deeply influence the options available to breeders in the future.

The purpose of this conference is to explore current issues in animal and plant breeding in order to allow geneticists, tree breeders and other tree improvement personnel an opportunity to discuss and consider this information within the context of tree breeding theory and application. Fundamental approaches to applied tree breeding, as it is today, are unlikely to change, but this exploration should prove useful in developing or refining existing breeding strategies for forest trees.

HOSTS

This joint conference of the Canadian Tree Improvement Association and the Western Forest Genetics Association, is hosted by the British Columbia Ministry of Forests and by the Centre for Forest Biology, University of Victoria.

WELCOME

On behalf of the B.C. Forest Service and the University of Victoria, I would like to welcome you to Victoria, British Columbia, and thank you for participating in the 1995 joint meeting of the Canadian Tree Improvement Association (CTIA) and the Western Forest Genetics Association (WFGA). This is the first joint meeting of these two organizations and we feel the timing could not be better: many tree breeding programs in Canada and the Pacific Northwest are dealing with similar issues in forest tree genetic resource management. These issues are also relevant to breeding programs in many parts of the world, and we especially want to welcome those of you who have traveled to North America for this meeting.

The theme for this meeting, "Evolution and Tree Breeding," was inspired by a 1989 book entitled "Evolution and Animal Breeding." It is a book in honor of the late Alan Robertson composed of several review papers by leading geneticists in the field of animal breeding (the book was edited by W.G. Hill and T.F.C. Mackay). When this book first came out several of us in the B.C. Ministry of Forests, Forest Genetics group, were impressed with the coverage of material related to the genetics of artificial selection and evolution. The organizing committee of this CTIA/WFGA conference thought that a meeting dealing with the same general issues, but relevant to the breeding of forest trees, would be of great value to practitioners of applied tree improvement. We have attempted to cover many of the issues brought forward in the "Evolution and Animal Breeding" book by inviting speakers who are leaders in genetics research, but who are also well aware of the special challenges that forest trees provide. And, as a special treat, we are lucky to have J.S.F. Barker, who is an author of an excellent paper entitled "Population Structure" in the "Evolution and Animal Breeding" book. Since the fields of quantitative and molecular genetics seem to be coalescing, we felt it would be important to add a component of molecular genetics to the meeting: hence, the sub-title of "Advances in Quantitative and Molecular Genetics for Population Improvement" was added to our main theme. The discussions on how these two areas of genetics research can compliment each other should prove interesting.

As with any meeting, it is the people and the discussions which we typically remember and find the most valuable for information exchanges. We hope this meeting will be no different, but that you will use the presentations, whether they are invited or contributed; as a starting point for developing new questions or ideas. The real intent of the conference is not to provide "answers" to operational tree breeding problems, but to summarize current knowledge in the field of genetics and tree breeding, so that breeders can develop their own strategies from a genetically informed basis, and to provoke thoughts for future work.

Many people are needed to put together a meeting such as this, and I would like to quickly acknowledge their help: John Russell, Michael Stoehr, and Pat McGuire (Organizing Committee), John Owens (University of Victoria sponsor), Jack Woods (field trips), Michael Carlson (art designs), Sally Aitken (WFGA Chair) and John Barker (Western Forest Product), Yousry El-Kassaby (Pacific Forest Products) and Glen Dunsworth (MacMillan Bloedel) for industrial sponsorship of field tours.

It was an added bonus to have the North American Forest Quantitative Genetics Group meeting in conjunction with this CTIA/WFGA meeting. The topic of "Inbreeding Depression in Conifers" is timely and will add to the technical aspects of this meeting greatly. I would like to thank Claire Williams for her efforts in organizing and bringing this group together here in Victoria.

Once again, the Tree Seed Working Group of the CTIA is holding a session, and I would like to thank Dave Kolotelo for putting together a program for those who work in the important field of seed production. This has always been a strength of the CTIA and WFGA organizations, in that it provides a forum for communication between operational and theoretical tree improvement personnel. The Wood Quality Working Group is also meeting as one of the concurrent sessions, and I hope the papers presented will lead to renewed interest in this important field.

Last, but not least, we wanted to take this opportunity to acknowledge the pioneering efforts of the late Dr. Alan Orr-Ewing, and dedicate this meeting in his honor. Reni Stettler and Chris Heaman have agreed to share some of their thoughts about Alan with us. Thanks to both of you.

If you have any questions or need assistance with any matters, please feel free to contact any one of us on the conference planning committee, and, above all, enjoy your stay in Victoria.

Alvin Yanchuk
Chair, Local Organizing Committee

Invited Symposia Speakers

"Ok now, 3 more big steps back"

(from left to right)

Gene Namkoong, Jim Brewbaker, Sue Carson, Stuart Barker, Floyd Bridgwater, Doug Shaw, Toby Bradshaw, Claire Williams, Rowland Burdon

EXPERIENCE FROM
THEORETICAL AND
EMPIRICAL WORK
IN
QUANTITATIVE GENETICS

QUANTITATIVE GENETIC MODELS : PAST, PRESENT AND FUTURE CHALLENGES

J.S.F. Barker

Department of Animal Science
University of New England
Armidale NSW 2351. Australia

Quantitative genetics as it has been applied in plant and animal breeding is essentially the statistical analysis of phenotypic differences among individuals for a trait or traits showing continuous or quasi-continuous (i.e. quantitative) variation. Our concern is with the inheritance of these differences - hence we call it genetics, but until very recently we have known essentially nothing about the genes involved. With the advent of molecular genetics, this is changing, and I will come back to that later.

To begin, however, it will be useful to review the basic concepts and assumptions, i.e. the basic model of quantitative genetics. This infinitesimal model, due to Fisher (1918), assumes that the inherited differences between individuals are due to many unlinked genes, each of small effect on the phenotype, and that these effects are additive. For a particular trait, the measurement made on an individual (its phenotypic value - P) is conceived as due to the combination of all the genes it carries (its genotypic value - G) and an environmental deviation (E),

$$P = G + E$$

For each genotype at a single locus, the genotypic value is the average phenotype for individuals with that genotype. However, it is genes, not genotypes that are inherited, so that it is the effects of alleles that we need to know. The inherited effect of an allele is its average effect, defined as the mean deviation from the population mean of individuals which received that allele from one parent, the other allele (from the other parent) coming at random from the population (Falconer 1989). Then the genotypic value can be partitioned into the additive or breeding value (A) and a dominance deviation (D). The breeding value is the sum of the average effects of the two alleles carried by an individual, and is twice the expected value of an individual's offspring, given random mating. The dominance deviation is the deviation of the genotypic value from the breeding value. For more than one locus, interlocus genotypic interactions (epistasis) may cause a deviation (I) from additive expectations, so that the total genotypic value for an individual is:

$$G = A + D + I$$

Extending these concepts to the differences, or variation, among individuals:

$$\begin{aligned} V_P &= V_G + V_E \\ &= V_A + V_D + V_I + V_E \end{aligned}$$

Fisher (1918) also derived the relationship between the inherited variance (specifically the additive variance or variance of breeding values - V_A) and the phenotypic similarity of related individuals, thus providing a means for estimating V_A.

For some population of individuals that have been measured for a particular quantitative trait, the fundamental question of interest is - What proportion of the observed phenotypic variance is inherited? That is, what is the value of V_A/V_P, which is defined as the heritability of the trait. Given known relationships among individuals in the population, V_A and hence heritability can be estimated. The heritability of a trait also may be defined as the proportion of parental superiority for that trait which is transmitted to progeny. Strictly, any estimate of heritability applies to the population for which it was estimated - a population with a particular set of genotypes, and with individuals exposed to a particular set of environments. But for most traits of interest in plant and animal breeding programs where many estimates of heritability are available, these estimates generally do not vary greatly, so that in the absence of information for a given population, an average may be used.

Knowledge of the heritability of a trait is of primary value in considering two inter-related questions - what is the optimum selection program for genetic improvement of the trait, and what are the predicted rates of genetic improvement (i.e. selection response). Before commenting further on these, there is an additional aspect of the infinitesimal model to be noted. Rarely in breeding programs are we interested in improving just one trait, and the traits of interest are not necessarily independent. In a way analogous to heritability estimation using the phenotypic similarity of relatives for a single trait, the phenotypic covariance for two traits in relatives is used to estimate the genetic correlation between these traits. Then just as heritability allows prediction of selection response for one trait, knowledge of a genetic correlation allows prediction of the correlated response in one trait when the second trait is under selection.

Prediction of selection response

For a single trait, the expected response to selection (R) is:

$$R = h^2 S$$

where h^2 = heritability of the trait

S = selection differential, i.e. mean phenotypic value of selected parents expressed as a deviation from the population mean.

Equivalent expressions are:

(1) $$R = i \sigma_P h^2$$

where i = intensity of selection, or the selection differential expressed in units of phenotypic standard deviations,

σ_P = phenotypic standard deviation.

(2) $$R = i\,h\,\sigma_A$$

where h = square root of heritability

σ_A = additive standard deviation, or standard deviation of breeding values.

In these prediction equations, R is of course the expected response per generation, but in breeding programs, years are the real unit of time, so that assessment of expected responses for alternative possible selection designs must be in terms of response per year:

$$R_y = R/L$$

where L = generation interval (in years).

Although the apparent emphasis here is on prediction of response per se, the real issue is the comparison of expected responses for different selection designs, the aim being to find the design that will maximize economic gain per year.

All of this application of quantitative genetics in developing breeding programs is based on the infinitesimal model, and there are two problems with this approach. The first problem, and I alluded to it earlier, is that the model makes a set of assumptions about the genetics of the traits, but in fact we work at the phenotypic level, making unknown changes in an unknown genotype, and where the pathways between the genotype and the phenotype are largely inside a closed and unknown black box. Any data on the real nature of the underlying genetic variation should allow increased efficiency of breeding programs. Such data for any trait would include (a quantitative geneticist's wish list):

(i) Number of loci contributing to heritable variation, or at least the number that have significant effects,
(ii) Number of alleles segregating at each locus,
(iii) Allele frequencies,
(iv) Nature and magnitude of allelic effects at each locus,
(v) Effects of dominance,
(vi) Nature and magnitudes of inter-locus interactions,
(vii) Linkage relations among loci, and
(viii) Mutation rates.

The second problem with application of the infinitesimal model and at first sight it might not be considered it a problem, is that in general it works. Experimental selection studies with laboratory animals (*Drosophila*, *Tribolium*, mice) have shown that in the short term (5-10 generations), average responses over replicate populations are generally (but not always - see Sheridan, 1988; Hill and Caballero, 1992), close to those predicted. Where deviations from the predicted response do occur in the short term, they are most likely due to changes in allele frequency at a limited number of loci, and not to any departures from normality of breeding values produced by linkage disequilibrium (Turelli and Barton, 1994).

In any case, in tree breeding, as in animal breeding, none of us are going to be around for long enough to see anything more than short term responses. But we need to recognize that what is good in the short term (maximizing response per generation or per year) may not be good in the long term! Long term response and limits to selection cannot be predicted on the infinitesimal model, because they depend on the factors in my geneticist's wish list.

Nature of quantitative genetic variation

Understanding the nature of quantitative genetic variation, and the forces that maintain that variation in natural populations is then a major challenge. While it is unrealistic to expect that we can fulfill the wish list for any particular quantitative trait, even limited information would allow a more effective modelling of the relations between genotype and phenotype, and hence the development of more efficient breeding programs.

How many loci?

The number of loci and their effects cannot be readily separated, because to some extent, they must be inversely related. For a given amount of genetic variation, if there are few genes, they must have large effects, while if there are many genes they must have small effects. In any case, all the genes are unlikely to have equal effects, and Robertson (1967) suggested that the distribution of gene effects is highly leptokurtic, with a few genes having large effects and most having small effects. Thus rather than the total number of loci, the more interesting and of course more tractable question is how many loci contribute to the greater part (say 80 - 90%) of the genetic variance. Shrimpton and Robertson (1988 a, b) found that 17 regions of the third chromosome of *Drosophila melanogaster* accounted for the difference in sternopleural bristle number between a high selection line and a low bristle number tester line. About a third of the difference was due to three or four regions with effects of one phenotypic standard deviation or more, and in agreement with the expectation of Robertson (1967), the distribution of effects was highly leptokurtic.

Genes of large effect on the selected quantitative trait have been found in many selection experiments (Mackay, 1990), and indirect evidence for genes of large effect has come from long term selection experiments (Hill and Caballero, 1992). Where alleles of large effect have been detected (primarily in *Drosophila* bristle number and mouse body size selection lines), it is not known whether these have arisen as new mutations, or are due to the changed genetic background enhancing their expression, or were present in the unselected population. In *Drosophila* abdominal bristle number selection lines, alleles of scabrous have been found a number of times. Flies homozygous for scabrous have large, rough eyes, some major bristles missing, but other bristles subject to twinning and there are extra small bristles. In one case, if scabrous had been present in the unselected base population, its frequency must have been less than 1 in 4000 (Hollingdale, 1971), suggesting that it had probably arisen as a de novo mutation during selection. Nevertheless, alleles of large effect for various traits have been found in domestic animal species, e.g. the dwarf gene in poultry (Guillaume, 1976), the Booroola, Inverdale and Javanese genes for prolificacy in sheep (Piper and Bindon, 1982; Davis et al., 1991; Bradford et al., 1986), the halothane gene for stress and performance traits in pigs (Webb et al., 1982), the double muscling gene in cattle (Hanset, 1982), and the callipyge muscling gene in sheep (Cockett et al., 1994).

For most quantitative geneticists, one of the implicit assumptions of the infinitesimal model has been that structural genes (i.e. genes whose nucleotide sequence codes for the amino acid sequence of a protein) are the basis of quantitative genetic variation. This assumption was questioned by Hohenboken (1982) and further discussed by Barker (1985a) in terms of variation in regulatory genes controlling enzyme activity, and possible control of complex physiological or morphological phenotypes, through regulation of fundamental developmental processes. On the other hand, Mackay and Langley (1990) have hypothesised that allelic variation at loci directly affecting the development of a particular trait is a major source of quantitative variation in that trait. The achaete - scute gene complex in *Drosophila melanogaster* has a central role in bristle development, and Mackay and Langley (1990) found that nearly 5% of the total quantitative genetic variation for bristle number is associated with DNA insertion variation at this locus. For the scabrous locus of *D. melanogaster*, DNA sequence polymorphisms among 47 chromosomes from a natural population contributed 13 and 8% of the total genetic variance for abdominal bristle number and sternopleural bristle number, respectively (Lai et al., 1994). Sites associated with variation in bristle number differed for the two bristle systems, and Lai et al. (1994) suggested that this may reflect the intricate regulation of expression of scabrous. Apparently none of these sequence variants exhibited the scabrous phenotype when homozygous, but selection (and possibly recombination) affecting these or other rare sequence variants may have contributed to the "mutational" origin of the scabrous phenotype in many abdominal bristle selection lines (Hollingdale, 1971, and references therein; Yoo, 1980 a).

Clearly genes of large effect should be considered in models of quantitative genetic variation and its maintenance in natural populations. In the development of breeding programs, finding and using such genes is of more immediate significance than asking what forces are operating to maintain their variation, and this is considered later in discussing molecular genetics.

Dominance and inter-locus interactions

As outlined earlier, the analysis of quantitative variation is commonly a partitioning of variance, with the genetic variance partitioned into additive, dominance and epistatic variance components. Clearly these are statistical concepts, and no assumptions are made about the mode of action of the genes affecting the trait. In particular, the existence of additive variance does not mean that any of the genes act additively. Nevertheless, it might be imagined that knowledge of the relative magnitudes of the three variance components would give some understanding of the importance of non-additive variation. However, even in laboratory populations under controlled environmental conditions, and using large, well-designed experiments with many different types of relatives, attempts to get good estimates of dominance and epistatic variance have not been very successful (Barker, 1974, 1979). Further, good estimates of these components still would be only statistical estimates that would not uncover the nature of gene action. These components are defined as deviations from additivity, so that even if most of the genetic variation is described as additive, there may still be complete dominance at some loci or strong epistasis among loci affecting the trait. For example, Lush (1945) demonstrated for two locus complementary epistasis, where the two alleles at each locus were at frequencies of 0.5, the total genetic variance would be partitioned as 4/7 additive, 2/7 dominance deviations, and only 1/7 epistatic deviations!

Analysis of means of parental, F_1, F_2, back-crosses and further generations derived from the F_2 and backcrosses, for divergent parental populations, can provide more information on gene action than partitioning of variance. Such generation means analysis or joint scaling test (Mather and Jinks, 1982) was initially developed for inbred parental lines, and extensively used for plant species, but the analysis is applicable to lines that are not homozygous, provided that close relatives are not mated in producing the various generations. But one should be warned before contemplating the setting up of an experiment to be analyzed in this way - all generations to be compared should be produced at the same time, and the results are specific just to the particular pair of divergent lines. An alternative approach to evaluating the importance of epistasis would be to use the general model of Kinghorn (1980), which is not limited to just two parental lines.

In contrast to analyses that are based only on phenotypic values, and are limited to assessing epistatic variance as a means of detecting epistatic gene action, direct measurement of epistasis and of its contribution to the variance components can be made when two-locus genotypic values are available (Cheverud and Routman, 1995).

In evolutionary quantitative genetics, the role of epistasis continues to be debated - primarily in the context of Sewall Wright's shifting-balance theory. Given substantial epistasis with respect to fitness, Wright (1977) argued that evolution would be faster in a structured population, i.e. one subdivided into partially isolated demes. A number of recent theoretical papers have examined various aspects of the theory (e.g. Moore and Tonsor, 1994; and references therein), but the importance of the shifting-balance process in adaptive evolution remains uncertain.

Wright's shifting-balance theory in fact was derived from his earlier work in animal breeding, where he first suggested (Wright, 1922) that selection would be more effective in a population subdivided into small lines, with regular selection between and crossing among these lines, than in a single large population. A number of experimental studies (reviewed by Barker (1989); López-Fanjul (1989), see also García et al., 1994) have compared selection responses in subdivided (with cycles of crossing or regular introgression) and undivided populations. Where selection was for abdominal bristle number in *Drosophila* or pupa weight in *Tribolium,* there was no advantage for subdivision - a result that should have been expected, given that genetic variation for these traits is primarily additive (Hill, 1989). However, where selection was for body weight in *Drosophila* (a trait expected to exhibit more epistasis) there were some advantages for subdivision. Further experimental evaluation is warranted for traits that do show significant epistasis. All of the experiments that have been done were initiated from unselected base populations. However, sublining with crossing or introgression may be an effective way to obtain further improvement in long term selected populations that are at or approaching selection limits. Such populations commonly (but not always, e.g. Roberts 1966) show high levels of residual genetic variability at the limit, often including strong epistasis (see reviews by Hill and Caballero, 1992; Moreno, 1994).

The use of several small replicate lines, each with the same selection intensity as a single large population, has been considered also in contexts other than for traits showing significant epistasis. With several small lines initially, and cycles of selection between lines, crossing the selected lines and starting a new set of small lines from the intercrosses, higher initial responses and selection limits might be attained than in a single large population. However, Madalena and Hill (1972), in a simulation study, compared these structures for an additive model, and for one with complete dominance and the recessive allele favoured by selection. For the additive model, subdivision and crossing may give some short term gains, but a lower limit. Where favoured recessive alleles were at low initial frequencies, however, subdivision and crossing schemes gave greater responses and a higher limit. More recently, Smith and Quinton (1993) examined subdivision and crossing schemes as compared with a single large population, comparing selection response and inbreeding - concluding that the response at the same level of inbreeding is greatest in a single large population.

Mutation

I noted earlier that one of the implicit assumptions of the infinitesimal model is that structural genes are the basis of quantitative genetic variation. An additional assumption has been that selection response for quantitative characters was due to changes in the frequency at such loci of alleles that were present in the base population when selection began. That is, the possible contribution of new mutations (in the broad sense including unequal crossing over, intragenic crossing over, chromosome rearrangements and transposable elements, in addition to single locus mutation) was ignored, or if considered at all, it was then dismissed as unlikely to play any significant role in selection response.

However, Yoo (1980 a, b) noted that mutation must have contributed to the long term (>80 generations) selection responses observed for abdominal bristle number in *Drosophila*, while Frankham (1980a) reviewed the evidence then available, and concluded that the contribution of mutations to genetic variation and selection response could not be ignored. Since then, the role of new mutations has been far from ignored, both in terms of the contribution to long term response of quantitative traits to directional selection, and to variation of quantitative traits within and among natural populations (see reviews by Hill and Keightley, 1988; Hill, 1990; Mackay, 1990).

Theoretical studies of the expected responses to artificial selection from new mutations have been due primarily to Hill and his colleagues in Edinburgh. For the simplest model of a homozygous base population and constant input of new variance by neutral additive mutants with effects symmetric about zero, the expected cumulative response is proportional to the effective population size, and the between line variance is proportional to the response. These predictions are modified in various ways for dominant or recessive mutants, asymmetric distributions of mutant effects, linkage or deleterious pleiotropic effects of the mutants on fitness (Hill, 1982; Keightley and Hill, 1990). In general, new mutants are not expected to contribute much to response for 20 or so generations, but then may contribute substantially (Franklin, 1982; Hill, 1982). Thus their potential contribution should not be ignored in designing and executing breeding programs (Hill, 1982).

Maintenance of genetic variation

In a finite population, genetic variation is lost due to drift of allele frequencies, with average heterozygosity decreasing at a rate of $1/(2N_e)$ per generation (N_e = effective population size).

Thus :

$$H_t = H_o (1 - 1/(2N_e))^t$$

where

H_o = initial heterozygosity

H_t = heterozygosity in generation t

(Crow and Kimura, 1970, Chapter 7), and at the limit, assuming no mutation or selection, $H_t = 0$. Apart from effects of finite population size on genetic variability for all traits, additive genetic variation for fitness, which is under strong directional selection, is expected to steadily decrease, eventually to zero in the absence of mutation and gene flow (input of variation from other populations).

Yet there is abundant genetic variability for most traits in natural populations, so that a central problem in evolutionary quantitative genetics is to determine what maintains genetic variation. Several hypotheses have been proposed, including mutation-selection balance, overdominance induced by genotype x environment interactions, frequency-dependent selection, spatial variation and pleiotropy (reviewed by Barton and Turelli, 1989). No one mechanism is likely to be all pervasive and no clear consensus has yet emerged, although Barton and Turelli (1989) suggest that "quantitative genetic variation for most morphological traits may be maintained by the pleiotropic effects of many diverse polymorphisms."

From the viewpoint of plant and animal breeding, where genetic variation is assumed and the concern is how best to utilize it, the factors maintaining genetic variation in natural populations may seem of little direct interest. However, increased knowledge and understanding of these factors could be very relevant to designing future breeding programs. One consequence of artificial selection is a reduction in fitness (Falconer, 1989), expected as a result of inbreeding due to finite population size. In addition, selection further reduces the effective population size, because of variation among families in the probability of selection (Robertson, 1961; Latter and Robertson, 1962), and this is expected also if the trait selected were subject to stabilising selection. Selection therefore increases the rate of approach to homozygosity at all loci, not just those affecting the trait under selection. If the factors maintaining genetic variation were understood, it may then be possible to manipulate a breeding program so as to minimize any reduction in fitness, while selecting to increase desired performance traits.

Effects of population size on selection response

Population size, as the number of individuals selected and used as parents in each generation, is under the control of the breeder, although constrained to some degree by operational factors. Thus it might be assumed that it would be advantageous to maximize population size so as to minimize loss of genetic variance from genetic drift and to minimize inbreeding depression.

However, population size effects need to be treated in conjunction with selection intensity, and further, we need to distinguish short term from long term responses. Short term response is maximized when the selection intensity is highest (i.e. small proportion selected), but at the expense of long term response. On the other hand, maximum response at the limit is expected from much lower selection intensities, so short term response is then reduced. In fact, maximizing the ultimate response depends on quite weak selection. Robertson (1960) predicted that maximum response at the limit would be expected for a proportion selected of 0.5. However, computer simulations by Hospital and Chevalet (1993) indicate that as population size (number of individuals measured) increases, the optimum proportion selected is shifted to values higher than 0.5. But if the primary objective is to maximize genetic gains in the short term, say 5 to 10 generations, then population size does not have much effect (Hill, 1985). However, the size of the initial sample (number of founders) used to initiate the breeding program is important. James (1971) showed that for a sample of N founders, the expected response in the first generation is proportional to $(1 - 1/(2N))$, so that short term responses would be reduced by $1/(2N)$, as compared with lines not subject to an initial bottleneck. Experimental studies with Drosophila have shown good agreement with this theoretical prediction (Frankham, 1980b).

In the long term, selection responses and limits depend on gene effects and frequencies (reviewed by Hill, 1985; López-Fanjul, 1989), but the limit is a function of Ni (N = population size, i = standardized selection differential), as shown by Robertson (1960). The expectation of increased long term response and limits with increased population size has been demonstrated in Drosophila (Jones et al., 1968) and mice (Eisen, 1975). Jones et al. (1968) used 20 to 80 parents each generation, but increased responses with increased population sizes were extended recently to 1600 parents by Weber (1990a, b).

Genetic variation between populations

The available material to initiate a selective breeding program or to be introduced into an existing program may include many breeds, strains or varieties, or in some cases, land races and natural populations. If such populations are, for geographic, ecological or human imposed reasons, genetically isolated, they will tend to accumulate different alleles as a consequence of mutation and the random process of genetic drift. Natural and artificial selection also will lead to differentiation if selective forces differ among populations, or to homogeneity if selective forces are similar. Thus both the levels of genetic variability and the actual alleles present may differ between populations.

Such genetic differences among populations are of concern for two reasons. Firstly, the genetic variance/covariance matrix for some set of traits of interest may differ between populations. This fact, or rather a failure to recognize it through extrapolation to other populations of estimates made in a single population, appears to be more a problem in evolutionary quantitative genetics (Barker, 1988; Barton and Turelli, 1989), than in plant and animal breeding, where estimation of genetic parameters should normally be an early step in any selection program.

Secondly, possible genetic differences among populations are important in the choice of populations for the exploitation of heterosis in crossbreeding programs, and in the choice of a base population for a selection program. For the former, heterosis is expected to be maximized for the cross where the two populations are most different genetically. However, empirical data must be used to decide which crossbred gives maximum performance, and this will be difficult to obtain where many possible populations are available. In the choice of a base population for a selection program, which population or populations will be best - for initial performance, and for maximizing genetic variability for future gains? If only a small number of potential base populations are available, it could be possible to evaluate all for performance and genetic parameters. More likely, however, there will be many potentially useful populations, and this will not be feasible.

One approach to these problems (Barker, 1980, 1985b) would be to determine the genetic relationships (or their inverse, genetic distances) among the populations. Genetic distance measures, based on allele frequencies at allozyme loci or molecular markers such as microsatellites, have been widely used in evolutionary genetic studies, to describe the genetic structure of populations or to determine evolutionary relationships among species. The allele frequency data obtained for each population also allows estimation of average heterozygosity, thus giving information on differences among populations in levels of genetic variability. Obviously such heterozygosities and genetic distances do not provide direct information on quantitative genetic differences between populations, but they do provide the best available objective information. Even if not used as a final arbiter in choice of populations for crossbreeding, genetic distances would allow objective choice of a restricted set of populations for further evaluation. In the choice of a base population for a selection program, distance measures should be used only as an initial guide, since distance measures cannot account for the consequences of artificial selection on economic traits, nor for natural selection on fitness. Final decisions must also take into account any available information on traits of economic importance, specific adaptive features and the presence of unique alleles or phenotypes. This approach should make a major contribution towards rationalization of the choice of strains or populations for utilization in breeding programs, and for conservation programs. In relation to domestic animals, a recent report to FAO (Barker et al., 1993) has recommended the establishment of a global program to estimate genetic distances among the breeds of each species.

Molecular genetics, major genes and breeding programs

The most likely distribution of gene effects for the loci controlling the expression of a quantitative trait (Quantitative Trait Loci or QTL) is one that is highly leptokurtic, and molecular genetic techniques are now providing the means to identify, and hence to exploit in breeding programs, the QTL of large effect (major genes or gene complexes within which linkage is not complete). Identification of such major genes allows the direct and immediate possibility of faster rates of genetic improvement. In addition, as the number of identified QTL for any particular trait increases, the developmental biology of the trait, or the pathways between genotype and phenotype, will be better understood, as will the extent of inter-locus interactions (Cheverud and Routman, 1995) and pleiotropy, thus providing a basis for improved quantitative genetic models, and further increasing the scope for manipulation of genetic variation in plant and animal improvement.

The basis of this approach is that it is now possible to identify large numbers of highly polymorphic molecular markers (such as Simple Sequence Repeat Polymorphisms, or variation in the length of microsatellites). The variation at such marker loci generally has no known or particular phenotypic effect, but using these loci, a saturated genetic map of the entire genome can be produced, with variable markers at least every 5-10 centi-Morgans. Given this density of marker loci, it is very likely that one or more markers will be in linkage disequilibrium with a QTL, resulting in a correlation between phenotypic value for a quantitative trait and marker genotypes.

Studies of agricultural plant species already have detected many QTL of moderate phenotypic effect (e.g. Doebley and Spec, 1991; Edwards et al., 1992, and references quoted therein), Bradshaw and Stettler (1995) have reported QTL with large effects on important traits in Populus, while Kinghorn et al. (1994) and Visscher and Haley (1995) have reviewed the identification and utilization of QTL in animal breeding.

In the future, the effect of this approach on breeding programs undoubtedly will be profound, but utilization of QTL will not replace current methods of quantitative genetics and breeding. The two are complementary, and much research is needed to ensure that they are integrated in an optimum fashion in designing and executing breeding programs.

REFERENCES

Barker, J.S.F. 1974. The state of information concerning deviations from additivity of gene effects. Proc. 1st Wld Cong. Genet. Appl. Livestock Production 1:373-383.

Barker, J.S.F. 1979. Inter-locus interactions. A review of experimental evidence. Theor. Pop. Biol. 16:323-346.

Barker, J.S.F. 1980. Animal genetic resources in Asia and Oceania - the perspective. Proc. SABRAO Workshop on Animal Genetic Resources in Asia and Oceania, pp.13-19. Tropical Agriculture Research Center, Japan.

Barker, J.S.F. 1985a. Potential contributions of genetics to animal production. Proc. 3rd AAAP Animal Science Cong. 1:22-32.

Barker, J.S.F. 1985b. Identifying the breeds to be evaluated. ACIAR Proceedings 5:161-166.

Barker, J.S.F. 1988. Quantitative genetics, ecology and evolution. In Proceedings of the Second international Conference on Quantitative Genetics, eds B.S. Weir, E.J. Eisen, M.M. Goodman and G. Namkoong, pp.596-600. Sinauer, Sunderland, Mass.

Barker, J.S.F. 1989. Population structure. In Evolution and Animal Breeding, eds W.G. Hill and T.F.C. Mackay, pp.75-80. CAB International, Wallingford.

Barker, J.S.F., Bradley, D.G., Fries, R., Hill, W.G., Nei, M. and Wayne, R.K. 1993. An integrated global programme to establish the genetic relationships among the breeds of each domestic animal species. Report of a Working Group, FAO Animal Production and Health Division, FAO, Rome, Italy. 32pp.

Barton, N.H. and Turelli, M. 1989. Evolutionary quantitative genetics : how little do we know? Annu. Rev. Genet. 23:337-370.

Bradford, G.E., Quirke, J.F., Sitorus, P., Inounu, I., Tiesnamurti, B., Bell, F.L., Fletcher, I.C. and Torell, D.T. 1986. Reproduction in Javanese sheep : evidence for a gene with large effect on ovulation rate and litter size. J. Anim. Sci. 63:418-431.

Bradshaw, H.D. Jr. and Stettler, R.F. 1995. Molecular genetics of growth and development in Populus. IV. Mapping QTLs with large effects on growth, form and phenology traits in a forest tree. Genetics 139:963-973.

Cheverud, J.M. and Routman, E.J. 1995. Epistasis and its contribution to genetic variance components. Genetics 139:1455-1461.

Cockett, N.E., Jackson, S.P., Shay, T.L., Nielsen, D., Moore, S.S., Steele, M.R., Barendse, W., Green, R.D. and Georges, M. 1994. Chromosomal localization of the callipyge gene in sheep (Ovis aries) using bovine DNA markers. Proc. Natl. Acad. Sci. USA 91:3019-3023.

Crow, J.F. and Kimura, M. 1970. An Introduction to Population Genetics Theory. Harper and Row, New York.

Davis, G.H., McEwan, J.C., Fennessy, P.F., Dodds, K.G. and Farquhar, P.A. 1991. Evidence for the presence of a major gene influencing ovulation rate on the X chromosome of sheep. Biology of Reproduction 44:620-624.

Doebley, J. and Stec, A. 1991. Genetic analysis of the morphological differences between maize and teosinte. Genetics 129:285-295.

Edwards, M.D., Helentjaris, T., Wright, S. and Stuber, C.W. 1992. Molecular-marker-facilitated investigations of quantitative tait loci in maize 4. Analysis based on genome saturation with isozyme and restriction fragment length polymorphism markers. Theor. Appl. Genet. 83:765-774.

Eisen, E.J. 1975. Population size and selection intensity effects on long term selection response in mice. Genetics 79:305-323.

Falconer, D.S. 1989. Introduction to Quantitative Genetics. 3rd ed. Longman, Harlow, Essex.

Fisher, R.A. 1918. The correlation between relatives on the supposition of Mendelian inheritance. Trans. R. Soc. Edinburgh 52:399-433.

Frankham, R. 1980a. Origin of genetic variation in selection lines. In Selection Experiments in Laboratory and Domestic Animals, ed. A. Robertson, pp.56-68. Commonwealth Agricultural Bureaux, Farnham Royal, U.K.

Frankham, R. 1980b. The founder effect and response to artificial selection in Drosophila. In Selection Experiments in Laboratory and Domestic Animals, ed. A. Robertson, pp.87-90. Commonwealth Agricultural Bureaux, Farnham Royal, U.K.

Franklin, I.R. 1982. Population size and the genetic improvement of animals. In Future Developments in the Genetic Improvement of Animals, eds J.S.F. Barker, K. Hammond and A.E. McClintock, pp.181-196. Academic Press Australia, Sydney.

García, N., López-Fanjul, C. and García-Dorado, A. 1994. The genetics of viability in Drosophila melanogaster : effects of inbreeding and artificial selection. Evolution 48:1277-1285.

Guillaume, J. 1976. The dwarfing gene dw : its effects on anatomy, physiology, nutrition, mangement. Its application in poultry industry. World's Poult. Sci. J. 32:285-304.

Hanset, R. 1982. Major genes in animal production, examples and perspectives : cattle and pigs. Proc. 2nd Wld Cong. Genet. Appl. Livestock Prod. 6:439-453.

Hill, W.G. 1982. Predictions of response to artificial selection from new mutations. Genet. Res. Camb. 40:255-278.

Hill, W.G. 1985. Effects of population size on response to short and long term selection. Z. Tierzüchtg. Züchtgsbiol. 102:161-173.

Hill, W.G. 1989. Sewall Wright and quantitative genetics. Genome 31:190-195.

Hill, W.G. 1990. Mutation, selection and quantitative genetic variation. In Population Biology of Genes and Molecules, eds N. Takahata and J.F. Crow, pp.219-232. Baifukan, Japan.

Hill, W.G. and Caballero, A. 1992. Artificial selection experiments. Annu. Rev. Ecol. Syst. 23:287-310.

Hill, W.G. and Keightley, P.D. 1988. Interrelations of mutation, population size, artificial and natural selection. In Proceedings of the Second International Conference on Quantitative Genetics, eds B.S. Weir, E.J. Eisen, M.M. Goodman and G. Namkoong, pp.57-70. Sinauer, Sunderland, Mass.

Hohenboken, W.D. 1982. Beef cattle breeding à la Jefferson. J. Anim. Sci. 54:667-679.

Hollingdale, B. 1971. Analyses of some genes from abdominal bristle number selection lines in Drosophila melanogaster. Theor. Appl. Genet. 41:292-301.

Hospital, F. and Chevalet, C. 1993. Effects of population size and linkage on optimal selection intensity. Theor. Appl. Genet. 86:775-780.

James, J.W. 1971. The founder effect and response to artificial selection. Genet. Res. Camb. 16:241-250.

Jones, L.P., Frankham, R. and Barker, J.S.F. 1968. The effects of population size and selection intensity in selection for a quantitative character in Drosophila II. Long term response to selection. Genet. Res. Camb. 12:249-266.

Keightley, P.D. and Hill, W.G. 1990. Variation maintained in quantitative traits with mutation-selection balance : pleiotropic side-effects on fitness traits. Proc. R. Soc. Lond. B 242:95-100.

Kinghorn, B.P. 1980. The expression of "recombination loss" in quantitative traits. Z. Tierzüchtg. Zuchtüngsbiol. 97:138-143.

Kinghorn, B., van Arendonk, J. and Hetzel, J. 1994. Detection and use of major genes in animal breeding. AgBiotech News and Information 6:297N-302N.

Lai, C., Lyman, R.F., Long, A.D., Langley, C.H. and Mackay, T.F.C. 1994. Naturally occurring variation in bristle number and DNA polymorphisms at the scabrous locus of Drosophila melanogaster. Science 266:1697-1702.

Latter, B.D.H. and Robertson, A. 1962. The effects of inbreeding and artificial selection on reproductive fitness. Genet. Res. Camb. 3:110-138.

López-Fanjul, C. 1989. Tests of theory by selection experiments. In Evolution and Animal Breeding, eds W.G. Hill and T.F.C. Mackay, pp.129-133. CAB International, Wallingford, U.K.

Lush, J.L. 1945. Animal Breeding Plans. 3rd ed. Iowa State University Press, Ames, Iowa.

Mackay, T.F.C. 1990. Distribution of effects of new mutations affecting quantitative traits. Proc. 4th Wld Cong. Genet. Appl. Livestock Prod. 13:219-228.

Mackay, T.F.C. and Langley, C.H. 1990. Molecular and phenotypic variation in the achaete-scute region of Drosophila melanogaster. Nature 348:64-66.

Madalena, F.E. and Hill, W.G. 1972. Population structure in artificial selection programmes : simulation studies. Genet. Res. Camb. 20:75-99.

Mather, K. and Jinks, J.L. 1982. Biometrical Genetics. 3rd ed. Chapman and Hall, London.

Moore, F.B.-G. and Tonsor, S.J. 1994. A simulation of Wright's shifting-balance process : Migration and the three phases. Evolution 48:69-80.

Moreno, G. 1994. Genetic architecture, genetic behaviour, and character evolution. Annu. Rev. Ecol. Syst. 25:31-44.

Piper, L.R. and Bindon, B.M. 1982. Genetic segregation for fecundity in Booroola Merino Sheep. Proc. 1st Wld Cong. Sheep and Beef Cattle Breeding 1:395-400.

Roberts, R.C. 1966. The limits to artificial selection for body weight in the mouse II. The genetic nature of the limits. Genet. Res. Camb. 8:361-375.

Robertson, A. 1960. A theory of limits in artificial selection. Proc. Roy. Soc. Lond. B. 153:234-249.

Robertson, A. 1961. Inbreeding in artificial selection programmes. Genet. Res. Camb. 2:189-194.

Robertson, A. 1967. The nature of quantitative genetic variation. In Heritage from Mendel, ed. R.A. Brink, pp.265-280. Univ. Wisconsin Press, Madison.

Sheridan, A.K. 1988. Agreement between estimated and realised genetic parameters. Anim. Breed. Abst. 56:877-889.

Shrimpton, A.E. and Robertson, A. 1988a. The isolation of polygenic factors controlling bristle score in Drosophila melanogaster. I. Allocation of third chromosome sternopleural bristle effects to chromosome sections. Genetics 118:437-443.

Shrimpton, A.E. and Robertson, A. 1988b. The isolation of polygenic factors controlling bristle score in Drosophila melanogaster. II. Distribution of third chromosome bristle effects within chromosome sections. Genetics 118:445-459.

Smith, C. and Quinton, M. 1993. The effect of selection in sublines and crossing on genetic response and inbreeding. J. Anim. Sci. 71:2631-2638.

Turelli, M. and Barton, N.H. 1994. Genetic and statistical analyses of strong selection on polygenic traits : What, me normal? Genetics 138:913-941.

Visscher, P.M. and Haley, C.S. 1995. Utilizing genetic markers in pig breeding programmes. Anim. Breed. Abst. 63:1-8.

Webb, A.J., Carden, A.E., Smith, C. and Imlah, P. 1982. Porcine stress syndrome in pig breeding. Proc. 2nd Wld Cong. Genet. Appl. Livestock Prod. 5:588-608.

Weber, K.E. 1990a. Increased selection response in larger populations. I. Selection for wing-tip height in Drosophila melanogaster at three population sizes. Genetics 125:579-584.

Weber, K.E. 1990b. Increased selection response in larger populations. II. Selection for ethanol vapor resistance in Drosophila melanogaster at two population sizes. Genetics 125:585-597.

Wright, S. 1922. The effects of inbreeding and crossbreeding in guinea pigs. III. Crosses between highly inbred families. USDA Bull 1121:1-60.

Wright, S. 1977. Evolution and the Genetics of Populations, Vol.3. University of Chicago Press, Chicago.

Yoo, B.H. 1980a. Long term selection for a quantitative character in large replicate populations of Drosophila melanogaster II. Lethals and visible mutants with large effects. Genet. Res. Camb. 35:19-31.

Yoo, B.H. 1980b. Long term selection for a quantitative character in large replicate populations of Drosophila melanogaster Part 3: The nature of residual genetic variability. Theor. Appl. Genet. 57:25-32.

MANAGING THE GENETIC VARIANCE SIMULTANEOUSLY FOR, HIGHER GAIN AND ADAPTIVE POTENTIAL

Gene Namkoong and Mathew P. Koshy

Department of Forest Sciences
University of British Columbia

One of the central dilemmas of plant and animal breeding has been that using the genetic variation has implied its destruction. This dilemma arises because the endowed state is assumed to be a unique and optimal condition that would be destroyed if it is changed or used. One of the paradigms of breeding agricultural crops is the achievement of an optimal state in either pure lines or hybrids of pure lines. In this paradigm the presently available varieties approximate an optimal productivity and new alleles from unimproved or wild relatives infused via backcrossing schemes are needed only to fine tune the productive varieties. Then, an emerging problem in breeding is that the naturally evolved variation is subject to erosion. Unless the environment or genetic system changes and different allelic effects are generated, lack of genetic variation will be the ultimate end either due to fixation or loss of alleles. Preservation of larger number of parents is often seen as a solution for this problem. But, the larger the number of parents, the lower is the selection differential and hence the lower is the gain from selection and in animal breeding theory, a selection proportion of about Q is optimal for maximum long term gain to be achieved (Robertson, 1960).

A different paradigm can be derived from a more dynamic view of the genetic resource. In both natural as well as artificial breeding systems, it is recognized that populations also affect their own environment and hence that both exogenous and autogenous sources of effects can continually generate changes in selection forces. Environmental changes are thus both a cause and an effect of evolution. In addition to environmental change as both cause and effect of evolution, the genetic system itself undergoes evolution in that as selection changes the physiology of plants, the effects of alleles on that physiology also change and hence an induced kind of epistasis and pleiotropy exist. Mutation rates on a per trait basis can also be quite high, and hence new allelic effects are constantly being added to populations even as some alleles go to fixation. As revealed by long term selection experiments, a steady state genetic variance can be created which is a resultant of the effects of selection, population size, and mutation rates (Hill and Keightley, 1988). We must consider that just as evolution has changed it means, it has also changed variances. Evolution is not driven by selection to an optimum fixed allelic state even in the absence of environmental change. There is thus, a dynamic to the genetic variance, some factors having an increasing and others a decreasing effect on the total variance in both natural as well as artificially bred populations. The genetic variance is thus an emergent property of genetic dynamics, subject to selection, mutation, and population size and is both a cause and effect of evolution.

One of the features of the genetic system is a population structure that limits complete or random intermating. In the absence of subdivisions of the mating population, variations in selection over time or geographic variables have relatively minor effect on maintaining patterns of genetic polymorphisms. Similarly, in the absence of selection differences among populations, small migration rates can ensure that alleles are widely distributed among all populations such that the probability of any allele frequency occurring in a population is as likely as any other. However, with selection differences confounded by restrictions in migration rates between populations, patterned polymorphisms can be readily maintained. Even if only seed migration is limited between selectively divergent stands but pollen is unrestricted, polymorphisms can exist where none could with complete random mating (Gregorius and Namkoong 1983; Namkoong and Gregorius, 1985), thus, random mating (Gregorius and Namkoong 1983; Namkoong and Gregorius, 1985), thus, the interplay between migration and selection can have effects that either alone could not produce.

As users and creators of genetic variation, breeders have several optional ways of influencing the size and distribution of genetic variance through both selection and controlled mating. In this paper, we will simplify environmental variables and will instead focus primarily on the genetic system itself and the breederis capacity to influence family and population structure to affect genetic variance while maintaining higher genetic gain.

GENETIC VARIANCE WITHIN POPULATIONS

Evolution of reasonably large populations under relatively constant environmental conditions may be expected to have often produced phenotypes that behave close to an average optimum. Selection may then be reasonably modeled as stabilizing selection for an intermediate optimum and aside from the occasional heterotic locus or frequency dependent selection for rare alleles, genetic variance is not expected to be maintained. In the long run, only mutation would be strong enough to maintain variation and several models have been investigated that indicate that high levels of genetic variance can be maintained despite linkage and unequal allelic effects. The various models put forward can be grouped into two distinct types with n additive polygenic loci subject to mutation and stabilizing selection. The model put forward by Lande (1976) and Fleming (1979) was an extension of the model described by Kimura (1965) which assumes a continuum of allelic effect at each locus with moderate frequency, while the model considered by Latter (1960) and Bulmer (1972, 1980) considers diallelic loci. Qualitative analysis and the solution to the former model demonstrate an equilibrium solution for genetic variance for given mutation rates and selection intensities which do not depend on the rate of recombination between the loci in the genome while the number of recombining loci has a strong influence on the amount of expressed genetic variability. The latter model shows that the equilibrium genetic variance is a function only of the mutation rate at that locus, being independent of the magnitude of the average effect of the gene concerned. In their analysis recurrent mutation, under natural selection favoring phenotypic intermediates could not account for the genetic variance commonly found in natural populations. Hence heterozygote superiority in fitness was suggested as a probable mechanism for conserving genetic variability. Later analysis of these models by Turelli (1984) has shown that the qualitatively different predictions of equilibrium genetic variance under the two above models are not due to the number of alleles assumed at a locus but are due to differing assumptions on relative magnitude of per locus mutation rates, the phenotypic mutation effects and the intensity of selection. The model described by Turelli (1984) based on the ehouse-of-cardsi model put forward by Kingman (1978) describes mutation-selection equilibrium for allelic effects under continuum of allele model of Kimura (1965) more accurately. Moreover, the model predictions also agree with the diabetic model by Latter (1960) and Bulmer (19?2, 1980). The genetic variance predicted under this model as well as under diabetic model of Latter (1960), and Bulmer (1972, 1980) is 4VsU where Vs is inverse of selection intensity, and U= (ui and u is per locus mutation rate. If a larger number of loci is involved, Barton (1986) has shown that additional stable equilibria with substantially larger variances can occur depending upon the history of the populations. The many stable equilibria, each giving a slight deviation from the optimum, coexisting in the population is possible since many different combinations of alleles could satisfy the optimum imposed by selection. Based on the ehouse-of-cardsi models of Turelli, with a biallelic locus and many loci involved in a trait, Barton (1986, 1989) has shown that the fitness surface may contain many local adaptive peaks. Various forces like sampling drift or random fluctuations in selection pressure can knock populations between these peaks. However, sampling drift and variation in allelic drift together may keep the population close to the optimum derived by Turelli (1984) and it should be a good predictor of the equilibrium genetic variance under mutation-selection balance in the case of a diabetic loci (Bulmer, 1989). These kinds of shifts between alternative equilibria may not have significant effects in the genetic variance within the populations. However, in the long run, it can lead to substantial reproductive barriers and speciation. In the short run, it may have significant implications on the management of genetic variance.

While the previous two models assume that selection affects the quantitative trait directly, the model suggested by Hill and Keightly (1988), and Barton (1990) considers pleiotropic effects. In these models, each locus affecting overall fitness is at an equilibrium under selection against alleles that lower fitness and mutation to those alleles. Several other factors other than mutation-selection balance may also be involved in maintenance of genetic variability. Slight heterozygote superiority, frequency-dependent selection due to competition between individuals with similar phenotypes, and selection for different optima in different populations with mating barriers etc. are some of them (Bulmer, 1989).

Continuous directional selection, even without any fluctuations can also result in eventually achieving a steady state genetic variance (Hill, 1982; Hill and Keightly, 1987; Hill and Rashbash, 1986; Keightley and Hill, 1988). When mutant effects are symmetrically distributed and population sizes are small, the variance maintained by directional selection and stabilizing selection are almost the same and it can be about half the level of the genetic variance in the original population as long as population sizes are kept above a few tens of monoecious individuals. However, at larger population sizes, while stabilizing selection reaches a limit of genetic variance, directional selection has no bounds for the variance maintained though natural selection may interfere to reduce this bound. This apparent difference may be due to the reproductive success of the positive mutants in the case of directional selection while in the case of stabilizing selection, both negative and positive effects on the traits decrease the reproductive success (Hill and Keightley, 1987). The early response to selection is dependent on the genetic variance that is residual in the initial populations. But with populations of only a few tens in size, most of the additive effect alleles would be fixed in the first ten or twenty generations. The continuous changes observed in long term selection experiments (Enfield, 1980; Yoo, 1980; Dudley, 1977) are therefore considered to be due to mutations. The continued response in pupal weight in Tribolium after 120 generations of selection (Enfield, 1980) and increase in oil content in maize after 76 generations (Dudley, 1977) may be due to new mutations generated during the course of the experiment though epistatic and pleiotropic effects can contribute. Therefore, the effects of directional selection is indicated to be no greater than the effects of stabilizing selection on the level of genetic variance and since genetic variance persists, continuous change in the population mean can be obtained.

GENETIC VARLANCE BETWEEN POPULATIONS

Since natural population subdivisions exist naturally to some extent, and subdivisions are constructed in breeding populations between base populations and among developed varieties, the variance between populations may reach an equilibrium, may steadily increase, or may perpetually experience periods of increase and decrease. If there is some continuous inter-population gametic migration, the level of the total genetic variance will depend on the divergence and directionality of selection as well as on the migration rate (Bulmer, 1980). The analyses of genetic variance are more complex due to the effects that different forms selection can take between populations and whether migration effects are confounded with selection differences. Selection can be stabilizing or directional and either type may be divergent among populations or identical for all. Selection among different populations may also be convergent and again, may be either stabilizing or directional. Continuous geographic clinal models have been described by Felsenstein (1977) and in more detail by Slatkin (1978), but most of these cases have not been thoroughly investigated theoretically or experimentally, but a few conjectures can be reasonable to draw. Clearly, one of the significant problems in theoretical population genetics remains in determining how the various forces of population size, selection, and migration, can operate to produce the changes or the equilibria in genetic variances and covariances within and among populations. The variance is not fixed either within nor certainly between populations, and hence to total genetic variance remains a variable factor that itself is subject to various evolutionary pressures.

MANAGING THE GENETIC VARIANCE

All of the forces of evolution are subject to management with the possible exception of the mutation rate although that too may soon be manageable. Breeders have long worked with fairly small population sizes and have been aware of its limiting effects when exclusionary practices are pursued for advanced varietal development. Therefore, considerable attention has been paid to population sizes and to the size of seed collections that intend to conserve genes. The effect of sample size on the probability of saving random alleles is known to be logarithmic and hence, after a few tens of samples, the sample sizes required to affect conservation increase exponentially (Namkoong, 1988). The problem this causes for breeders is that selection intensity is a function of the proportion selected and hence requires that breeding populations carry relatively few parents per generation. Hence there is a conflict between using the genetic variance in breeding populations, and using it up if the only way we manage genetic variance is by population size alone.

Aside from affecting the mutation rate, the two remaining factors that breeders can influence are selection and migration. Breeders generally use closed populations to develop advanced varieties but various forms of hybridization and backcrossing are regularly used, first to increase the genetic variance and then to fix certain allele combinations. However, long term selection experiments within manageable breeding population sizes, demonstrate that genetic divergence can accumulate and contribute to a persistence of genetic variance and hence that selection between populations can readily increase the between population component of the total genetic variance. Since common breeding practices can generate large differences between base populations and selected varieties in very few generations, and if the total genetic variance is proportional to the mean difference between populations or its square, the management of genetic variance can be powerfully influenced by the kinds of population divergence that breeders select. Thus, managing the between population component of genetic variance can be a very powerful tool for breeders to use.

The genetic variance between selectively diverged populations would be first dependent on the residual genetic variance in the common base population from which they originally diverge. Considering first one selected population and one population that could be maintained without selection, the fixation of favorable alleles would eventually eliminate all useful genetic variance. However, with high mutation rates on a per trait basis, allelic variants can be generated and fixed at different loci and can create a mutation - selection balance in the genetic variance while those alleles are still in a transient state in the population being selected. If we consider that in the unselected population, there would be no selection pressure on those alleles, both positive and negative effect alleles would be selectively neutral. With independent mutations in non-interbreeding populations, the loci are independent, and the genetic variance would be the sum of the variances at the multiple loci. With variance between populations, there is a genetic variance that can be created if the populations are hybridized and free recombination in the F2 or later generations is realized. The increased variance realized in the hybrid population has been termed the Segregation variance? Lande (1981).

For populations that have been allowed to drift, but with the same stabilizing selection, the levels of genetic variance within each of the base populations may remain same but the magnitude of segregation variance will depend upon the kind of genetic variance underlying a quantitative character provided by mutation. When populations at different adaptive peaks on the fitness surface are crossed and F2 generated, it will produce higher levels of genetic variance. The various factors which influence the shift in peaks and its rate are of high significance and need further scientific elucidation. If variation is maintained by numerous alleles of moderate or small effect, segregation variance will increase linearly with time of separation. Because selection imposed only a single constraint on the mean of the character, most of the alleles at the underlying loci are free to drift. When variation is maintained by low frequency alleles, the increase in segregation variance is likely to be small (Slatkin and Lande, 1994). However, if the two populations have stabilizing selection but for different optima, the effects on F2 genetic variance are difficult to analyze.

We can trace how different models of mutation and gene effects contribute to this variance and can show that certain differences in effects emerge in the relative sizes of the variance in hybrid populations derived from no selection, selection with replicate populations, and divergent selection. If multiple alleles can exist at each locus, then divergent selection will increase the realized segregation variation over that of replicate population hybrids and both will be larger than the variance in the base population. Similarly, Koshy et al. (1995), has shown that with backward and forward mutations at a single locus, the genetic segregation variance of divergently selected populations will be larger than from hybrids of replicate populations. Interestingly however, Roberds et al. (1995), have shown that if the loci segregating are different in the divergent populations and only two alleles per locus can exist, then the segregation variance would be identical for divergent as for replicate population selection systems. We conjecture that if mutation is of Turelliis ehouse-of-cardsi form, that the difference in genetic variance between divergent and replicate population hybrids will be less than if Landers multivariate normal approximation were valid. In all cases, the genetic variance is maintained even though selection continues, and presumably the variance would continue to increase until some form of outbreeding depression is incurred, and non-additive gene actions dominate the response to selection.

GENETIC MANAGEMENT OPTIONS

With the capacity to alter levels of genetic variance by controlling between population as well as within population variances, it is also possible to control the genetic covariances by other means than only selecting within single populations to break correlations. The whole matrix of variances and covariances can be managed by breeders and breeders can choose to deploy different levels of genetic variance as well as to use them for breeding advance. Several new research questions are now generated by this option. In some cases, increased variance is desirable because the array of ecological or economic need are wider than can be accommodated well by present levels of variance. For example, when climate change may require higher levels of tolerance to climate extremes or temporal variations, it may be advantageous to have higher variances deployed and to have wider variances available among different breeding populations.

Genetic management options are also increased by multiple population management in a manner analogous to searching for multiple adaptive peaks in natural selection. It is well known that several populations of small size can more completely search an adaptive surface for multiple peaks of adaptation than can a single population of any size. The existence of multiple peaks is dependent on certain kinds of fitness epistasis in natural population evolution, but this can be mimicked in breeding for non-linear economic values when value is a function of several component traits. Even if gene actions are independent, a multiple trait value function can create allelic ifitnessl values such that multiple epistatic value peaks exist. To explore such complex surfaces by breeding, multiple populations can use different gene and trait combinations to ultimately achieve higher values than possible wlth single population.

In forestry, where improvement almost always involves a complex of multiple traits (Namkoong, 1976), a total value epistasis can be generally expected even if individual trait gene actions are largely additive between loci. Just as non-additivity between traits can cause an interaction in a composite value function (Namkoong, 1985), the non-additivity of growth, resistance, and wood quality can create a complex value surface for gene effects. Multiple populations are therefore useful for increasing ultimate values as well as for increasing genetic variances. Simultaneous improvement of the populations in the direction of the interest of the breeders and maintenance of higher levels of potential for genetic variance are thus simultaneoulsy possible by this strategy. We suggest that, by managing genetic variance, breeders will have many more options for improving the value and adaptability of future forest than we can now dream of.

LITERATURE CITED

Barton, N. H. 1986. The maintenance of polygenic variation through a balance between mutation and stabilizing selection. Genet Res. 47:209-216.

Barton, N. H. 1989. The divergence of a polygenic system subject to stabilizing selection, mutation and drift. Genet. Res. 54:59-77.

Barton, N. H. 1990. Pleiotropic models of quantitative variation. Genetics 124:773-782.

Bulmer, M. G. 1972. The genetic variability of polygenic characters under optimizing selection, mutation and drift. Genet Res. 19: 17-25.

Bulmer, M. G. 1980. The Mathematical Theory of Quantitative Genetics. Clarendon Press, Oxford.

Bulmer, M. G. 1989. Maintenance of genetic variability by mutation-selection balance: a childis guide through the jungle. Genome 31 :761-767.

Dudley, J. W. 1977. 76 generations of selection for oil and protein percentage in Maize. pp. 459-473. In Proceedings of the International Conference on quantitative Genetics. Eds. E. Pollak, O. Kempthorne, and T. B. Bailey Jr., Iowa State University Press, Ames, Iowa.

Enfield, F. D. 1980. Long term effects of selection; the limits to response. pp. 69-86. In Selection Experiments in Laboratory and Domestic Animals. Ed A. Robertson, CAB Slough.

Felsenstein, J. 1977. Multivariate normal genetic models with a finite number of loci. pp.227-46. I¢ Proceedings of the International Conference on Quantitative Genetics. Eds. Eds. E. Pollak, O. Kempthorne, and T. B. Bailey Jr., Iowa State University Press, Ames, Iowa.

Robertson, A. 1960. A theory of limits in artificial selection. Proc. R. Soc. B153, 234249.

Slatkin, M. 1978. Spatial patterns in the distribution of polygenic characters. J. Theor. Biol. 70:213-228.

Slatkin, M. and Lande, R. 1994. Segregation variance after hybridization of isolated populations. Genet. Res 64:51-56.

Turelli, M. 1984. Heritable genetic variation via mutation-selection balance: Lerchis zeta meets the abdominal bristle. Theor. Pop. Biol. 25:138-193.

Yoo, B. H. 1980. Long term selection for a quantitative character in large replicate populations of Drosophila melanogaster. Genet Res 35: 1-17.

FUTURE DIRECTIONS IN TREE BREEDING: SOME QUESTIONS OF WHAT WE SHOULD SEEK, AND HOW TO MANAGE THE GENETIC RESOURCE

Rowland D. Burdon

Forest Research Institute
Private Bag 3020
Rotorua, New Zealand

ABSTRACT

Forest tree breeding has delivered major genetic gains, despite various early setbacks, and increasing areas are now being established or reforested with genetically improved stock. These very successes bring some problems. As the easily-achieved genetic gains become captured, thought must be given to the types of genetic gain to be sought in the future, and to genetic improvement of species which, while they must be used, have hitherto been regarded as less tractable. Genetic gains in productivity have so far derived from selecting individuals that are outstanding competitors, quite unlike gains usually made in crop breeding. Longer-term gains in forest trees may well come from exploiting the divergences between competitive ability and crop efficiency, which could strongly favour clonal forestry systems. Some species that have traditionally been regarded as climax species and, as such, less promising for domestication, now show signs of exceptional genetic variability for early growth, but these prospects for genetic gain may be tempered by the need to tackle some adaptive problems.

The prospect of breeding for improved wood properties is, in some quarters, attracting a new upsurge of interest. Certain processing characteristics can show remarkable genetic variation, but the costs of evaluating candidates for such properties pose some problems which although formidable are fascinating and not necessarily insuperable.

The levels of domestication already achieved are placing new demands on genetic management. While great advances have been made in covering a diversity of breeding goals, the appropriate stance to take towards inbreeding is still debatable. Inbreeding has strong theoretical attractions as a breeding tool, but in few tree species does it seem practicable to use it very aggressively. Maintenance of gene resources, which underpin pedigreed breeding populations, is widely favoured, partly incidental to ecosystem conservation for *in situ* resources, and partly as a backup for tree breeders' needs. No satisfactory substitute is seen for naturally occurring genetic variation. Management of such gene resources, both *in situ* and *ex situ*, can pose considerable problems, especially with wind-pollinated species that become massively domesticated. No cheap and really attractive solution is in sight, but it is to be hoped that with better knowledge and a sufficient commitment to the species that appropriate solutions can be afforded. In this, public opinion may be helpful. A very tantalising question is the extent to which mutation can be relied on to replenish or augment genetic variation within populations that are not very large.

INTRODUCTION

I thank the organisers most sincerely for the invitation to come. This caps a fascinating visit. While I am honoured to be speaking now, for what input I can make, I fear that the learning process has been a little one-sided, in my favour.

I will begin with a few reflections on where forest tree breeding stands in relation to other plant breeding and to animal breeding, in respect of its inherent nature and the stage of its development. I will then review briefly some developments that reflect the increasing maturity of forest tree breeding. Then I will review in greater depth some of the more complex issues that tree breeders will need to face, in two areas:

- Where we should look for future genetic gains
- How should we manage genetic material, in relation to what has just been said (Namkoong & Khoshy 1995)

Despite some past forays into inbreeding, forest tree breeding has generally been dominated by the outbreeding behaviour of most trees. That sets it apart from much of plant breeding which has been based on inbreeding which produces the uniformity in stabilised cultivar varieties. In general, tree breeding has tended to link more closely with animal breeding than with most other plant breeding. There were some unfortunate early borrowings of mating designs from animal breeding. The salient features, however, that link the two disciplines have been: the essentially obligate outbreeding; and the resources that often have to be committed per individual, yet the needs for large bodies of data to acquire the requisite genetic information, and for recording and analysing data on individuals that are necessarily scattered in space if not also in time. Detailed constraints differ; for instance, animals have typically much shorter generation times, but they have other important reproductive constraints and very finite life spans. Early evaluation is a greater issue with forest trees, but animals pose the problem of sex-limited traits.

Vegetative propagation is often an option with forest trees. Moreover, with the geographic distribution of many tree species and the clear importance of genecology we may have inherently a better framework for predicting hybrid performance. We can often identify complentarities between populations in site tolerances. Given sites where the complementarities are required, and the tolerances being inherited favourably enough in hybrid combination, hybrids can show both superior performance on such sites and broad site tolerances.

Animal breeding has, along with some other areas of plant breeding, generated much of the quantitative methodology, it has contributed much to the teaching of tree breeders, and has even produced a number of them. We are therefore very fortunate to have here such a distinguished figure from animal breeding as Stuart Barker. Yet forest tree breeding, in the wider sense, has made some contributions of its own. Along with other plant breeding it has made its own contributions to studying genotype-environment interaction and, very recently, the presence of the haploid megagametophyte in conifers has been exploited to spectacular advantage in genomic mapping. Much less formally, it is contributing to some lively behind-the-scenes debate on requisite population sizes for long-term breeding. May the intercommunication continue and increase.

For most forest trees, selective breeding gives significant gains in growth rate, or in the standard of tree form, or in resistance to pests or diseases, if not in all three. Some of the gains have come very easily, turning attention to what future gains are readily obtainable, whether different types of genetic gain should be sought in future, and how they should be sought.

Various setbacks have admittedly occurred. Some well-executed programmes have come to nothing because they involved the wrong provenance base, or even the wrong species. Early on, hybridisation was sometimes pursued without due attention to choice of parents within species, or a critical consideration of what hybridisation really stood to achieve. Maturation effects made many early clonal trials or 'tree shows' misleading. Pollen contamination, delayed graft incompatibility, and various other problems seriously held up the delivery of genetic gain from many early seed orchards. Using too narrow a genetic base, in premature searches for near-perfect parents, did not help either.

Nevertheless, there have been major achievements. In many places, genetically improved stock is meeting much if not all of the planting requirements, often significantly beyond the first generation of improvement.

In managing genetic material to assure continued genetic gain and flexibility, great progress has been made. Yet the question whether or how inbreeding should best be coped with, or even aggressively pursued in breeding populations, warrants further attention. At another level, the management of the back-up genetic resources, which are held in reserve for the very long term and the greatly unexpected, can be much complicated by the sheer success of a breeding programme. The need for such back-up resources therefore warrants critical examination, particularly in the light of what mutation may have to offer.

My coverage will be selective, largely because I want to focus on some problems, or areas of special interest. Much of what I say will not be new or really my own. Hopefully, it will acquire value from its context.

DIRECTIONS FOR FUTURE GENETIC GAINS

Crop Productivity

Gains in growth rate are almost always sought by tree breeders, and are generally made. Faster early growth can accelerate site occupation in a plantation, and thereby increase mean annual increment (m.a.i.), while it can give desired log sizes earlier which is also economically attractive. Yet, provided a dense canopy is maintained, differences in m.a.i. can be modest compared with differences in individual-tree growth rates, and differences in current annual increments can be much less still -- slower-growing material can, at a given age, have much the same per-hectare volume production, but spread over more but smaller stems. This reflects what may well be some fairly fundamental limits to the photosynthetic capacity of a foliage canopy.

It is instructive to consider how most gains in productivity have been made in crop plant breeding. Two main points must be considered. Firstly, most of the dramatic gains have occurred in harvest index, the proportion of biomass that can be used (Evans 1993). In cereals, for instance, grain yield has increased with little increase in total dry matter production, given comparable moisture and soil fertility. In forest trees, where the stem represents such a high proportion of biomass, the scope seems much less. Areas for potential improvement include (1) reduced reproductive activity, although in some species this is largely suppressed by intense competition, and, speculatively, (2) more efficient turnover patterns in foliage. Big genotypic differences may exist among young trees in stemwood/branchwood ratios (St Clair 1984), but these differences can narrow with age, while the early allocation patterns may be significant more as investment strategies than in their own right; a genotypically determined pattern that is desirable in a mature tree may not be what is needed for fast initial growth or early site occupation, and *vice versa*.

Secondly, we should note in selecting for growth rate in forest trees we almost invariably select for effective competitors within in a species (Burdon 1982, Libby 1987). This contrasts with crop breeding, where the cultivars are typically poor competitors, albeit with highly desirable biomass allocation, their success depending on being isolated from competition from other than peer genotypes. For longer-term genetic gains in production in forest trees we will probably need to look increasingly to exploiting divergences between crop productivity and competitive ability. It would certainly push us towards using clonal blocks, unless we can assemble mixes of clones or even families of similar competitive ability. Tests of clonal blocks would be needed to help define the appropriate ideotypes for achieving this goal -- while we may theoretically model an ideotype, experimental verification will be needed, because with the complex and non-linear functions involved the cumulative effects of errors of estimating the individual parameters are likely to be major. Empirical screening of a large array of candidates for final per-hectare production is obviously prohibitive, which makes ideotype definition paramount. Early evaluation of new genotype, however, may be particularly difficult in this respect.

Clonal blocks will have some other advantages as a tool for research and selection, notably screening for resistance to wind or snow damage, which are often features more of stands than of individual trees.

Gain Projections

Genetic gains in growth rate are typically evaluated as single-tree data quite early in the rotation. Various workers are now addressing the problem of translating those gains into final whole-crop production gains (*see* Carson & García, in MS). That has obvious advantages for demonstrating payoffs from genetic improvement, and for yield projection and forest planning when genetically improved stock is used. Of greater concern for the tree breeder, however, is that this is a prerequisite for attaching an economic weight to the measured genotypic gains in growth, and that the trade-off pervasive between growth and wood density needs to be quantified at the level of the crop. It is where negative or adverse genetic correlations exist that it becomes crucial to determine both the strength of the tradeoffs and the comparative economic weights of the traits involved.

Shifts in Successional Status?

Commercial forestry is increasingly becoming plantation forestry, which is necessarily growing trees as pioneers. Species that are naturally pioneers typically have rapid early growth, which is commercially attractive. Yet climax species often show very high current annual increments after slow starts. The biggest successes with plantation forestry have often been with species that are not just pioneers, but carry through to an effective climax status under regimes of infrequent disturbances, e.g. fire. Examples include *Pinus radiata*, Douglas-fir, and various "wet sclerophyll" eucalypts. What is attractive is the prospect of growing climax species in plantations, if they can be persuaded to make rapid early growth, which would have many advantages over using such species as final-crop components in planted mixtures. Good site preparation can obviously help greatly, yet we are seeing prospects of major genetic gains in early growth rates. Western hemlock is a case in point. Very often it comes in as an apparent climax species, yet has small seeds and often profuse seed production which are often more characteristic of a pioneer species, and can invade areas rapidly after fire or storm damage. What is fascinating is the presence of very wide variations in early growth among progenies (J.N. King in prep.). While the evidence is still very tenuous, this suggests that the species contains a broad spectrum of genetic variation, ranging from genotypes that are adapted, by virtue of rapid early growth, to take possession of the site as pioneers, at one extreme, through to genotypes that are adapted to survive under a canopy with intense root competition, but which may be inherently slower growing. By selecting for a population of pioneers, very big gains seem possible.

A similar case may exist in interior spruce, which also shows great variation among progenies in growth rate (Kiss & Yeh 1988, Yanchuk & Kiss 1993), and is not usually of pioneer status. There the situation is less straightforward, because it tends to be attacked by the white pine stem weevil, so selection for weevil resistance will need to accompany selection for growth rate if the species is to be a satisfactory pioneer. This will reduce the expected rate of genetic gain for growth rate, but even with this 'coadaptive drag' the prospects may be very attractive. In some species, however, there may even be a negative coadaptive drag, in that regeneration following a major disturbance can be healthier, e.g. with less dwarfmistletoe, than in undisturbed areas.

Geographic Transfer of Material

Transfers of material, geographically or successionally, can often be very successful, although early growth can sometimes flatter to deceive. Geographic transfers may often succeed because local material is already suboptimal, presumably through lags in climatic adaptation, or limitations of the genetic bases from which areas were colonised, or both. The natural niches of species are defined by multiple factors which, by imposing extreme multi-trait, natural selection, will tend to prevent adaptive responses beyond those limits; however, if only a single factor is pre-eminent it would effectively come under the full weight of single-trait natural selection, which is conducive to a rapid response that will cover the tracks of the process. Hopefully, under artificial regeneration we can remove a complex of limiting factors and thereby permit significant adaptive responses. For each species, however, the situation must be treated on its own merits. At one extreme, Douglas-fir can be transferred northwards over considerable distances in the Coast ranges; at the other, the stem weevil problem has made certain geographic transfers of Sitka spruce very sensitive. With successional niche transfers there could be special needs to protect natural climax species from grass competition in establishing plantations.

Disease and Pest Resistance

Intensive management will often make increased demands upon resistance to diseases and pests, through growing species in monoculture, changing a species, successional status, or growing a preferred species to its environmental limits, depending on the case. While much more can be said, I will make just one observation. With disease resistance there is often the spectre of resistance being overcome by genetic shifts in a pathogen. However, the availability of DNA markers for Quantitative Trait Loci should now make it possible to dissect pathosystems, identifying gene-for-gene interactions that may exist between host and pathogens. Thereby it should be possible to deploy multiple resistance factors in cultivar populations so as to greatly enhance the prospects of resistance being durable.

Whether multiple resistance factors in single genotypes are preferable to using a mix of different genotypes carrying different factors is still moot, but would depend on the biology of both host and pathogen.

Wood Properties

With rapid gains in tree form, in which a reasonable breeding goal may often be approached soon, and with very finite scope for gains in production (given the constraints imposed by the environment and the fundamental shoot architecture of the species), thought must be given to what other gains should be sought. As time goes on we shall have obtained the most easily captured gains, but with our increasing skill and knowledge we hope to maintain the rates of gain despite moving into more difficult areas.

Wood properties represent a case in point. They have been studied for a long time, but have tended to be involved in numerous trade-offs, e.g. between the properties required for different pulp and paper-making characteristics, or between wood density and stem volume production. With these trade-offs, and the scope for manipulating pulp properties by log segregation or in the mill, the wood users have often hesitated to give tree breeders a clear mandate. Nevertheless, wood density has become increasingly accepted as selection criterion, while certain chemical pulping traits are now being adopted in screening eucalypt clones in Brazil (Fonseca et al. 1995).

Wood properties, while typically showing significant tree-to-tree genotypic variation and inherently high heritabilities (if they can be measured on candidates with enough precision), are usually expensive to measure. This is especially so for mechanical pulping properties, which are extremely expensive to evaluate directly. Yet the 'Clone 55' example with *Pinus radiata* is a case of a particular genotype having substantially superior mechanical pulping properties (Corson et al. 1989), almost like a different species. This was discovered rather fortuitously, but exemplifies the challenge to identify easily-measured basic properties that determine processing or end-product characteristics. With a significant variant fortuitously identified, it appears that the basis for it has been traced (Donaldson 1993). However, major variants in respect of other processing/end-product properties that are very costly to evaluate may be far more difficult to identify empirically, particularly if, as is suspected, uncommon alleles of large effects are involved. It becomes an issue for both practical screening and management of genetic resources. Tenuous as it may seem, a promising approach might be to identify what basic properties govern the differences among species in processing and utilisation characteristics, and seek basic-property variants with resemblances to other, preferred species.

High hopes are held for genetic engineering for improving wood properties. However, not only do promising DNA sequences have to be identified for transformation, but also their efficacy after incorporation needs to be demonstrated, plus the lack of adverse side effects on the mechanical stability and other components of field fitness.

MANAGEMENT OF GENETIC MATERIAL

With the success of forest tree breeding, the commercial forest estate is becoming progressively dominated by plantings that are genetically improved. The ongoing genetic improvement, and the maintenance of the genetic variability for longer-term genetic gain and general flexibility are now classically achieved in the breeding population which underpins the production population (i.e. seed orchards or clonal stool beds, etc.) and which contains greater genetic variability but a somewhat lower level of genetic improvement, which reflect the broader parentage or genetic base. An effective breeding population size of upwards of 200 (generally 300-500) is a widely accepted guideline (White 1992). In turn, the breeding population is typically underpinned by the gene resources, which represent a still broader genetic base (with a population size around an order of magnitude or so greater than the breeding population) but at the cost of less (if any) genetic improvement. While this entails an 'improvement gap', that gap is typically minor in forest trees relative to the daunting gap between crop plant cultivates and their wild relatives. It is desirable, however, to manage the gene resources to prevent this gap from becoming too great in the future (Burdon 1988).

Breeding Populations

Management of breeding populations has generated a considerable literature (e.g. Eriksson et al. 1993). A major development has been the multiple-populations concept, put forward by Namkoong (1976), which addresses a diverse array of breeding goals within various subdivisions of the breeding population. In so doing, it addresses uncertainties as to what breeding goals are appropriate for the future and amplifies the genetic variation within the species by the multidirectional selection creating divergences in allele frequencies between the subunits. It enhances the probability of retaining alleles if they have selective advantages within any subunit; at the same time, this subdivision leaves the probability of retaining currently neutral alleles essentially unchanged relative to an indifferented population of the same size, yet having the potential to create concentrations of the low-frequency alleles within subdivisions. Its effectiveness can be enhanced by controlled-pollination in seed orchards (or for producing clones for mass propagation), by allowing absolute control of inbreeding (Burdon 1986b, 1988), or by allowing specific crosses between parents in any desired combination of subpopulations. While conceived to address uncertainties concerning future technical requirements, this scheme can be adapted to breeding for range of climates (Eriksson et al. 1994). There are many other aspects of breeding population management e.g. choice of mating designs in relation to genetic parameters and biological constraints, designating elite subunits, and other variable representation of parents (Lindgren et al. 1989, 1993), but I will leave this topic, except for where it impinges on the issue of inbreeding which I will address.

Inbreeding as a Phenomenon and a Tool

Some inbreeding naturally occurs in most forest trees even if they are fundamentally outbreeders. It can take the form of self-fertilisation or, in natural stands, mating among nearby relatives. It appears that many of the resulting embryos abort, being homozygotes for embryo lethals but, because of archegonial polyembryony, will not necessarily result in empty seeds (Williams & Savolainen 1995). Even so, the viable seeds often show appreciable inbreeding which is reflected in positive F statistics. Selective elimination of homozygotes, however, tends to occur during the life of a stand such that the parents for the next generation typically show F statistics around zero or even very slightly negative (Bush & Smouse 1992). This selective elimination would come at little cost in a 'soft' selection situation wherein more seedlings of a species appear than there is space for mature trees (unless, perhaps, one cares to argue that it reflects over-investment in reproduction).

Where stands are planted with a mix of segregant genotypes, as with seedlings or vegetative multiplication, the unmasking of the 'soft' genetic load (involving mildly deleterious alleles) is not desirable because we do not want to have to plant a proportion of genotypes that are bound to fail. In theory, some of the load can be addressed by nursery culling, but increased culling rates are unattractive while the load may not be clearly expressed in the benign environment of the nursery.

In a breeding population, mild inbreeding will tend to hamper the initial process of 'forwards' selection, since the inbreeding depression will tend to mask the additive gene effects of prime interest. How much theoretical inbreeding can be tolerated in this context is uncertain, given that homozygosity should build up at something less than the theoretical rate, and that inbreeding depression for a given degree of relatedness will be highly specific to parents and individual offspring. Testing candidates in outcrosses for 'backwards' selection should give unbiased estimates of breeding values, but not before inefficiencies have been incurred in the choice of candidates, and it may often represent an additional cost. We thus need to know much more about the actual impacts of low to moderate levels of theoretical inbreeding in breeding populations. Fortunately, this now looks possible with the development of new DNA marker technology.

For clonal forestry systems some inbreeding in the candidate material may be quite easy to tolerate (Burdon 1986b). This is because the selection of clones tends to entail progressive culling, in which it may be possible to screen and eliminate many candidates at low cost early in the process. Hopefully, any inbreds can be largely eliminated in this way.

Historically, enthusiasm for inbreeding as a breeder's tool for forest trees has fluctuated. It appears initially to have been driven by a hope of emulating the successes of the hybrid maize breeding programmes. The most vigorous proponent of this approach was the late Alan Orr-Ewing, and although it yielded no practical results with the Douglas fir, the experience was highly instructive, largely because the approach was given a thorough trial. In full hindsight, it is now clear why some features of the hybrid maize approach are less attractive for forest tree breeding. For forest trees the uniformity that could only be obtained in maize by using inbred lines could easily be obtained in some species by vegetative propagation systems. Moreover, extinction rates of inbred lines that could be tolerated in maize could not be tolerated with the work content of crossing and the generation times for forest trees. And, even for maize, the superiority of the intense inbreeding followed by outcrossing, relative to alternative systems, was not incontestable (cf Vencovsky et al. 1988).

Interest in inbreeding is now reviving in forest trees, but it is worth reviewing the circumstances in which inbreeding may be accepted or even actively embraced as a tool for managing breeding populations, along with the rationales:

1. Acceptance based on the fact that in any closed, finite population some inbreeding will eventually occur. The most passive approach is to constitute and maintain the smallest breeding units, e.g. addressing a particular breeding goal or climatic zone, at a size, say, an effective number (Ne) of 30-50 parents, that should lead to minimal inbreeding depression and associated biases in estimating breeding values, in either the population unit or in a seed orchard constituted therefrom. With a multiple-populations approach such a level of inbreeding may be accepted as a price of the potential benefits of specialisation and a progressive increase in the expression of genetic diversity.

2. A slightly less passive approach is embodied in sublining (Burdon & Namkoong 1983) which, by subdividing the population into parallel unrelated units, entails accepting an earlier build-up of inbreeding in order to guarantee outcrossing in the future. With control-pollinated orchard systems this requires only two sublines, which can duplicate the multiple-populations structure (Burdon 1986b, 1988). If there is no fine subdivision according to breeding goals the build-up of inbreeding even within the two sublines can be very slow.

3. The more active forms of inbreeding can involve the adoption of very small breeding groups, say 10 or less (cf McKeand & Bridgwater 1992) or in the most extreme case, self-fertilisation. The rationale for self-fertilisation if it works can be severalfold (Williams and Salovainen 1995):
    - purging of deleterious alleles
    - maximising future options for outcrossing
    - maximising expression of additive genetic variation in the population though assuring perfect assortative mating for all traits simultaneously.

Feasibility, however, is crucial and for many species, such as Douglas-fir, it is not effectively met. The compromise, which can be expected to dilute the advantages listed above, is to intermate in small groups, say 4-10, which will quite soon necessitate some inbreeding. From the outset, even with no initial relatedness, appreciable genetic diversity among such groups is inevitable, if only through drift, or genetic sampling error, for some traits. Indeed, such schemes should paradoxically impose an upper limit on the erosion of the genetic base.

On the inbreeding front we are looking at some exciting developments which have been addressed in more detail in the preceding NAQFGG meeting. At the research level, the new suite of PCR-based DNA markers affords powerful new tools (Fu and Ritland 1994) to confirm, refine or refute our notions of the detailed phenomena underlying the observed behaviour upon inbreeding. This will have wide ramifications for our understanding of evolutionary process. At a more practical level, there is the case of western red cedar (Russell et al. in prep.) in which active use of strong inbreeding, even selfing, seems to be feasible, and appears very attractive for capitalising upon the modest but apparently well worthwhile genetic variation that now appears to exist.

Gene Resource Management

As mentioned earlier, it is widely advocated that breeding populations should be underpinned by gene resource populations (although breeding material can also provide insurance against loss or depletion of native populations). At the crudest level this provides further back-up. Moreover, *in situ* genetic resources can be retained essentially incidental to maintaining the ecological reserves that may exist in their own right or in parks, with ready ethical and political justification -- although appropriate management may be easier said than done, technically and politically.

The Role of Biotechnology

Biotechnology can provide very powerful tools that can be used in various ways for both breeding and the underpinning research. It must, however, be regarded as providing enhancements of the traditional breeding infrastructure, viz controlled crossing, progeny tests etc., rather than providing substitutes (Burdon 1992). Indeed, much of the development of the biotechnology will, at least in the short to medium term, require field-based material often far beyond what is needed for classical breeding. Thus biotechnology must be regarded as an intensification of domestication which is all about being prepared to increase inputs into growing the species in order to obtain better returns (Burdon 1994).

Specifically for managing gene resources, biotechnology can be used in various ways. It may be needed to help propagate very scarce material and it can help to achieve accelerated deployment when material needs to be incorporated into the breeding population or even orchards. The use of genetic markers, moreover, can be invaluable for checking the identity of material and obtaining measures of genetic diversity. For the latter purpose, however, the diversity observable in genetic markers (usually PCR-based DNA markers) must be well calibrated against the genetic variation of practical interest as revealed in field evaluation trials (which may, however, be supplemented by laboratory trials). We now know that important discrepancies are far from rare (e.g. Libby & Critchfield 1987, Millar & Libby 1991), although they seem unlikely to arise uniformly for a wide range of markers in any one species.

A crucial question is whether biotechnology can substitute for the presence of large numbers of individuals in the field, which contain diversity because of both independent geographic origins and the numbers of unrelated parents that ensure retention of low-frequency alleles from within populations. Now that there is the prospect of using genetic transformation to incorporate specific genes from other species or even 'off the shelf' genes, the question is not trivial. An intuitive reaction of many biologists is that it would be dangerously arrogant, to assume that biotechnology can provide a solution in this area. We have seen many humbling cases where biotic factors in the form of quarantine failure, or adaptive genetic changes on the part of pest or pathogens driven by mutation and new sets of natural selective forces, have led to the re-emergence of problems that have looked to have been solved -- this has occurred frequently in medicine, in insect pest control, in breeding crop plants for resistance to fungus diseases and, more recently, in controlling weeds by herbicides.

A very large gene resource offers the most complete assurance of wide genetic diversity in terms of allele retention and the scope for recruitment by mutation (accepting that the large majority of the mutations will be unfavourable). At the same time a field planting of the resource may be deployed where biotic and climatic challenges can arise in a form that is fully relevant, being likely to give a highly appropriate resolution of genetic differences, and helping to secure any favourable mutations. Furthermore, in situ conservation of ecosystems (again, while it may be easier said than done) not only preserves the genetic variation of the prime target species but also promises the conservation of all the webs of interdependent species.

Management Conundrums

What is problematic will be how to manage the gene resources, as I have just described, be they *in situ* or *ex situ*.

The basic concept is that they should be (1) unrelated to breeding populations, so as to cause no problems with unwanted inbreeding and (2) a source of new genetic variability. To be a source of new genetic variability they should *inter alia* contain even greater reserves of genetic variation. That, in turn, is likely to require retaining numbers of trees that are too large to be perpetuated by controlled crossing. I have proposed (Burdon 1986a, 1988) that such material be maintained by open pollination with mild to moderate mass selection for general silvicultural merit (viz growth rate, health and tree-form traits). Substantial areas, in solid blocks, would give some protection against pollen contamination and give scope for significant selection intensities without seriously eroding the genetic base. The resulting genetic improvement will in turn reduce the opportunity cost, per hectare, of growing the gene resource material relative to growing seed orchard or equivalent stock, and so on (Fig. 1).

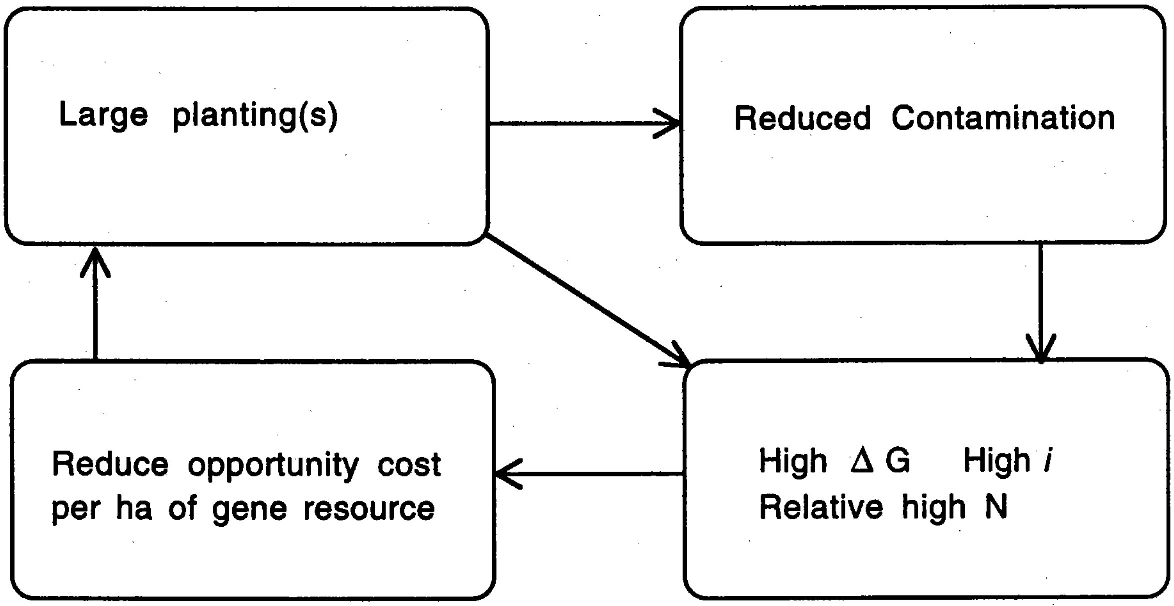


Figure 1: Feedback loop for proposed gene resource management scheme (Burdon 1986b, 1988)

Here, however, I must recant, although not as lucratively as Dr. Benjamin Spock. A potentially serious flaw in the proposal arises with wind-pollinated species that are cultivated on a massive scale. Such species produce enormous pollen clouds, with appreciable components of long-distance transport and even some heavy local 'dumping' of pollen from afar (Lindgren et al. 1995). Even large blocks of gene resource plantings may incur appreciable contamination, and this contamination will come increasingly from intensively select but quite narrowly-based material of seed orchard origin. If intensive genetic improvement is succeeding, the offspring resulting from such pollen contamination could enjoy a considerable selective advantage under the proposed gene resource management regime. Moreover, this contamination process would be expected to compound over consecutive generations (until or unless some new agent imposed new and very different selection pressures). Thus, a little contamination could eventually go a long way (Fig. 2). The significance is likely to be sensitive to a number of parameters that are largely conjectural.

For *in situ* gene resources, which may take the form of set-asides, parks, or Gene Resource Management Units the problem is likely to be much less acute, but it could still be major where a single 'keystone' species is grown, using intensively improved planting stock.

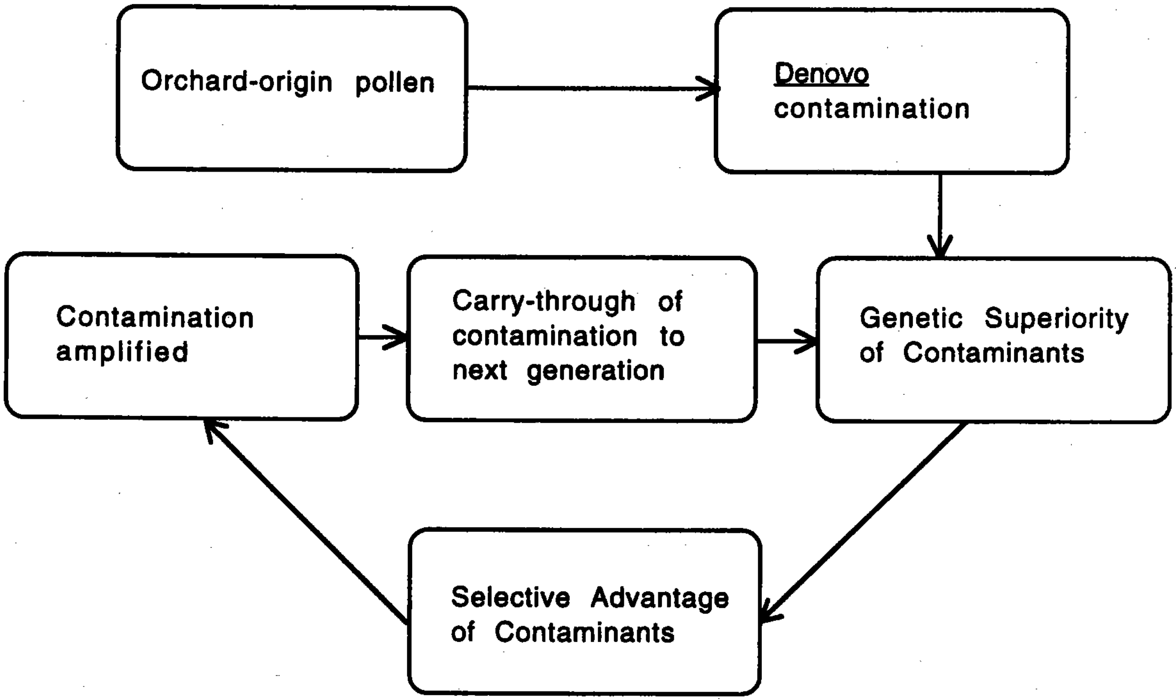


Figure 2: **Feedback loop for pollen contamination process in gene resource material**

Either way, both main planks of the original concept of the gene resource, namely (1) as a source of new, unrelated germplasm and (2) as a greatly broader pool of genetic variability to work with, could be seriously compromised.

Now look at our options. These include the following, not all of which are mutually exclusive:

1. Accepting the contamination and hoping for the best
2. Evaluating the extent of the contamination by
   (i) ascertaining pollen contamination rates
   (ii) modeling through progressive contamination assuming varying level of pollen contamination and genetic superiority of orchard stock
3. Screening candidate seed parents in the gene resource for contaminant status.
4. Extending controlled pollination and pedigree into the gene resource(s) (tantamount to substantially expanding the breeding population(s))
5. Deploying gene resource plantings away from pollen contamination
6. Deploying 'buffers' around the core gene resource plantings, which represent compromise levels of genetic improvement and breadth of genetic base
7. Relying very finite breeding populations with mutational accretion of variation
8. Relying on mutational accretion of genetic variation in the production population(s).

Some comments are needed on the various options:

1. ***Accepting the contamination.*** This entails strong assumptions as to its significance or lack of significance, although it may be vindicated if option 2 is exercised. The worst-case scenario, however, is very undesirable. Nevertheless, some relatedness to existing elite stock may be tolerable if the gene resources can play a decisive role in overcoming a future problem. One possibility is that with in situ resources the natural selective pressures may diverge so much from the pressures of artificial selection that the contaminants are at a severe disadvantage. This would certainly preserve the gene resources, but at the cost of an ever-increasing 'improvement gap'.

2. ***Evaluating the extent of the contamination.*** This will require a significant outlay, particularly in the development and application of DNA marker systems of adequate power for the purpose, but it still seems necessary unless one or more of options 4, 5, 6 and 7 would be exercised. It can be used in conjunction with options 1 and/or 3.

3. ***Screening candidate seed parents for contaminant status.*** A pre-requisite for this would be exercising option 2, which would be expensive in itself. This option, while it would avert the expense of controlled crossing, would still require major follow-up expenditure.

4. ***Extending controlled pollination and pedigree into gene resources.*** This is tantamount to enlarging the breeding population in order to dispense with the need for separate gene resource populations. To do this on a scale adequate to fulfil the classical role of gene resources would clearly be extremely expensive, and unlikely to be accepted, even if technically justified.

5. ***Planting gene resources away from pollen contamination.*** This in principle circumvents all the problems that I have raised above. On the other hand, it may necessarily entail planting gene resources of preferred species substantially off-site, which could prevent their being exposed to appropriate selection pressures.

6. ***Deploying buffer zones*** around the core gene resource plantings or ecological cores of *in situ* resources. This, as far as I know, is a fairly new proposal, which has yet to be fleshed out in detail. The prime purpose would be to protect the gene resources from the massive contamination by the quite narrowly based element of highly improved commercial stands. Candidate material for *ex situ* cases would be breeding population elements that are not closely related to orchard parents, but there are still the logistical problems of propagating such material on an adequate scale, and the buffers still confer no watertight guarantee against contamination. Set-asides of *in situ* material that are not to be logged at all might be bordered by forest that is naturally regenerated.

7. ***Relying on very finite breeding populations, with mutational accretional of variation.*** Against the inevitable loss of alleles in finite populations is the possibility, which is being increasingly raised (e.g. Namkoong & Khoshy 1995), that mutation can contribute significantly to useful genetic variation even with very finite populations (say, 50 or less). While the very large majority of mutations may indeed be adverse, this more optimistic view of the role of mutation stems partly from interpretation of long-term selection experiments (which have prompted various theoretical studies), and partly from a more inclusive view of mutation (cf Barker 1995) which can embrace various genetic events other than the classical mutations observed from ionising radiation or chemical mutagens. Such 'quasimutational events' may well be far more frequent and less consistently deleterious than classical point mutations. Hence, while rates are very speculative, mutation in the broad sense may offset much of the allele loss without populations having to be very large. Directional selection will help greatly in serving such new alleles, so a multiple-populations selection scheme

should tend to secure a broad spectrum of such mutations. However, random losses should continue unchecked for alleles that have no immediate advantage in any of the population subunits. Moreover, the finite population sizes will not be conducive to the multiple independent occurrences of equivalent new alleles that may be wanted for deploying material that is neither inbred nor narrowly based.

8. ***Relying on mutational accretion of variation within commercial stands.*** This flies in the face of the notion that commercial stands are eventually a dead-end for breeding, which has now become conventional wisdom. Yet it is in commercial stands that trees are deployed (a) in the sheer numbers that are likely to ensure that the mutational events not only occur but do so independently in a number of genetic backgrounds and (b) in situations where advantageous mutations are most likely to be expressed and thereby captured much more efficiently. Even so, the process may operate only in respect of the very rare mutants of large favourable effects. To offset the obvious disadvantages of recourse to such material may require (i) that it be rather more broadly based, in terms of numbers of parents, than would otherwise be indicated and (ii) that pedigree reconstruction be undertaken on candidates to ensure both a range of unrelated candidates and to control their use in related matings.

Possible Resolution?

Thus, if gene resources are maintained to underpin pedigreed breeding populations, it may often be crucial to ascertain the pollen contamination levels, which may offer reassurance but will not directly solve anything if reassurance is not obtained. For the longer term, the ideal for managing gene resources would be to have commercial plantations that are fully non-reproductive. Apart from preventing pollen contamination of gene resources it has the major attractions of (i) eliminating a diversion of high-grade biomass into unusable reproductive tissues and (ii) meeting a likely restriction on the release of Genetically Modified Organisms. A fully satisfactory solution may, however, be very difficult. Use of an appropriate anti-sense DNA sequence would seem to offer much the best prospect of avoiding unacceptable side- effects on field fitness. This, however, requires that the sequence be identified. By far the easiest sequence(s) to identify would be syntenous genes that are common to a wide range of organisms, yet such genes may not have the requisite role specificity.

On present knowledge the practical problems of maintaining large back-up gene resource populations can be formidable. Yet to deny these problems is likely to be as futile as denial of the importance of a fire ecology in perpetuating many populations and communities. On the other hand, Namkoong & Khoshy (1995) have pointed out the potential of mutation (*sensu lato*) to offset random allele losses and, with appropriate population management, to even expand the variability. Still uncertain, however, is how far we can rely on mutation to achieve this. We see around us the cases where mutation has delivered genetic variation that was crucial. The cases where it has not, have disappeared, except sometimes into the fossil record. Moreover, accretion of fresh alleles in finite populations will be far less efficient if the populations are not deployed where the new alleles have an immediate selective advantage.

The technical ideal would still be to retain the large gene resources, partly for pure back-up, and partly with the prospect of serving complementary purposes. Yet the difficulties and costs of maintaining such resources, and the probable need for a disaster to prove their worth, are powerful disincentives for financial managers, which may become even stronger in the context of the tantalising capabilities of mutation. Nevertheless, public opinion, driven largely by environmental watchdog groups, may well force the forestry community into maintaining the gene resources. Public opinion can be a scourge of the industry, often with misconceptions that even-aged stands are totally unnatural, but here it may well prove an ally for us to do what is right.

FINAL REMARKS

Now to some final comments. I am a biologist, and many of you are basically biologists. Yet tree breeding will depend greatly on appropriate institutional structures; there seems no way that the isolated backyard breeders can contribute much to the mainstream of future tree breeding.

Tree breeding has produced a distinctive type of institution, the Cooperative Tree Breeding Program and variations thereof. This has reflected both the difficulties usually faced by any one company (or other agency) trying to mount a fully viable, self-sufficient programme, and the benefits of pooling breeding material across territorial boundaries of no biological significance. This type of institution, while it has served tree breeding very well, is facing severe strains arising from rapid advances in biotechnology. The strains arise in two ways, through accentuating the divergences between member companies in the levels of technology to which they are committed, and through encouraging proprietary appropriation of technology. The latter is far removed from the original concept of a cooperative, but the new technology can create what are arguably grey areas. When once proprietary appropriation takes hold it can easily encourage a defensive spiral among all parties, much to the detriment of the scientific communication that can benefit all. Moreover, individual aspects of new biotechnology almost all entail high risks of either failure or early obsolescence, so the risk spread of collaborative arrangements can actually be very attractive, so long as there is commitment among all parties.

Management of genetic material will certainly need broad collaboration if it is to succeed. It would be tragic if that collaboration broke down in pursuing other gains without an adequate risk spread.

ACKNOWLEDGMENTS

I thank Gene Namkoong and Alvin Yanchuk for helpful comments on the draft.

REFERENCES

Barker, J.S.F. 1995: Quantitative genetic models: Past, present and future challenges. Proc. CTIA/WFGA Conf., Victoria, BC, Aug 1995.

Burdon, R.D. 1982. Breeding for productivity - Jackpot or will-o'-the wisp? Pp 35-51 in Proc. 7th North Am. For. Biol.Workshop, Lexington, KY.

Burdon, R.D. 1986a: Gene Resource Management Plan. Ch 4 (pp 51-69) in Shelbourne, C.J.A., Burdon, R.D.; Carson, S.D.; Firth, A.; Vincent, T.G. Development Plant for Radiata Pine Breeding. Special publication, New Zealand Forest Service, Forest Research Institute, Rotorua.

Burdon, R.D. 1986b: Clonal forestry and breeding strategies - a perspective. Pp 645-59 in Proc. IUFRO Genetics Group Meeting, Williamsburg, VA, USA, Oct. 1986.

Burdon, R.D. 1988: Recruitment for breeding populations: objectives, genetics and implementation. Pp 555-72 in Proc. 2nd Internat. Quant. Genet. Conf. (Ed. Weir, B.S. et al) Raleigh, NC, USA, June 1987. Sinauer.

Burdon, R.D. 1992: Tree breeding and the new biotechnology - in damaging conflict or constructive synergism? Pp 1-7 in Proc IUFRO Meeting on Breeding Tropical Trees, Cartagena & Cali, Colombia, Oct 1992. CAMCORE.

Burdon, R.D. 1994: The place of biotechnology in forest tree breeding. Forest Genetic Resources Information 22: 2-5. FAO, Rome.

Burdon, R.D.; Namkoong, G. 1983: Sublines and multiple populations. Silvae Genetica 31: 81-5.

Bush, R.M.; Smouse, P.E. 1992: Evidence for the adaptive significance of allozymes in forest trees. New Forests 6: 179-96.

Carson, S.D.; García, O. (in MS): Genetic gain in *Pinus radiata* expressed as growth rate multipliers. Sumbitted to For. Sci..

Corson, S.R.; Foster, R.S.; Richardson, J.D. 1989: New Zealand grown spruce and radiata pine can have similar TMP properties. Appita 42: 345-9.

Donaldson, L 1993: Cell wall fracture properties in relation to lignin distribution and cell dimensions among those genetic groups of radiata pine. Wood Sci. & Technol. 29: 51-63.

Eriksson, G.; Namkoong, G.; Roberds, J.H. 1993: Dynamic gene conservation for uncertain futures. For. Ecol. & Manage. 62: 15-37.

Evans, L.T. 1993: Crop Evolution, Adaptation and Yield. Cambridge Univ. Press.

Fonseca, S.M.; Oliveira, R.C.; Silveira, P.N. 1995:: Industrial tree selection: Procedures, risks, costs and benefits. Pp 14-19 in Proc. CRCTHF/IUFRO Conf. "Eucalypt Plantations: Improving fibre yield and quality" (Ed Potts, B.M. et al.) Hobart, Tasmania, Australia, Feb. 1995.

Fu, Y.B.; Ritland, K. 1994: Evidence for the partial dominance of viability genes contributing to inbreeding depression in *Mimulus guttatus*. Genetics 136: 323-31.

Kiss, G.; Yeh, F.C. 1988: Heritability estimates for height for young interior spruce in British Columbia. Can. J. For. Res. 18: 158-62.

Libby, W.J. 1987: Do we really want taller trees? Adaptation and allocation as tree-improvement strategies. H.R. MacMillan Lectureship, University of British Columbia, Vancouver, B.C., Canada.

Libby, W.J.; Critchfield, W.D. 1987: Patterns of genetic architecture. Annales Forestales 13: 77-92.

Lindgren, D.; Libby, W.J.; Bondesson, F.L. 1989: Deployment to plantations and proportions of clones with special emphais on maximising gain at a constant diversity. Theor. Appl. Genet. 77: 825-31.

Lindgren, D.; Wei, R.-P.; Bondesson, F.L. 1993: Optimal selection from families. Heredity 70: 619-21.

Lindgren, D.; Paule, D.; Xihuan, S.; Yazdani, R.; Segeström, U.; Wallin, J-E.; Lejebro, M.L. 1995: Can viable pollen carry Scots pine genes over long distances? Grana 34(26295/138-94).

McKeand, S.E.; Bridgwater, F.E. 1992: Third-generation breeding strategy for the North Carolina-Industry Cooperative Tree Improvement Program. Pp 234-40 in Proc. IUFRO Meeting on Breeding Tropical Trees, Cartagena & Cali, Colombia, Oct 1992. CAMCORE.

Millar, C.I.; Libby, W.J. 1991: Strategies for conserving clonal, ecotypic and disjuct population diversity in widespread species. Pp 149-70 in Falk, A.D.; Holsinger, K.I. (Ed.) "Genetics and Conservation of Rare Plants". Oxford Univ. Press

Namkoong, G. 1976: A multiple index selection strategy. Silvae Genet. 25: 199-201.

Namkoong, G.; Khoshy, M.P. 1995: Managing the genetic variance. Proc. CTIA/WFGA Conf. Victoria, BC, Aug. 1995.

Russell, J.H.; Burdon, R.D.; Yanchuck, A.D. (in prep): Inbreeding depression and experimental selfed populations in western red cedar.

St. Clair, J.B. 1984: Genetic variation in tree structure and its relation to size in Douglas-fir. I. Biomass partitioning, foliage efficiency, stem form and wood density. Can. J. For. Res. 24: 1226-35.

Vencovsky, R.; Miranda F., J.B.; Souza, C.L. 1988: Quantitative genetics and corn breeding in Brazil. Pp 465-77 in Proc. 2nd Int. Quant. Genet. Conf. (Ed. Weir, B.S. et al.), Raleigh, NC, USA, June 1987. Sinauer.

White, T.L. 1992: Advanced-generation breeding populations: Size and structure. Pp 208-22 in Proc. IUFRO Meeting on Breeding Tropical Trees, Cartagena & Cali, Colombia, Oct 1992. CAMCORE.

Williams, C.G.; Savolainen, O. 1995: Inbreeding depression in conifers: Implications for breeding strategy. For. Sci. 41(4).

Yanchuk, A.D.; Kiss, G.K. 1993: Genetic variation in growth and wood properties and its utility in the improvement of interior spruce in British Columbia. Silvae Genet. 42: 141-8.

APPLIED CONSIDERATIONS
FOR
BREEDING POPULATION STRUCTURE
AND
SELECTION CRITERIA

EXPERIENCES IN BREEDING STRAWBERRIES AND FOREST TREES

Douglas V. Shaw

Pomology Department
University of California, Davis

At this time I have spent approximately 10 years in each of two professions: the genetics and breeding of forest trees and strawberries, respectively. My objective here is to provide an overview of my experience in both professions, with special reference to research results that interface between the two. My current position is with the University of California, and I have statewide responsibility for strawberry breeding and cultivar development. I'll begin with a brief overview of the strawberry industry in California, the basic design of the cultivar development program, and some homologies and contrasts. Then I'll follow with some discussion of specific research results obtained from strawberry that might impact forestry.

In many respects my current industry is rather localized, as there are few places in the world where the implementation of highly productive strawberry cultural systems is possible. California has 23,500 acres of strawberries, produces about 80% of the fruit in North America, and 25% of the world's fruit. Our annual crop is worth just over 700×10^6 USD and we ship some quantity of fresh fruit 12 months of the year. Annual nursery plant production for strawberry transplants used in California is about 650×10^6 plants, with an equal number exported to other parts of the world using CA cultivars (which include Italy and Spain).

Strawberry is a hybrid–derived crop of relatively recent origin, and relatively few breeding cycles have passed since its domestication. Wild relatives of the domestic strawberry are octoploid and dioecious, but modern populations are diplodized, monoecious, and handled using population breeding methods. Production populations are single genotypes, vegetatively propagated using runner plants. Key similarities between forest trees and strawberries include: improvement using cross– fertilized populations, limited numbers of improvement cycles since domestication, and a large number of different environments requiring adapted genotypes. Important features of California strawberries that differ from those of forest trees include: production dependent on annual planting, short generation intervals, manipulability of the production environment, production using vegetative propagules, the importance of physiological tricks/culture, and the number of different important traits requiring simultaneous improvement. Perhaps most importantly, control of native environmental variation is critical in forest tree testing programs; in strawberries the number of traits requires for each individual genotype effort predominates the testing system design.

Despite some obvious differences, I have identified four research topics with results that might impact forest tree breeding. First, strawberry appears at first approximation very sensitive to inbreeding, and like forest trees this is a concern for long–term sustained progress. However, although very small populations have typically been used and severe depression is easily demonstrated for current crosses among relatives, no accumulated depression due to pedigree F over multiple cycles of weak inbreeding and strong selection can be detected. Second, perhaps the most obvious success in strawberry breeding has been for improvement in productivity. The dramatic improvement in California strawberry yield can be traced to several factors including extending the length of the fruiting season and modifications to resource partitioning. Despite the accumulated selection response, additional progress seems likely. Third, improvement in strawberries depends heavily on indirect selection. Similar to experiments with forest trees, genetic and environmental variance parameters change over the "rotation". In general I have found that information about performance during relatively small proportions of the season seem adequate for selection of genotypes at early stages of the program. Fourth, and again an issue common with forest tree breeding, the choice of test environments is a critical concern for strawberries. Individual strawberry environments are perhaps easier to control, but on the whole as diverse as those encountered in forestry. Several examples including selection based on seedlings vs vegetative runners are demonstrated.

EXPERIENCES IN BREEDING MAIZE AND FOREST TREES

James L. Brewbaker

Department of Horticulture, University of Hawaii
3190 Maile Way
Honolulu, HI 96822

The discussion focusses on concurrent breeding of tropically adapted vegetable ("super-sweet") maize and the tropical fodder-legume, leucaena. Generalizing, this combination of horticulture (agronomy) and forestry has a refreshing array of advantages, and no compelling disadvantages. It does compel the view that in the tropics, education and research in horticulture and forestry can be co-institutionalized to advantage.

The UH maize breeding program involves bi-monthly plantings year round, pollinating in 2 months and harvesting in 4. Leucaenas are planted annually, pollinated in year 2 and harvested 3 months later. Evaluations are divided into trials for fodder, on 3-month harvest, and wood trials on 2 to 4-year harvest regime.

Most tropical supersweet maize derives from the 3-decade UH breeding program, based on an unusual gene, brittle-1, in the unusual background of tropical field corns. Temperate corns have been of little use, due to a narrow gene base with high susceptibility to most corn diseases and pests. Fifteen major composites and inbred-based synthetics have been released, together with many inbreds and their highly-heterotic hybrids, of which 3-way hybrids have been most successful. Recurrent mass selection under conditions ensuring selection advance (diseases, stresses, etc.) is favored over systems with greater effort and greater gain. Inbred development has only been successful from inbred-based synthetics. Backcross conversions are employed to add genes for color.

Most improved tropical leucaena derives from the 3-decade UH program, based on arboreal accessions of the self-fertile polyploid, L. leucocephala, and crosses with two other polyploids and a few of the 13 diploids in this genus. Several Latin American expeditions contributed to the thousand accessions growing in Hawaii, and all species were intercrossed dialley. About 90 viable hybrids have been under study. Four stages in genetic improvement have been (1) elite accessions (selfed lines), (2) composites for forage based on cross with L. pallida (psyllid-resistant), through four cycles of recurrent mass selection, (3) pure-line-based synthetics for wood production that exploit half the (high) heterosis for yield and (4) 3-way crosses using self-sterile clonal singlecrosses that exploit all of the heterosis. Seedless and highly heterotic triploid clones K1000 and K1001 represent an emerging fifth stage, as cloning becomes commercially practical.

Some of the great genetic achievements of the century have impacted rather little on these programs, although academically intriguing; radiation-induced mutation, chemical mutagenesis, induced polyploidy, isozymic polymorphism, tissue culture, translocation and RFLP-aided gene mapping, etc.

A LOCUS BASED MODEL FOR ADVANCED GENERATION BREEDING

Floyd Bridgwater

USDA Forest Service
Texas A&M University
Texas, U.S.A.

Many tree breeding programs have adopted some form of multiple population strategy (sublining) to manage coancestry. Several questions arise about how these populations should be formed and managed. Computer simulation of the breeding strategy adopted by the North Carolina State University-Industry Cooperative Tree Improvement Program suggests that:

1. Selecting the best individual from the four best families (self or outcross) ranked on expected breeding values is an alternative that will give good genetic gains and result in relatively moderate rates of increase in coancestry within sublines.
2. Assigning parents to sublines at random, or disassortatively, rather than by positive assortment will increase within-subline genetic variance and result in greater increases in breeding value for the first generation of selection and mating.
3. It will be possible to enrich elite populations from the mainline population for 3-4 generations without reducing gains in the elite breeding program.

A fourth, important decision was the choice of small sublines, size 4. These very small sublines were adopted for three reasons.

1. Smaller sublines reduce the chance that rare alleles included in the population will be lost.
2. Small sublines can be combined in future cycles if necessary to create larger sublines while the converse is difficult if not impossible.
3. Smaller sublines help to maintain population variability (genetic diversity) in the long-term.

Comparison of the population variances for 20 generations for subline sizes 4, 6, 8 & 12 show that with the model assumptions and subline size 4, population additive variance increases for 7 to 8 generations before slowly declining. The additive genetic variances averaged over all sublines decrease much more rapidly in the larger sublines. This occurs because selection within small sublines implies a lower selection differential and smaller genetic gains. Thus, the additive genetic variance is reduced more rapidly in larger sublines.

These relationships serve to illustrate that maintaining genetic diversity comes at the price of reduced genetic gains in the short-term. However, trading short-term genetic gains to maintain genetic variability in the long-term means that the breeding population will remain viable for many generations.

CONSERVATION, PUBLIC POLICY AND SCIENCE[1]

Clark S. Binkley

Dean
Faculty of Forestry
University of British Columbia
Vancouver, BC V6T 1Z4
604/822-2467 tel
604/822-8645 fax
binkley@unixg.ubc.ca e-mail

I have had warm feelings for the CTIA ever since your meeting in Ottawa in 1991 when I met Gene Namkoong and began to convince him that BC was the most exciting place in the world for him to continue his career. As a result, you can all share some of the credit for bringing him to Canada.

Despite these warm feelings, tonight I am going to violate the first rule of dinner party etiquette: never discuss politics or religion. Conservation in BC inevitably involves the former, and usually touches on the latter.

My talk tonight will develop three points: (i) the necessity of conservation, or, more precisely, the *inevitably* of conservation if economic development continues, (ii) the specific challenges conservation brings to public policy, and (iii) the unique contributions science can make to conservation policy.

I. Why is Conservation Necessary?

Especially in Canada, Nature defines much about human culture and spirituality. And perhaps this has always been so. Nearly two millennia ago, the Roman philosopher Seneca observed

> If you have ever come upon a grove that is full of ancient trees which have grown to unusual height, shutting out the view of the sky by a veil of pleated and intertwining branches, then the loftiness of the forest, the seclusion of the spot, and the thick, unbroken shade in the midst of open space will prove to you the presence of God.

The ecologist Gordon Orians has linked the emotional resonance that some forested landscapes produce to our evolutionary past in the plains of East Africa. There, he argues, early humans had neither the luxury of time nor the benefit of formal scientific analysis to identify those landscapes which would be suitable for human habitation. Instead, those individuals who picked up the proper cues from the vegetation and landforms survived to become our ancestors. Gordon claims that our attraction to certain landscapes derives from the evolutionary fitness they once conferred. He has conducted detailed allometric analyses comparing *Acacia* savannas with renowned English landscape paintings and successful landscape designs, and his findings are consistent with this evolutionary hypothesis explaining our landscape preferences.

---

[1] Presented at the Canadian Tree Improvement Association Meeting, 29 August 1995, Victoria, BC. References to material quoted or cited available on request.

Of course, it is dangerous for an economist to make evolutionary arguments among a group of geneticists, so I will quickly move on to my main point--the economic necessity of conservation. The well-known Brundtland Report articulates half of the argument: a healthy environment is necessary for producing a healthy economy and a healthy economy is necessary for producing a healthy environment. When stated this way, it is obvious that Brundtland's "sustainable development" is a supply-side argument: the *supply* of economic development and environment quality are inextricably linked. As such, this line of thinking can give only half of the story.

The other half comes from the demand side. The objective of economic development is to increase personal incomes. The demand for conservation products and services is highly correlated with income levels. Good empirical evidence substantiates this claim for some features of the environment--clean air, clean water and outdoor recreation--and it probably is more widely applicable as well.

If economic development is successful, then conservation will inevitably become more important economically. So, it is economic activity that enables us to be concerned about conservation. The contrary idea--that we must reduce economic activity to insure sustainability--will be self defeating. With decreases in income will come a decline in society's demand for conservation and an increase in environmental destruction. When Larry Summers worked for the World Bank, he made a similar observation and argued that the best way to enhance environmental quality was to speed the passage of countries through the highly-polluting middle-income stages of development.

As an aside, this argument has an important microeconomic implication for those involved directly with industry: if companies are successful in increasing profits, they will also increase societal demand for conservation. As a consequence of this demand-side link between conservation and economic prosperity, successful long-run business strategies must embrace high levels of environmental performance.

II. What Challenges does Conservation Create for Public Policy?

Public policy becomes necessary when the normal mechanisms for mediating interactions among humans fail. In liberal western democracies, markets are one of the most ubiquitous mechanisms for allocating resources among competing human claims. Most of the products or services we collectively call "conservation" operate outside formal markets. They may be pure public goods such as aesthetically pleasing landscapes or the preservation of endangered species. Or society might simply have chosen not to allocate them through markets. Examples in BC include clean water flowing from a forested watershed--we sell water licences for far less than their economic worth--and recreational hunting and fishing which are allocated through markets in most of Europe and much of the US.

Because conservation operates outside formal markets, the "price" of conservation activities is zero. As a result, society systematically *overconsumes* features of the natural environment and *underproduces* them. When the *supply* of conservation products and services is large compared to their *demand*--as it was for most of BC's history--the cost of establishing conservation institutions exceeds the benefits of better management and doing nothing makes perfect sense. Now, however, the situation has changed. The cost of inaction has become too great, and society can no longer ignore the losses from inefficient resource allocation. The situation literally *demands* public policy.

There is a spectrum of policy options for protecting these critical conservation values. At one extreme we could elect to create private property rights in conservation values and leave production and allocation decisions to the market place. At the other extreme, we could protect these values through pure "command and control" regulation. In BC we have clearly opted for a fairly extreme version of the latter approach. In part our preference for regulation has to do with what the Nobel-prize winning economist Patrick Buchanan called "rent seeking." Each of us--whether from industry or from environmental organizations--now enjoys the benefits of low-cost or free access to conservation products and services, and we naturally dislike the idea of actually having to pay for something we now get for free. It is cheaper for us to lobby government to achieve the ends we desire than to contract with the owner of a private property right to produce the conservation values we wish to consume. I doubt we will adopt private property/market solutions to conservation policy problems until we have played out all of the problems with the command and control approaches, so I will not discuss privatization options further tonight.

III. How can Science Support Conservation Policy?

A regulatory regime for producing the increasingly valuable conservation products and services places some specific and significant demands on science. Some of these same demands would exist with market approaches to the public policy problem, but the decentralized nature of markets tends to disperse the need for scientific information and to diversify the risks of incorrect answers.

The first demand is to forecast the consequences of specific policy interventions. This generally requires clarifying cause-effect relationships of the "if/then" type--*if* we establish a landscape corridor, *then* a specific suite of species will persist on the landscape. And perhaps more importantly the converse: if we do *not* establish the landscape corridor, then this same suite of species will not persist. These sorts of predictions are the ordinary stuff of science, so they would seem to present no great difficulties. But they do.

In the first place, the great bulk of science in Canada is driven by hypotheses of *theoretical* importance to a particular discipline rather than by hypotheses of *practical* utility to conservation and land management. We tend to study photosynthesis in algae or *Arabidopsis thalnia* rather than in spruce. As a result, there are too many untested but plausible management hypotheses which claim an undue amount of attention in policy debates.

In the second place, definitive experiments for many management-relevant hypotheses will require large landscapes to conduct. In BC we literally have no land devoted exclusively to landscape-scale experimentation. This inadequacy would not be so significant if we truly adopted the principles of adaptive management--explicit experimentation in actual management practice, replication of experiments, controls, and long-term monitoring--but we have not. Indeed our regulatory regime tends to homogenize land management practices, and will therefore slow our rate of learning. Nor are we permitted to "test to failure" as engineers or doctors do to understand quickly and powerfully the limiting conditions in the systems we manage.

The second demand on science is for open information. Liberal democracies require public policy processes which reflect democratic principles. By its nature, science is democratic. Scientific information emanates from a process in which any suitably educated person can participate. No divine connection or familial relationship with a powerful politician is required. As a consequence, the authority of science coincides with Canadian polity. To secure this authority simply requires the ordinary process of science--publication in the open literature, third-party replication of findings, and rigourous, unbridled debate of methods and conclusions.

Yet even here we have recidivism. For example, in the United States the government biologists who provide the internal agency information used for listing species as threatened or endangered are not permitted to defend their results publicly. Broadly applied, this principle would produce the "scientist as shaman", a result which is consistent with neither good science nor good public policy.

The third demand is to create new options. Politicians are generally neither stupid nor venal, so "obvious" scientific solutions to conservation problems usually create one or more serious, unacceptable economic, political or social side effects. Inexpert in the social sciences or humanities, bench and field scientists are, not surprisingly, unaware of such constraints on policy intervention. Social scientists involved in the policy process spend a great deal of time trying to figure out ways around these side effects so that otherwise sound practices can be implemented.

The wonderful creativity of science can provide wholly new approaches to such tough problems. These "outside the box" approaches can starkly shift the terms of debate. Again this is the ordinary stuff of science, but demands that scientists work on many approaches which turn out not to be useful, approaches which in some sense are failures because they are never adopted. To embrace rapid learning is to embrace rapid failure.

The possibilities of creating wholly new approaches is sometimes used as a justification for curiosity-driven research. The argument is fine as long as the work focuses on problems of practical importance. To extend my analogy, what do we learn about spruce culture from studying *Arabidopsis* that we would not better learn from studying spruce? The Finns and Swedes seem to do a much better job of harnessing the power of disciplinary science to serve forestry and conservation than we do in North America.

IV. Summary and Conclusions

Conservation is more important now than it was in the past. And it will be more important in the future than it is now. Only if the world's economy stumbles will conservation slip down the social agenda. For this reason, those of use who are interested in conservation should work hard to see that economic development occurs.

That most elements of conservation fall outside formal markets demands astute public policy. In BC--and indeed nearly everywhere in North America--society has opted for "command and control" regulatory approaches. These approaches logically arise from interest-group politics, but may be extraordinarily cumbersome and costly means to achieve desirable conservation outcomes.

These regulatory approaches place significant, specific demands on science, demands which are generally consistent with the scientific process but demands which are not always met by conservation science as currently practiced. There is a need for rigourous hypothesis testing at meaningful spatial and temporal scales (perhaps through real adaptive management approaches), a need for spirited replication and debate, a need for deploying a wide range of options, and a need for "testing to failure."

We scientists generally enjoy our new importance in the policy debates. But with the increased reliance on science comes an increased risk of enormous failure. The costs of failure will be measured in a new totalitarianism where opinion and personality provide the answers that science has failed to deliver. And the costs of failure will be measured in a diminution of the natural world when inept policies derived without due attention to cause-effect relationships are religiously enforced.

THE USE OF
MOLECULAR MARKERS
IN
QUANTITATIVE GENETICS
AND BREEDING

MOLECULAR GENETICS OF GROWTH IN HYBRID POPLARS

H.D. Bradshaw, Jr. and R.F. Stettler

College of Forest Resources
Box 354115
University of Washington
Seattle WA USA 98195

Linkage maps of the *Populus* genome were used to detect quantitative trait loci (QTLs) responsible for variation in growth potential among 54 F2 offspring of an interspecific hybrid between *P. trichocarpa* and *P. deltoides*. "Major" QTLs for height, diameter, and volume growth were found on several linkage groups, accounting for approximately half the genetic variance in these traits. QTLs for morphological correlates of growth, such as cumulative leaf area, often were coincident with QTLs for the growth traits themselves. The explanatory power of individual "growth" QTLs tended to decline over time, suggesting that changes in QTL pattern and magnitude of effect may be a consequence of stand development and/or year-to-year variation in climatic conditions. QTLs detected in this small F2 family in a single growing environment were tested for their effect in two much larger F2 families grown in contrasting environments east and west of the Cascade Range. Some QTLs were found to be important across families and environments, while others were not. Those QTLs with stable patterns of expression across families and environments are logical targets for marker-assisted selection in the genetic improvement of hybrid poplar for wood, fiber, and biomass.

THE USE OF MOLECULAR MARKERS IN TREE BREEDING

S. D. Carson, T. E. Richardson, and M. J. Carson

New Zealand Forest Research Institute
Private Bag 3020, Rotorua, New Zealand

Beginning with the discovery of the structure of the DNA molecule in the early 1950s, the technology generated by molecular biology has expanded exponentially, including the development of DNA-based molecular markers. These molecular markers reveal differences in the DNA sequences among individuals. The technology is beginning to be applied in tree breeding programmes, and there is tremendous potential to augment the genetic gain that we capture through conventional tree improvement methods by better understanding the genetics of our major species, including radiata pine (*Pinus radiata* D. Don). Molecular marker technology will not replace classical tree breeding methods, but instead will result in genetic gain additional to what can be achieved by conventional methods.

Molecular markers are new tools which will increase genetic gain in production forests. Gain from conventional tree breeding methods will continue to be made in the future, but will be enlarged through

- correct identification of clonal seed orchard ramets through fingerprinting,
- increased availability of high quality control-pollinated crosses because of our ability to evaluate less expensive pollination techniques,
- certainty of parentage in planting stock by certification of identity through the Seed Certification Service,
- reduction of risk from genetic uniformity through an increased understanding of the changes in diversity brought about by domestication,
- higher gain from traditional selection and breeding techniques through better choice of breeding and selection strategies as a result of more accurate prediction models, and
- additional gain from marker aided selection on top of that attainable through traditional methods, especially for traits that are expensive to measure (for example, wood property traits).

SELECTING ON RECOMBINATION RATES IN MAIZE

Claire G. Williams

Texas A&M University
College Station, TX 77843

Recombination generates new phenotypic variability in recurrent plant breeding programs. The rate of recombination itself is genetically variable, under the control of both genic and chromosomal mechanisms. Selecting on the rate of recombination is an appealing idea which dates back to the 1920's but is being re-visited due to availability of new marker technology, an interest in preserving marker-QTL linkages and the continuing frustration of linkeage drag. I discuss results from a survey of recombination rate variability and from the development of an experimental system designed to restrict recombination. The system will ultimately be used to test the effect of restricting recombination on phenotypic traits.

NORTH AMERICAN
QUANTITATIVE FOREST GENETICS

INBREEDING DEPRESSION IN CONIFERS: IMPLICATIONS FOR BREEDING STRATEGY

Claire G. Williams[1] and Outi Savolainen[2]

[1] Texas A&M University, College Station, Texas, USA
[2]University of Oulu, Oulu, Finland

Use of selfing as a breeding tool for conifers is controversial, this topic is addressed with a review of genetic models, theory and experimental results based on a wide range of plants and animals. Some supporting evidence is available from conifer studies.

For most conifers, selfing will not be the best method for reducing inbreeding depression in small subpopulations or elite lines of deleterious alleles, sib- or random-mating is a better option in the early generations of conifer domestication. Possible exceptions are conifer species which have few lethal alleles. Few organisms have been studied which have more lethal equivalents than conifers so slower rates of inbreeding than selfing are needed initially t o prevent large losses to low offspring survival and adult fecundity.

Inbred breeding populations will also require large numbers of replicate lines and progeny per replicate because the probability of extinction for each line is expected to be high. Like maize, few valuable lines will result from selfing in the initial generations. If inbreeding depression is based on deleterious mutations then it is hypothesized to declines with stringent selection against deleterious alleles (purging). After the initial purging phase, selfing would be efficient. Advantages of selfing include perfect assortative mating, increased selection efficacy among lines and increased uniformity within lines.

Theoretical predictions for inbreeding depression in conifers have outpaced experimentation. Operational breeding programs will not provide needed data on changes in inbreeding depression but the inbreeding assumptions for breeding strategies must be tested experimentally. We advocate using *experimental inbred populations* to maximize direct use of inbreeding depression as a breeding method. It provides first-hand results and lends confidence to long-term population management decisions. The greatest value will be to reveal unforeseen problems, preventing irreversible mistakes. As an example, we outline a plan for a rapidly-cycled experimental inbred population for *Pinus taeda* L. which combines early selection, rapid screening for adult fecundity and traditional genetic testing. Inbreeding depression research is central to the success of long-term population management. It has become more powerful with integrated classical genetics-molecular approaches, accelerated breeding techniques and computer simulation models.

EFFECT OF INBREEDING ON SUPERIOR LOBLOLLY PINE

Steve McKeand and J.B. Jett

North Carolina State University
College of Forest Resources
Raleigh, NC, USA

The impact that various levels (F=0, 0.125, 0.25, and 0.5) of inbreeding have on growth, disease, and quality traits have been assessed at 6 years of age for loblolly pine *(Pinus taeda* L.) from the Atlantic Coastal Plain and Piedmont provenances in the southeastern United States. Ten second-generation clones from each of the geographic regions were sewed (F=0.5), mated to two full-sibs (F=0.25), two half-sibs (F=0.125), and two unrelated clones (F=0). Seedlings were grown in a greenhouse for five months, and then were established in field trials, four in the Coastal Plain and four in the Piedmont in 1988.

There were no major effects of inbreeding level on germination of filled seeds or on seedling growth in the greenhouse, and very few mutants were observed for any inbreeding level. After six years in the field, there was a general linear depression for growth traits and relatively minor impacts on form, disease resistance, and survival. There were highly significant family x inbreeding interactions for growth. Inbreeding depression was very severe in some family lines, while in one family inbreeding actually stimulated growth. Implications for advanced generation breeding programs and opportunities for future research are discussed.

INBREEDING EFFECTS ON THE SEED PRODUCTION, NURSERY PRODUCTION AND EARLY FIELD GROWTH OF DOUGLAS-FIR

Jack H. Woods

British Columbia Forest Service
Cowichan Lake Research Station
Mesachie Lake, BC, Canada

Inbreeding effects are of concern for both operational forestry (seed orchards, nurseries and field plantations) and breeding programs (obtaining sufficient seed, effects on testing and selection). This study uses 9 founder clones, from the south-coastal area of British Columbia, and their progeny, to create inbred levels from F=0.0 to F=0.75, where F is the inbreeding coefficient. Data were collected on filled seed production, nursery losses and early field growth.

Seed production is strongly influenced by inbreeding, with an average reduction in filledSseed per cone of about 19% per 0.1 increase in F. for F values between 0.0 and 0.5. Seed germination was slightly higher for outcross families (96%) than for inbred families (91%), but differences were not significant among inbreeding levels ranging from 0.125 to 0.5. First year nursery growth (6-15 styroblock containers) was not significantly reduced for inbreeding levels as high as F=0.5, however, mortality and culling levels avere much higher for the inbreds. Combined losses due to mortality and culling resulted in a nearly linear increase of from 23 percent for outcross families to 82 percent for self families.

Seedlings were planted on 2 test sites. Difficulties in obtaining good quality seedlings from all inbred families were experienced due to the high culling levels for the inbreds. This was successfully controlled, however, as height and root-collar diameter of planted trees at age 1 (from seed) showed no differences across the inbreeding levels from F=0 to F=0.5. As the plantation aged, significant differences in growth occurred, with a loss of 5.8% in height per 0.1 increase in F by age 5. This inbreeding effect held across all 9 common ancestor groups. Variances did not change with the different inbreeding levels.

INBREEDING DEPRESSION IN LOBLOLLY PINE: BIOMASS COMPONENTS

George R. Askew[1], Barbara McCutchan[2], and Michael Taras[1]

[1]Forest Resources, Clemson University, Georgetown, SC, USA
[2]Westvaco Corporation, Sommerville, SC, USA

Sixteen families with controlled inbreeding levels of 0 (outcross), 0.125 (maternal half-sib cross), 0.125 (paternal half-sib cross), and 0.25 (full-sib cross) were analyzed for variation in foliage, bark, and bole weight differences at age 6 years. The trees were grown at 8'x8" spacing from containerized seedlings with no chemical weed control, fertilization or irrigation. Randomly selected trees from each family were harvested, separated into their respective components and weighed green in the field. Sub-samples were collected, dried and reweighed for dry weight analyses. No correlation was found between the inbreeding levels and the dry weights or the fresh weights. Some variation among the inbreeding levels was observed with respect to several components: green limb weight, total tree weight, and dry limb weight. In those cases, the full-sib cross and the outcross were similar and the half-sib crosses differed from both the full-sib and half-sib crosses. There was no evidence, at age six, that inbreeding levels of 0.25 and less had a measurable adverse effect on the growth of loblolly pine.

LINKAGE OF MARKERS TO VIABILITY GENES CONTROLLING INBREEDING DEPRESSION

Yong-Bi Fu[1] and Kermit Ritland[2]

[1]**University of British Columbia**
Faculty of Forestry
Vancouver, BC, Canada

[2]**University of Toronto**
Department of Botany
Toronto, ON, Canada

Understanding the behaviour of purging deleterious genes controlling inbreeding depression in populations of forest trees is of both theoretical and practical importance and requires detailed genetic information on these deleterious genes. Acquiring this information requires effective genetic methods. Here we introduce a graphical, marker-based method for inferring the relative role of different genetic mechanisms (i.e., complete recessivity, partial recessivity, additivity, partial dominance, complete dominance, underdominance and overdominance) in the expression of viability genes reducing fitness in self-fertile organisms. This method requires only selfing a parent heterozygous for a number of genetic markers and analyzing the segregation pattern of marker genotypes in selfed progeny. We applied this method to data of 2577 selfed progeny of 31 plants of *Mimulus guttatus* and found that, in the chromosomal segments identified by isozyme markers, partial dominance played a predominant role in the expression of viability genes. Alternative statistical models for analyzing such data are described and their properties discussed.

COMBINING PHENOTYPIC AND MARKER DATA TO DETECT INBREEDING DEPRESSION

Brad Potts, C.M. Hardner, R.E. Vaillancourt

Collaborative Research Centre for Temperate Hardwood Forestry
Department of Plant Science
University of Tasmania
Hobart Tasmania

Eucalyptus globulus has a mixed mating system but seed set is severely reduced following self-pollinations. Selfing results in marked inbreeding depression for growth and increases variation between and within families consistent with exposure of deleterious recessive alleles. High genetic loads have been suggested from high levels of segregation distortion detected with molecular markers in a sewed F2 population. Experiments are underway to investigate (i) the correlation between self, outcross and open-pollination performance, (ii) the inheritance of self-incompatibility (iii) differences within and between populations in inbreeding depression and (iv) the level of inbreeding depression at different degrees of relationship. The relationship between spatial proximity of parents in native stands and cross success is also being examined. Seed set was not reduced in pollinations among neighboring trees suggesting that neighbors are not closely related. However, after two years in the field, depression in the growth of both selfs and nearest-neighbor crosses had begun to develop, consistent with the onset of later age inbreeding depression.

WITHIN AND BETWEEN POPULATION VARIATION FOR INBREEDING DEPRESSION IN *PINUS SYLVESTRIS*

Katri Karkkainen

Department of Genetics
University of Oulu
Oulu, Finland

The magnitude of inbreeding depression in a population is dependent on the mutation rate and on the intensity of selection against the mutations. Inbreeding depression can be reduced by increased levels of inbreeding. The average magnitude of inbreeding depression is very high in Scots pine, probably due to high mutation rates of deleterious genes. However, there are differences between individuals and populations in the level of inbreeding depression. The northernmost populations of Scots pine exhibited lower levels of early inbreeding depression than more southern ones. Differences between populations in the level of inbreeding depression may be caused by increased levels of self-fertilization in the northern populations. These results from natural populations can help to evaluate whether inbreeding can be used in the breeding programs of Scots pine.

INBREEDING DEPRESSION AND EXPERIMENTAL SELFED POPULATIONS IN WESTERN REDCEDAR *(THUJA PLICATA)*

John H. Russell[1], Rowland Burdon[2], Alvin Yanchuk[1]

[1]British Columbia Forest Service
Research Branch
Victoria, BC, Canada

[2]New Zealand Forest Research Institute
Rotorua, New Zealand

Western redcedar *(Thuja plicata)* has a high selfing rate in a seed orchard environment (65% of filled seed). As well, effective inbreeding depression (IBD) tends to be minimal in features of the early stage of the life cycle, which include cone abortion on the parents, filled seed, germination and nursery growth. However, upon field planting, IBD has become prevalent averaging approximately 7.5% for height at age 4. There is, however, some variation between clones in IBD with most traits measured, yet there was no apparent maladaptation of selfs, upon outplanting, with respect to survival, frost tolerance and form traits.

With modest, though far from negligible genetic variation in western redcedar, there is a particular call to capitalize on the theoretical advantages of inbreeding as a breeding tool. Moreover, western redcedar responds favourably to giberellins, flowering early as age one year. Thus, with the potential for short generations coupled with minimal IBD, this species is ideal for developing a model system of experimental selfed populations. This may also serve, on a pilot-scale, for operational breeding.

Select and random selfed lines with appropriate outcrossed controls are currently being developed to investigate the feasibility of using inbreeding as a tool in breeding western redcedar, and to test, in this situation, some of the assumptions underlying quantitative genetic models used for forest trees. From a base of 15 single-pair matings, 30 seedlings selected for early height and 30 random lines are to be selfed for up to 6 generations, with a 2- or 3-year generation interval. Each generation, the selfed seedlings will be outcrossed with a 28-clone polymix of comparable, current-generation, unrelated pollen. At a minimum, every other generation will be outplanted in field trials for long-term observation.

MOLECULAR GENETICS

OF

DISEASE

MOLECULAR ANALYSIS OF HOST-PATHOGEN INTERACTION OF WHITE PINE BLISTER RUST PATHOSYSTEM

Abul K.M. Ekramoddoullah

Pacific Forestry Centre
Canadian Forest Service
and
Centre for Forest Biology
University of Victoria,
Victoria, British Columbia, Canada

This study is being approached in three different ways. In one approach, i.e. pathogenesis-related proteins, resistant (Rr) and susceptible (rr) sugar pine (*Pinus lambertiana*) seedlings were inoculated with white pine blister rust (WPBR) fungus and foliar protein patterns were analyzed by 2-dimensional gel electrophoresis in conjunction with densitometry and computer-assisted gel image analysis. In resistant seedlings, both enhancement and suppression of the biosynthesis of proteins were observed. By contrast, only suppression in the biosynthesis of proteins was observed in susceptible seedlings. Two acidic proteins, 36.7 kDa and 28.1 kDa, were detected in large amounts.

The 36.7-kDa protein was suppressed in susceptible seedlings at day 3 while the 28.1-kDa protein was enhanced in resistant seedlings at day 9. In another approach, bark proteins of western white pine (*Pinus monticola*) trees displaying slow canker growth - a form of white pine blister rust resistance - were analyzed. A 10.5-kDa protein unique to this resistance type was identified and partially characterized. In the third approach, a series of monoclonal antibodies (Mabs) to WPBR was generated. Although, most Mabs were shown to be cross-reactive to western white pine proteins, two Mabs were specific to WPBR. These two Mabs will be used to identify immunocytochemically the pathogen in resistant and susceptible trees during initial infection. In addition, a "cold" protein was shown to be differentially expressed in resistant and susceptible white pine trees, indicating its potential use as a marker for resistance.

GENETIC VARIATION IN THE BLISTER RUST FUNGUS, *CRONARTIUM RIBICOLA*

E.E. White[1], M. Gitzendanner[2], B.M. Foord[1], B.Westfall[2], G. Dupper[2], P.D. Hodgkiss[2], and B.B. Kinloch[2]

[1] Pacific Forestry Centre,
Victoria, B.C.
[2] USDA Forest Service
Institute of Forest Genetics
Placerville, Ca.

Disease resistance mechanisms depend on the interaction of genes of both host and pathogen. Consequently knowledge of the genetic structure and breeding system of the pathogen is important for planning genetic improvement and deployment strategies for disease resistance in trees.

DNA markers, isozymes and virulence phenotype have been used to examine genetic variation in the blister rust fungus (*Cronartium ribicola* J.C. Fisch).

The ribosomal RNA intergenic spacer shows variation in aeciospores from individual rust cankers, indicating that the fungus is heterothallic and sexual reproduction occurs. This is confirmed by Mendelian segregation at this locus and at highly variable high copy number loci hybridizing to a genomic DNA clone. Mendelian segregation also occurs at loci defined by PCR amplification with random primers (RAPD markers), and at isozyme loci. Gene recombination can be expected in the fungus, with the potential to create new virulence phenotypes. Considerable variation occurs within populations for all molecular marker types. Virulence to the major gene resistance in sugar pine may not conform to normal patterns for nuclear inheritance.

GENETIC MAPPING OF A DOMINANT GENE FOR RESISTANCE TO WHITE PINE BLISTER RUST IN SUGAR PINE

**D.B. Neale, B.B. Kinloch, D.M. Harkins, A.D. Mix,
W.L. Nance and M.E. Devey**

**Institute of Forest Genetics
Pacific Southwest Research Station
USDA Forest Service
Albany and Placerville, CA**

Our goal is to understand the molecular basis of resistance to white pine blister rust (*Cronartium ribicola* Fisch.) in sugar pine (*Pinus lambertiana* Dougl.) and other white pines. Molecular cloning of a resistance gene from pine would greatly facilitate such investigations. One important step towards cloning a resistance gene is to genetically map the gene to be cloned. We have recently mapped a single dominant gene, R, for resistance to white pine blister rust in sugar pine[1]. The approach we used relied on three important factors: (1) the ability to assay for genetic markers in the haploid stage of the host's life cycle by using the seed megagametophyte, (2) a simple and clearly defined pathosystem[2], and (3) the use of random amplified polymorphic DNA (RAPD) markers. We used the technique of bulked segregant analysis[3] to screen 800 RAPD primers for linkage to R using DNA from megagametophytes of one teset cross (Rr x rr) and from four open-pollinated families of heterozygous (Rr) mother trees. Ten RAPD loci were found to be linked to R, the closet being within 0.9 cM. More recently, we have screened and additional 1200 RAPD primers to the four open-pollinated families to identify more tightly-linked markers. The newly identified markers will be used to construct high-resolution maps for each of the individual families. Finally, we are pursuing a strategy to clone R based on homology to other plant resistance genes that have recently been cloned.

(1) Devey et al. (1995) *Proc.Natl.Acad.Sci.* 92:2066-2070.
(2) Kinloch et al. (1970) *Science* 167:193-195.
(3) Michelmore et al. (1991) *Proc.Natl.Acad.Sci.* 88:9828-9832

DEVELOPMENT OF DEFENSE-RESPONSE RELATED PROBES AS DNA MARKERS IN CONIFERS

Susan M. Baker and Eleanor E. White

Canadian Forest Service
Pacific Forestry Centre
Victoria, BC, Canada

Molecular markers associated with defined resistant phenotypes provide powerful tools which can be used to aid in early selection of stock in tree improvement programs. Since in most of our forest species we do not have defined resistant phenotypes, one approach to isolating useful markers for resistance would be to study genes known to be involved in other plant defense responses. In our lab we have been developing defense-response related DNA probes which may be useful molecular markers in our tree improvement programs.

Degenerate oligoneucleotide primers based on consensus sequences found in two defense-response related genes, chalcone synthase and phenylalanine ammonia lyase, were used to amplify western white pine total DNA. The amplification products obtained were cloned and have been used to study the genomes of several conifer species.

Chalcone synthase occurs as a high copy number, multi-gene family in the conifers probed. It has been found to be polymorphic within and between species. Phenylalanine ammonia lyase appears to occur at low copy numbers and may be only a single gene.

GENERAL TOPICS

OF

TREE IMPROVEMENT

EFFECTS OF SELECTION FOR INCREASED GROWTH RATES ON THE ANNUAL DEVELOPMENTAL CYCLE OF COASTAL DOUGLAS-FIR

S.N. Aitken

Department of Forest Science
Oregon State University
Corvallis, Oregon, USA 97331

Artificial selection for faster tree growth can result in indirect responses in genetically-correlated adaptive traits. The potential impacts of selection for growth on spring and fall cold hardiness, bud and shoot phenology, and frequency of second flushing in breeding populations of coastal Douglas-fir (*Pseudotsuga menziesii* var. *menziesii*) are examined. In one population in the Coast Range of Oregon, growth is uncorrelated with spring and fall hardiness, and bud phenology. However, in a Cascade population, faster-growing genotypes tend to deacclimate earlier in the spring and break bud sooner than slower-growing genotypes. Studies with other populations indicate a positive genetic correlation between frequency of second flushing in late summer and growth rate, and also between the occurrence of second flushing and cold injury, thus selection for growth may result in an increase in second flushing and with it increased risk of fall cold injury. Selection and deployment strategies for mitigating unfavorable effects of selection for growth on these annual developmental cycle traits are discussed.

A COMPARISON OF HERITABILITY ESTIMATES BETWEEN GROWTH CHAMBER AND FIELD GROWN *POPULUS TREMULOIDES* STECKLINGS FOR GROWTH AND GAS EXCHANGE TRAITS

Barbara R. Thomas, S. Ellen Macdonald and Bruce P. Dancik

University of Alberta
Department of Renewable Resources
751 General Services Building
Edmonton, Alberta, T6G 2E1, Canada

Thirty clones of trembling aspen (*Populus tremuloides* Michaux) were collected from across Alberta and multiple cuttings were obtained from the roots for each clone, under greenhouse conditions. These stecklings were grown at two field sites (High Level and Calgary) for two years where caliper and gas exchange measurements were taken four times. A three month growth chamber experiment was also conducted on the same clones with conditions programmed to mimic 30 year average temperatures from the field sites. A comparison of broad-sense heritabilities will be presented for caliper, net assimilation and water use efficiency for both growth chamber and field grown material.

AGE TRENDS IN DOUGLAS-FIR GENETIC PARAMETERS

G.R. Johnson and R.A. Sniezko

U.S.D.A. Forest Service
Forestry Sciences Laboratory
Corvallis, OR, USA

Growth data from 19 open-pollinated progeny tests were used to examine changes in genetic parameters over time. Twelve progeny tests from the Vernonia breeding cooperative (NW Oregon) were assessed at ages 7, 10, 15, 20, and 25 years from seed. Seven progeny tests from the Umpqua breeding cooperative (coastal Oregon) had data available to age 20. Narrow sense heritabilities increased in a curvilinear fashion to age 25 for DBH. Narrow sense heritabilities for height increased from age 7 to 10 and then changed very little. DBH and height heritabilities were very similar, except at age 7 when height had larger heritabilities. Age-age correlations of age-15, -20 and -25 data were all greater than 0.90. As expected, the age-7 and -10 data had weaker correlations with the older data. Examinations of selection efficiency showed age-15 to optimize gain per year when selecting for age-25 gain. Age 15 is optimal from a practical point since it is not always possible to obtain good information on diameter and tree form at earlier ages. In addition, genetic correlation patterns across sites did not show any significant change after age 15.

THE QUALITY OF YOUNG SCOTS PINE STEMS IN PREDICTING THE BREEDING VALUE

Martti Venäläinen[1], Tapani Pöykkö[2], Juhani Hahl[2]

[1]The Finnish Forest Research Institute
Punkaharju Research Station
FIN-58450 Punkaharju, Finland
[2]Foundation for Forest Tree Breeding
Haapastensyrjä Breeding Centre
FIN-12600 Läyliäinen, Finland

First instructions for grading exported sawn goods were established in Finland already in 1936. Since 1960 the instructions have been the same in the Nordic countries, the revised version of them was released in 1994. The main factor causing lower quality is the knots.

The wood quality has been one objective of the Scots pine breeding programme from the beginning, year 1947. Numerous components of external and internal quality and the inheritance of measurable traits have been studied. However, the breeders have missed a method which would be quick, cheap and accurate enough to screen a sample of 250 000 trees, the progenies of 1 500 plus-trees.

In this presentation the nordic standards for the quality of Scots pine timber will be briefly described. A method to predict the quality and relative value of a stem, 40 - 50 years from now will be introduced. This method is arising from the concept of working ideotype and it is based on the ocular assessing of the branches and stem form of 8 - 12 meters tall, 15 - 20 years old trees. Possibilities to use this method to predict the breeding value of the candidates for the open-nucleus breeding populations will be discussed.

GENERAL

BIOCHEMICAL

FOREST

GENETICS

MULTILOCUS MARKER-BASED INFERENCE OF EPISTASIS FOR GENES INFLUENCING INBREEDING DEPRESSION IN *MIMULUS GUTTATUS*

Yong-Bi Fu and Kermit Ritland

Department of Botany, University of Toronto
Toronto, Ontario M5S 3B2 Canada

We introduce a multilocus, marker-based method for inferring fitness interactions between loci affecting inbreeding depression in self-fertile organisms. It is based upon selfing a parent heterozygous for several unlinked, codominant markers, and analyzing the fitness of multilocus progeny genotypes. If loci causing inbreeding depression are partially or completely linked to the marker loci, then viability selection is manifested by distorted segregation of markers, and fecundity selection by dependence of the fecundity character upon the marker genotype. To characterize this selection, fitness is regressed on the proportion of loci homozygous for markers linked to deleterious alleles; epistasis is then detected by non-linearity of the regression. Other modes of selection can be incorporated via a bivariate regression involving both homozygote and heterozygote marker genotypes. The advantage of this marker-based approach is that "purging" is minimized and specific chromosomal segments are identified; its disadvantage lies in low statistical power, particularly when linkage is not strong. We applied this method to empirical data from *Mimulus guttatus* and found the predominance of multiplicative fitness interactions influencing fecundity, as well as evidence for synergism reducing viability.

ALLOZYME GENOTYPE AND GROWTH RATE IN DOUGLAS-FIR -- REVISITED

Bruce C. Bongarten[1], Nicholas C. Wheeler[2] and Keith S. Jech[2]

[1]Warnell School of Forest Resources
The University of Georgia
Athens, GA, USA;
[2]George R. Staebler Forest Research Center
Weyerhaeuser Company
Centralia, WA, USA

Weyerhaeuser's first-generation, full-sib progeny tests of Douglas-fir afforded the opportunity to examine the relationship between allozyme genotype and growth rate in a population that a) is wholly outcrossed, b) has minimized environmental variation, c) contains a large number of parents (over 300) and progeny (over 50,000), d) is replicated (bi-parental and diallel testing programs were conducted and analyzed independently), and e) is nearing half rotation age (15-17 field seasons).

Using full-sib family means and expected allozyme genotypes of progeny determined from parental genotypes, we found that the presence of rare alleles (frequency less than 0.05) was associated with reduced growth rate, particularly when two rare alleles were present at a single locus. The expected volume loss associated with the presence of two rare alleles at a locus was approximately 5%. On the other hand, alleles that occur with intermediate frequency (0.08 to 0.25) appeared to be slightly heterotic when combined with a common allele (frequency over 0.5). Overall, allozyme genotypes at 28 loci accounted for less than 1% of the variation in mean full-sib family volume. The model, and the aforementioned elements of it, were, nevertheless, statistically significant.

GENETIC DIVERSITY OF OLD-GROWTH AND REGENERATED WHITE SPRUCE - IMPACT OF SILVICULTURAL MANAGEMENT

O.P. Rajora, M.S. Sekhon and B.P. Dancik

Department of Renewable Resources
University of Alberta
Edmonton, Alberta, Canada T6G 2H1

For ecosystem sustainability and stability, it is essential to maintain genetic diversity in forest populations. Forest practices relying on natural and/or artificial regeneration systems can significantly affect genetic variability in subsequent generations. We attempted to determine the impacts of silvicultural practices on maintaining genetic diversity in white spruce (*Picea glauca* (Moench) Voss). Random amplified polymorphic DNA analysis was conducted to determine the genetic diversity levels of adjacent natural old-growth, naturally-regenerated and planted white spruce stands, and open-pollinated progeny of 30 selections made for tree improvement purposes. The estimates of genetic variability parameters suggested reduced genetic diversity in plantations and tree improvement material. The genetic variability of the old-growth populations and natural regeneration was comparable. Implications for alternative silvicultural practices and gene conservation will be discussed.

PROTOPLAST FUSION IN LARCH (*LARIX SP.*)

R. Pattanavibool[1], K. Klimaszewska[2] and P. von Aderkas[1]

[1] Centre of Forest Biology
Department of Biology
University of Victoria
Victoria, BC, Canada
[2] Petawawa National Forestry Institute
Chalk River, Ontario, Canada

Protoplast fusion is a powerful technique used to create plant cells with novel nuclear or cytoplasmic genetic constitutions. Moreover, larch embryogenic tissues are easily grown as suspensions of embryonal masses which provide material for protoplast isolation and regeneration. We describe experimental putative somatic hybrids between a diploid embryogenic line of tamarack (*L. laricina*) and a line of hybrid larch (*L.* x *eurolepis*). The protocol for protoplast fusion will be outlined and discussed. There are many advantages in choosing these two species: 1) they are naturally compatible species, give us a high possibility to recover compatible heterofused cells, 2) each has a unique DNA banding pattern which is useful for detection of the hybridity of heterofused cells and, 3) they differ in their behaviour *in vitro*. Tamarack is able to produce mature somatic embryos while under the same conditions, the hybrid larch is incapable. The selection system is based on complementation of metabolic inhibition of tamarack and the lack of ability to produce mature embryos of the other source. Ideally, only the heterofused cells regenerate.

QUANTITATIVE

GENETICS

OF

DISEASE

THE EVOLUTION OF FUSIFORM RUST DISEASE: HOST RELATIONSHIP ON SLASH PINE AND NEARBY TROPICAL RELATIVES

R.C. Schmidtling, R.L. Doudrick, and C.D. Nelson[1]

Southern Institute of Forest Genetics (SIFG)
USDA Forest Service
Saucier MS 39574.
[1] Present address: International Paper Co.
Bainbridge, GA 31717

Seedlings of eight sources of slash pine (*Pinus elliottii* var. *elliottii* and var. *densa*) and two each of Caribbean pine (*Pinus caribaea* var. *caribaea*) and West Indian pine (*Pinus occidentalis*) were inoculated with *Cronartium quercuum* f. sp. *fusiforme*, the causal agent of fusiform rust disease. Percent infection and pycnial sporulation of *C. q. fusiforme*, differed significantly within sources of slash pine and among species. Percent infection was highest on seedlings from the most southern source of slash pine from Florida (92%) and lowest on *P. occidentalis* (30%). Among slash pine, more pycnia sporulation occurred on seedlings from sources nearest the origin of inocula, with the exception of those of the source from furthest south that showed abundant sporulation of pycnia similar to the nearby sources of *P. c. caribaea*. These two sources occur outside the present natural range of the *Quercus* species which are the alternate host. No sporulation occurred on *P. occidentalis*. Because sporulation is necessary for the spread of the disease, the results suggest strong selection on natural inoculum for infection and fertility among and within sources of slash pine. Breeding strategies currently recognize families of slash pine that minimize damage due to fusiform rust disease but new strategies might consider limiting pathogen reproduction and utilizing germplasm from resistant south Florida sources.

ANALYSIS OF RISK IN CLONAL FORESTRY USING THE TIME-TO-FAILURE MODEL

James H. Roberds

USDA Forest Service, Southern Research Station
Raleigh, NC, USA

The time-to-failure model (Bishir and Roberds 1994, Math. Biosci. 125:109-125) is a stochastic model developed to study effects of a number of variables on risk of plantation failure in clonal forestry. Number of clones deployed in plantations is one such variable that managers have at their disposal to control risk of failure due to an unforeseen catastrophic event. Using the time-to-failure model, the effect of increasing the number of clones used has been investigated and some general results obtained from this analysis are reported in this presentation. Theoretical developments are described which demonstrate that 30-40 clones per plantation provide approximately the same protection from risk as large numbers of clones.

NATURAL SELECTION OR INTROGRESSION? GEOGRAPHIC VARIATION OF LODGEPOLE PINE RESISTANCE TO WESTERN GALL RUST

Harry X. Wu and Cheng C. Ying

Research Branch
B.C. Ministry of Forests
31 Bastion Sq.
Victoria, B.C.
V8W 3E7

Incidence of western gall rust (*Endocronartium harknessii* (J.P. More) Y. Hiratsuka) was investigated in a lodgepole pine (*Pinus contorta* spp. *latifolia*) provenances/families trial in Prince George, British Columbia. The most interesting findings in this study are (1) the strong relationship between pest incidence and procenance distance to western limit of jack pine and provenances from jack-lodgepole pine hybrid zone are very resistance to western gall rust;
(2) provenances from relative high elevation are more susceptible to western gall rust.
(3) provenances from gall-ridden regions had relative high resistance to gall rust. The question is whether resistant developed in lodgepole pine are due to jack pine introgression or natural selection or both. The possibility of these evolution forces on shaping western gall rust resistant was evaluated and corresponding breeding strategies to deal with different evolution mechanism responsible for developing gall rust resistant were discussed.

ESTIMATION OF GENETIC PARAMETERS AND BLUP BREEDING VALUES FOR PINE NEEDLE BLIGHT (*DOTHISTROMA SEPTOSPORA*) AND STEM DEFORMITY IN *PINUS RADIATA* IN SOUTH-EASTERN AUSTRALIA

I.R. Bail[1] and S. F. Jarvis[2]

[1] : Centre for Forest Tree Technology
Victoria, Australia
[2] : Southern Tree Breeding Association
South Australia, Australia

Pine needle blight caused by the fungus *Dothistroma septospora*, and severe Stem Deformity caused by establishment on highly fertile ex-agricultural soils, have had a major effect on the productivity of *Pinus radiata* plantations in south-eastern Australia. (Marks et al. 1989, Aus. For. 52:10-19, Bail and Pederick 1989, Aus. For. 52:309-320, Hopmans et al., Plant and Soil in press). Both disorders have expressed strong genetic control, and tree breeding is seen as the integral part of a long-term solution to both these problems. Extensive progeny trials were established between 1985 and 1991 on ex-agricultural sites where a high incidence of infection by *Dothistroma septospora* and severe Stem Deformity were expected. The trials were assessed progressively from age 3 years onwards when symptoms were clearly displayed. Genetic parameters are essential elements for the development of an appropriate breeding strategy, and have not been estimated for these traits from such an extensive trial series in Australia. This study reports a combined analysis of all sites using an individual tree mixed model BLUP analysis. The parameters reported here will be used in the development of breeding objectives for the rapid improvement in resistance to both traits for members of the Southern Tree Breeding Association.

GENERAL

MOLECULAR

GENETICS

CLONING, SEQUENCING, AND MAPPING THE PHYTOCHROME GENE IN DOUGLAS-FIR

Kimberly A. Marshall and David B. Neale

Institute of Forest Genetics
USFS, 800 Buchanan Street
Albany, CA 94710

We are studying the structure and expression of phytochrome (phy) genes in Douglas-fir (*Pseudotsuga menziesii* (Mirb.) Franco) to satisfy our long-term goal of understanding phytochrome's role in controlling bud phenology and shoot elongation. To clone a phytochrome gene, we constructed primers for PCR based on consensus phytochrome DNA sequences of oat, zucchini, and Arabidopsis (Sharrock, Lissemore and Quail, Gene 47: 287-295 and Sharrock and Quail, Genes and Development 3:1745-1757). These primers were used to amplify 369 and 1104 bp products from Douglas-fir genomic DNA. Subsequently, we amplified the 369 bp fragment from total RNA of dark-grown Douglas-fir seed. The 369 and 1104 bp fragments have been cloned and their DNA sequences have been determined. Sequence comparisons confirmed that these PCR products are most like phytochrome A, since they contain conserved domains unique to the phytochrome A genes published in Genbanks' sequence database. Southern hybridizations of genomic DNA with the 1104 bp phy PCR probe suggests that there are at least six loci. This exceeds the number of phytochrome genes so far reported for all other species to date, however, we do not yet know how many of the Douglas-fir phy loci are functional. A Northern blot of dark-grown seed mRNA, hybridized with the 369 bp product, revealed a single band at approximately 4000 base pairs, the expected size of the phy mRNA. In the future, we hope to clone other phytochrome genes in Douglas-fir and use these as probes to study light-regulated and tissue-specific expression of phytochrome genes.

IDENTIFICATION OF QUANTITATIVE TRAIT LOCI CONTROLLING PHENOLOGY TRAITS IN COASTAL DOUGLAS-FIR USING MOLECULAR MARKERS

Jermstad, K.D.[1], Bassoni, D.L.[1], Jech, K.S.[2], Wheeler, N.C[2]. and Neale, D.B.[1]

[1]Institute of Forest Genetics
PSW-USDA Forest Service
Placerville, CA
[2] Weyerhaeuser Company
505 N. Pearl Street
Centralia, WA

We have a long-term interest in identifying the genes controlling bud phenology in coastal Douglas-fir [*Pseudotsuga menziesii* (Mirb.) Franco var. *menziesii*]. We are using a map-based approach for locating these quantitative trait loci (QTL) for bud phenology in a 3-generation outbred pedigree. Clonal replicates have been made from 250 progeny and outplanted to test plantations in Washington and Oregon. Phenotypic data is currently being recorded for date of bud flush, date of bud set, increment growth and lammas growth. An additional 48 progeny from the same mating has been utilized for the construction of a low-density linkage map using restriction fragment length polymorphisms (RFLPs) and random amplified polymorphic DNAs (RAPDs). One hundred-fifty RFLP loci and 28 RAPD loci have thus far been analyzed for linkage. Multi-point linkage analysis with JoinMap software Version 1.4 (Stam, 1993) using a LOD score of 3.0 resulted in 14 linkage groups, with the largest group comprised of 10 marker loci. Our strategy is to select markers at evenly distributed intervals for application to the larger set of 250 field progeny for detection of QTL controlling bud phenology.

QUANTIFYING NATURAL HYBRIDIZATION BETWEEN THE GENETICALLY CLOSELY RELATED BLACK SPRUCE AND RED SPRUCE WITH SPECIES-SPECIFIC RAPD FINGERPRINTS

M. Perron and J. Bousquet

Centre de recherche en biologie forestière
Faculté de foresterie et de géomatique
Université Laval
Sainte-Foy, Québec, Canada G1K 7P4

Species-specific molecular markers were designed to assist in the identification of closely related black spruce (*Picea mariana* [B.S.P.] Mill.) and red spruce (*P. rubens* Sarg.) in northeastern North America. These markers were then used in natural populations of the sympatric zone to assess the levels of interspecific hybridization. The species-specific genetic markers were developed using RAPD and a combination of bulk sample and individual tree analyses of trees from outside the sympatric zone. Most of the markers obtained were shared by the two species, but a small number of markers were found monomorphic or nearly monomorphic and specific to either *P. mariana* or *P. rubens*. These markers remained species-specific when F_1 progenies derived from independent intraspecific crosses were screened. These markers were then found to co-segregate in hybrids derived from independent interspecific crosses here used as controls. For the survey of natural hybridization, trees were sampled from 14 natural stands where the two species coexist and from 7 provenances from outside the sympatric zone here used as controls. The trees were scored for qualitative morphological traits and RAPD markers. While no hybrids were observed in control populations, hybrids were found within populations from the sympatric zone, and their frequency was variable.

ASSOCIATIONS OF NUCLEAR, CHLOROPLAST, AND MITOCHONDRIAL VARIANTS WITHIN POPULATIONS OF JACK PINE (*PINUS BANKSIANA*) AND LODGEPOLE PINE (*PINUS CONTORTA*)

D.B. Wagner[1], T. Li[1], J. Dong[1], D.R. Govindaraju[1], R.C. Hamelin[1], R.N. Patel[1], and B.P. Dancik[2]

[1]Department of Forestry
University of Kentucky
Lexington, KY, USA
[2]Department of Renewable Resources
University of Alberta
Edmonton, AB, Canada

We have studied nuclear-dicytoplasmic associations among nuclear (Mendelian), chloroplast (paternal), and mitochondrial (maternal) markers in six natural populations of jack and lodgepole pines in Alberta, Canada. A total of 2520 individuals were sampled, representing two allopatric lodgepole pine populations, two putatively allopatric jack pine populations, and two populations in a sympatric region of natural hybridization. Allozymes served as nuclear markers, while DNA restriction fragment length polymorphisms were employed as organellar markers (Dong and Wagner 1994, Genetics 136:1187-1194). Surprisingly, we observed mitochondrial variants typical of jack pine in only one of the two putative jack pine populations (and not at all in sympatry). All mitochondrial variants found in the other five populations were typical of lodgepole pine. The unusual jack pine population that contains lodgepole pine mitochondrial variants, as well as the two sympatric-region populations, all harbor chloroplast variants of both species. In contrast, the chloroplast variants observed in the other three sampled populations appear in each case to be those only of the expected species. These results, together with the within-population spatial distributions of variants and cytonuclear associations that will be reported, suggest genetic interactions that constrain hybridization and introgression between jack and lodgepole pines. Such interactions may have general implications for germplasm conservation and interspecific gene transfer programs.

GENERAL TOPICS IN FOREST GENETICS

THE EFFECTS OF EMBRYO COMPETITION WITH MIXED MATING ON THE GENETIC LOAD

R. G. Latta

Dept. of E.P.O. Biology
University of Colorado
Boulder, Colorado, 80309, USA

Competition among developing embryos is thought to favour outbred progeny. However, removal of inbred offspring may prevent the expression of deleterious mutations as homozygotes, and shelter such mutations from selection, permitting them to increase in frequency. This possibility was assessed using models of genetic load. The effect of embryo competition on load depends upon the characteristics of the mutations involved. Mutations which are not expressed in the embryo (e.g., chlorophyll deficiency mutations) are sheltered from selection by the removal of inbred embryos, and increase in frequency relative to the case without embryo competition. Moreover, if embryo competition is discontinued after several generations, expression of the accumulated load causes a temporary, but pronounced drop in fitness. By contrast, mutations which are expressed in the embryo may affect the outcome of competition, and thus experience an additional episode of selection relative to the case with no competition. Genetic load at these loci may thus be reduced, although the exact outcome depends upon the expression of the mutations in heterozygotes. The mechanism of embryo competition must be known before the long term effects of management practices which enhance embryo competition (e.g. Supplemental Mass Pollination) can be predicted.

POLLEN SCAVENGING IN SPRUCE AND EVOLUTION OF THE CONIFER POLLINATION DROP

C. John Runions * and John N. Owens

Centre for Forest Biology
University of Victoria
P.O. Box 1700
Victoria, British Columbia, Canada, V8W 2Y2

The pollination drop of interior spruce (*Picea glauca* or *P. engelmannii* and their hybrid) is large enough to collect pollen adhering to the ovule integument as well as adjacent surfaces of the seed cone. Scavenging of pollen from cone surfaces adjacent to the integuments is sometimes facilitated by rainwater which can float pollen into the opening of the micropyle. This observation lead to the new hypothesis that rainwater may have functioned in ancestral conifers like the pollination drop does in modern ones. Pressure for selection of a secreted pollination drop, required in pollen scavenging, would have mounted once ancestral conifers radiated into or became remnant in more arid regions where rainfall was a less certain feature of the environment. Cladistic analyses of Pinaceae based on morphological characters agree with those based on genetic analysis only when the pollination drop is viewed as derived in certain genera rather than ancestral and lost in certain other genera. This condition is satisfied if rainwater, instead of a pollination drop, is considered a component of the pollination mechanism in Permian conifers.

THE EFFECT OF NURSERY ON EARLY GROWTH OF SPRUCE SEEDLOTS

C.D.B. Hawkins

Red Rock Research Station
R.R. #7, RMD #6
Prince George, BC, Canada V2N 2J5

Seed orchard and natural stand collection (wild) interior spruce (*Picea glauca, P. engelmannii,* and their hybrids) seedlots were cultured at seven nurseries in British Columbia in 1994. A seed orchard and a wild seedlot were paired from each of three BC collection zones. Two nurseries were in the lower mainland, two were in the southern interior, and three were in the central interior of BC. Culture was unique to a nursery. A summer crop of all seedlots, from all nurseries but one, was planted in the first week of July 1994 at two sites. One was near Salmon Arm and the other near Prince George, BC. A fall crop, all seedlots and nurseries, was placed in storage and planted at both sites in early May 1995. Nursery-induced morphological variation by seedlot is described. Early plantation survival and performance by seedlot is discussed with respect to nursery.

TOWARDS *IN VITRO* FERTILIZATION IN CONIFERS: I. INDUCTION OF POLLEN TUBES BY MEGAGAMETOPHYTE EXTRACTS

Tok Takaso, Patrick von Aderkas, John Owens & Nicole Dumont-BéBoux

Centre for Forest Biology
Department of Biology
University of Victoria,
Victoria, British Columbia, V8W 2Y2 Canada

An ovular secretion, which we consider essential to sexual reproduction, is examined in *Pseudotsuga menziesii*. This secretion begins soon after egg formation, about 1 week before fertilization. Secreted fluid fills the micropylar canal and causes: 1) pollen transfer from the distal end of the micropylar canal to the nucellar apex; 2) dissolution of intine materials; 3) pollen distortion that may relate to prezygotic selection; and, 4) induction of pollen tubes. The origin of the secreted fluid is interpreted from an experiment using homogenates. Elongated pollen supplied with a homogenate of the megagametophyte showed immediate movement of starch grains and body cell and some pollen formed a pollen tube. Only a few or no pollen responded to homogenates of the nucellus or the integument. If ovules are dissected before the megagametophyte secretion and kept in a humid place, the micropylar canal becomes filled with fluid. This fluid is an artifact resulting from dissection, but it also induces pollen tubes. A few examples are shown of cultured pollen tubes penetrating neck cells of Douglas fir archegonia of cultured megagametophytes.

ASEXUAL PROPAGATION FOR JACK PINE GENETIC IMPROVEMENT

Jerome I. Klein[1], Campbell G. Davidson[2], and Robin D. Browne[2]

[1]Natural Resources Canada
Canadian Forest Service
Winnipeg, MB
[2]Agriculture and Agri-Food Canada, Research Centre
Morden, MB

Asexual propagation can enhance gain capture in tree improvement programs by enabling use of controlled pollination in the production population. Use of this strategy for jack pine has been impeded by lack of success from asexual propagation. A propagation protocol able to produce 50 plants from 1 seed in 4 years, would allow meeting the jack pine stock requirements for southeastern Manitoba with 50,000 control-pollinated seeds, harvestable from pollinations on 250 seed orchard trees. Techniques were developed that provided numbers of proliferated dwarf shoots and rooting percentages that come close to reaching the set goal. Tissue culture shoots from cotyledon explants, showing spontaneous rooting, present another promising approach. Rooted cuttings appeared not inferior to seedlings in field performance, which will be formally tested in a field trial with full-sib families, clones in families, and propagation methods as treatment factors. Canadian Forest Service funding of this work will end in 1996, but provincial or private-sector sponsors would have a high likelihood of an attractive return on their investment.

BREEDING

STRATEGIES

FULL-SIB FORESTRY IN PLANTATION CONIFERS

M.J. Carson and D. Lindgren

New Zealand Forest Research Institute Ltd.
Private Bag 3020
Rotorua, New Zealand

The concept of full-sib forestry (FSF) is defined as 'both seed and pollen-parent are controlled and known for plants used for forestry'. Full-sib forestry can be either a precursor of, or substitute for clonal forestry, and can be accessed using cost-efficient controlled-pollination and vegetative propagation techniques. Compared to the use of open-pollinated orchard stock, FSF offers many advantages through:

- Greater pedigree control in production and deployment.
- Increased flexibility, for more rapid and specific responses to changing plant demands, and for fine-tuning of families for alternative sites, management systems and end-uses.
- Reduced dependence on seed orchards.
- Ability to exploit additional sources of genetic and non-genetic variation for increased gains.

FSF can be implemented from almost any breeding programme, but breeding strategies may also be refined to optimise gains from FSF. Potential changes include:

- Reductions in numbers of sublines.
- Increased use of small, multiple populations ("elites").
- Improved coordination of archiving, progeny testing and deployment.
- Increased emphasis on fast multiplication of pollen, seed and vegetative sources.
- Greater utilisation of specific combining ability, and between-family variance.
- More efficient deployment of single genes.

THE EFFICIENCY OF MULTIPLE ELITE POPULATIONS

P.A. Jefferson, S.C. Concheyro,
D. Garrick and L.D. Gea

New Zealand Forest Research Institute Ltd.
Rotorua, New Zealand

ABSTRACT

Previous deterministic **single-trait** simulations of genetic gain in small (20-50 parent) elite versus large main breeding population sublines have shown somewhat higher gains from production populations derived from the small elite versus large main sublines.

In this study stochastic simulation of gains in **multiple traits** in small elite and large main breeding population sublines have been undertaken to confirm the trends from the deterministic single-trait simulation. The stochastic simulations involve traits showing both positive and negative genetic correlations. Gains per unit time are calculated for each trait for several different strategies.

OPTIMIZING SUBLINE SIZE FOR BREEDING POPULATIONS

L.D. Gea, P.A. Jefferson, D. Lindgren, T.M. Mullin,
C.J.A. Shelbourne

Sublining constitutes one of the critical features on the organization of a breeding population. Subdividing the population reduces the impact of genetic drift, as sampling of gametes within each subline is independent of that in the remainder of the population and different alleles become fixed in the different groups.

Since the accumulation of inbreeding is restricted to within sublines orchard parents selected from different sublines will always be unrelated. Breeding schemes with small sublines are more efficient in preserving status effective number through a large number of generations than breeding systems with large sublines.

Trade-off between diversity (status number) and gain for sublines sizes from 4 to 200 over a number of generations have been analysed and their implications for managing breeding populations discussed.

Very small sublines increase the aggregated status number of the breeding population. Medium to large size sublines showed a comparatively small reduction in aggregated status number over generations but showed larger increases in gain compared with small sublines. Inbreeding in small sublines becomes so big that it is likely to cause fertility problems and disturb selection considerably. Small sublines will probably not be a sustainable long term breeding strategy.

DIVERSE NUCLEUS BREEDING: SELECTION IN DIFFERENT DIRECTIONS FROM ONE CENTRAL MULTIPLE-TRAIT SELECTED BREEDING POPULATION

Hubert Wellendorf

Hørsholm Arboretum, Denmark

A new structure of forest tree breeding populations is suggested as an extension of the nucleus breeding concept. Starting point is a large breeding population in which a balanced multi-trait selection scheme is applied. From this central population, a restricted number of nucleus or elite populations are created with diverse breeding objectives, including one with the original multiple-trait criteria.

Examples of diverse breeding objectives might be resistance against specified damaging agents, wood quality concerning well-defined traits or adaptation to adverse environmental 'niches.' The relevance of such a system is dependant of the general forestry and tree improvement situation in actual regions. Forestry may benefit from such a system, as they can obtain more specialized strains from diverse nuclei, including the one with the balanced breeding objectives. The system will fit especially well into a differentiated type of forestry where environmental mapping is applied.

Combining gains from two specialized nuclei is a challenge for marker aided selection - MAS. Once a restricted number of quantitative trait loci - QTL - are mapped for the specified traits in a few outstanding nuclei-members, screening for the rare favourable combinations in between-nuclei crosses appears to be feasible. Follow-up by some type of vegetative propagation as for instance somatic embryogenesis might be necessary for large-scale application.

SUMMARY OF A SLASH PINE (*PINUS ELLIOTTII* VAR. *ELLIOTTII*) BREEDING STRATEGY FOR THE CONTINUED IMPROVEMENT OF ITS F1 HYBRID WITH CARIBBEAN PINE (*PINUS CARIBAEA* VAR. *HONDURENSIS*).

M.J. Dieters, D.G. Nikles, and P.G. Toon

Queensland Forest Research Institute
MS 483 Fraser Rd., Gympie, Qld. 4570, Australia

The F1 hybrid between slash and Caribbean pines has proven to be the superior taxon for planting in south-eastern Queensland (Australia); exhibiting fast growth, wide adaptability, resistance to wind throw, and desirable wood properties. Similar results have been demonstrated by experimental plantings of this hybrid in many other locations in the tropics and sub-tropics.

The breeding strategy for slash pine in Queensland has two unusual aspects. Firstly, in Queensland, slash pine is no longer grown in commercial plantations, and so the breeding program is directed solely towards the production of improved F1 hybrids with Caribbean pine. Secondly, in contrast with traditional programs which use multi-clonal open-pollinated orchards to provide seed for commercial plantations, the F1 hybrid is currently propagated commercially via tested full-sib families. Further, it is anticipated that by the year 2005 the entire planting program will be based on tested F1 clones. Therefore, the breeding strategy presented is targeted towards the production of superior F1 hybrid families for use in clonal forestry. Some key elements of this strategy are: i) an initial screening of parents for general hybridising ability, ii) a multiple population structure, iii) an elite nucleus, iv) vegetatively multiplying the base population to improve within family selection, v) the use of BLP/BLUP to combine both pure- and hybrid-breed information, and v) a sub-project to conserve genetic diversity.

ABSTRACTS OF
WORKSHOP CONTRIBUTIONS

Genetics
of
Wood Quality

PHENOLOGICAL VARIATION IN HEIGHT AND DIAMETER GROWTH, AND ITS RELATIONSHIP TO WOOD DENSITY IN PROVENANCES AND FAMILIES OF LOBLOLLY PINE.

K.J.S. Jayawickrama, McKeand, S.E. and Jett, J.B.

North Carolina State University
Raleigh, NC 27695–8002

This study includes eight open–pollinated families from each of four different provenances in a trial (two locations) in southwest Georgia. The provenances are: Atlantic Coastal Plain, Gulf Hammock (FL), Lower Gulf Coast and Upper Gulf Coast. Our goal is to see how wood properties are affected by the timing of initiation and cessation of height and diameter growth. The trees were measured from summer to fall in 1993 and 1994 (when the trees were in their fifth and sixth growing seasons) and the cambium of each tree wounded weekly in late summer and early fall. These wounds provide a record of whether earlywood or latewood was being formed at the time of wounding.

There were very significant differences between provenances for the date of cessation of height growth in fall. The fast growing Gulf Hammock provenance grew the longest while the slowest growing Upper Gulf source was first to stop growing. Provenances were also different for the date of cessation of diameter growth, and the order was the same as for height. Families within provenances were different for cessation of both height and diameter growth. Microscopic evaluation of wood sections is in progress and results will be presented at the meeting.

DNA MARKERS FOR SELECTING HIGH WOOD DENSITY IN DOUGLAS-FIR

J.E. Carlson and Hesham Agrema

Biotechnology Laboratory and Department of Forest Sciences
University of British Columbia
Vancouver, B.C. Canada V6T 1Z3

We constructed single-tree genetic linkage maps for two important clones from Jack Wood's coastal Douglas-fir tree improvement program, clones DF60 and DF69, using RAPD markers and haploid megagametophyte DNA. Data on quantitative traits such as wood density were collected over a 15 year period for F1 progeny in reciprocal cross families from clones DF60 and DF69, by the BC Ministry of Forests. The genetic linkage map for clone DF60 contained 132 linked DNA markers evenly distributed over 2143 cM of 13 linkage groups. The map for clone DF69 contained 153 linked DNA markers evenly distributed over 2335 cM on 13 linkage groups. Markers from these single-tree maps were moved onto a map of their F1 progeny, and additional markers scored, to yield a linkage map for the F1 with 136 linked RAPD markers on 13 linkage groups, covering 2030 cM. Using the linkage map and the phenotypic data for the F1 progeny, we were able to identify QTLs on 5 linkage groups bearing significant association with wood density (LOD scores higher than 2.0). These QTLs accounted for 73% of the phenotypic variance for wood density in the F1 progeny tests. Use of markers for these and other QTLs in marker-assisted selection in Douglas-fir will be discussed.

WOOD DENSITY CONSIDERATIONS IN BLACK SPRUCE (*PICEA MARIANA)* TREE IMPROVEMENT PROGRAM

S.Y. Zhang

Forintek Canada Corp.
319, rue Franquet
Sainte-Foy, PQ, Canada G1P 4R4

Keywords: black spruce, wood density, growth traits, variation, correlation.

Based on 15-year-old black spruce trees from 40 half-sib families, this study examined the genetic variation and inheritance of wood density, its relationship with growth traits (tree height and bole volume) and the variation of the relationship with genotype and environment implications of these genetic parameters and results for wood quality improvement were discussed.

Compared with growth traits, wood density shows a smaller phenotypic variation, but is under stronger genetic control. In general, wood density in this species shows a moderate negative genetic correlation with growth traits. Index selection for multiple traits is therefore essential in order to achieve optimal genetic gains. The results indicate that by selecting for dry mass weight a remarkably higher genetic gain in gross fibre yield would be obtained than by selecting for bole volume alone (+14.15% and +9.28%, respectively). Furthermore, selection for dry mass weight would result in less reduction in wood density, and while holding wood density at zero change, it is still possible to obtain huge genetic gain in gross fibre yield.

Although a moderate negative relationship between wood density and growth traits exists in this species, the relationship of wood density with growth traits, to some extent, varies with genotype and environment. A non-significant or even a weak positive relationship can still be found in some families. The negative relationship in this species tends to be weaker in the families growing in a more favourable environment. This study indicates that in this species it is still possible to select some families which not only grow fast but maintain high wood quality.

TOWARDS GENETIC IMPROVEMENT OF END-PRODUCT VALUE IN RADIATA PINE

C.J.A. Shelbourne, R.P. Kibblewhite,
B. Manley and J.M. Uprichard

New Zealand Forest Research Institute Ltd.
Rotorua, New Zealand

ABSTRACT

Log and wood properties dictate the suitability and acceptability of radiata pine for different manufactured end-products. An extensive, multi-disciplinary programme of research at NZFRI has the aim of determining and documenting the technical requirements for different end-products in terms of critical wood characteristics. It will do this by developing predictive relationships linking log, wood and chemical properties with the characteristics and value of different end-products. By studying individual trees of different clones or families the magnitude of the effects of genotype, environment and silvicultural management can also be quantified.

The first study, largely completed, involving two trees from each of eleven 16 year-old clones, quantified variation between clones and between trees-within-clones in wood and chemical properties, and related these to whole-tree pulp and paper properties from mechanical (TMP) and kraft processes.

A second study of 25 thirteen year-old seedling trees, preselected on parental and phenotypic information for extremes of fibre dimensions and wood density, will evaluate relationships between a number of wood and chemical properties with end-product characteristics of sawn wood, and TMP and kraft pulp and handsheet properties.

Understanding linkages between wood properties, processing conditions, and end-product values will enable an appraisal of processing options to maximise economic returns. This knowledge will also assist forest managers to target silviculture and tree breeders to make appropriate economic weighting of selection criteria in selection indices for choosing parents for the breeding population and seed orchards and clones for clonal forestry.

ASPEN IMPROVEMENT STRATEGIES FOR WESTERN CANADA - ALBERTA AND SASKATCHEWAN

Bailian Li

Department of Forest Resources
University of Minnesota
North Central Experiment Station
Grand Rapids, MN, USA

A breeding plan is developed to improve trembling aspen (*Populus tremuloides*) and hybrids for fiber and wood production in Alberta and Saskatchewan for members of the Aspen & Larch Genetics Cooperative at the University of Minnesota. Phenotypic selections are being made from three major geographic regions representing the range of the cooperator's lands, e.g., northern Alberta, southern Alberta, and east-central Alberta and western Saskatchewan. A simple recurrent breeding program is proposed with a nested polycross mating design. A combined family and within family selection will be used to form a new breeding population for advanced generation breeding. Initial focus of the aspen hybrid breeding program is to identify the types of hybrids suitable for planting in northern latitudes. Hybridization will concentrate on crosses between local trembling aspen selections and northern sources of *P. tremula* and *P. davidiana*.

ABSTRACTS OF
WORKSHOP CONTRIBUTIONS

Tree Seed
Working Group

HOW SEED ORCHARD CULTURE EFFECTS SEED QUALITY: EXPERIENCE WITH THE SOUTHERN PINES

James P. Barnett

USDA Forest Service
Southern Research Station
Pineville, LA 71360, USA

Tree improvement programs have significantly influenced the quality of southern pine seeds produced when compared to collections from native stands. Studies show that seed orchard management practices such as fertilization increase seed size and reduce seed dormancy. This results in the need for less complex pregermination treatments. Repeated cone collections from the same clones facilitate collections according to ripening (cone specific gravity), which can improve seed germination and storage. However, cultural practices may result in seed properties that are more sensitive to damage during processing procedures and result in lower quality unless special care is provided during this stage of handling. The effect of orchard management practices on seed quality also varies by species, with loblolly pine (*Pinus taeda* L.) being less affected than longleaf pine (*P. palustris* Mill.).

LOSS OF MBC (METHYL BROMIDE CHLOROPICRAN): MINIMIZING THE RISK OF DISEASE IN DOUGLAS-FIR BARE-ROOT NURSERIES BY MANAGEMENT OF SEED VIGOR, SEED-BORNE PATHOGENS AND FAMILY

Chuck Masters

Weyerhaeuser
The George R. Staebler Forest Resources
Centralia, Washington 98531 U.S.A.

Methyl bromide chloropicran (MBC) is currently used in Douglas-fir bare-root nurseries for disease and weed control. Its benefits are highly touted by nursery managers, but its use has been implicated in the degradation of the ozone layer. The plan is to terminate or significantly curtail its use by 2001. As a result, some sources suggest that reductions in nursery yield from cancellation of MBC may be as great as 30%. It is expected that disease mortality may increase only slightly due to the availability of alternate fumigants. However, fumigation alone should not be a long term solution. Integrated pest management approaches must become more common place; minimizing the use of pesticides. The discussion centers on an integrated system that focuses on the inter-relationship between seed vigor, seed-borne pathogens,, and the family (genetic relationships). The story begins at the seed orchard with factors affecting seed vigor. Opportunities for direct control are discussed, offering recommendations that describe an integrated system.

CLONAL VARIATION IN CONE AND SEED PRODUCTION IN BLACK SPRUCE AND WHITE SPRUCE SEED ORCHARDS AND MANAGEMENT IMPLICATIONS

Gregory W. Adams and Hartmut A. Kunze

J.D. Irving, Limited
Sussex Tree Nursery, R.R. #4
Sussex, New Brunswick
E0E 1P0 CANADA

ABSTRACT

Variation in cone and seed production across a range of clones in black spruce and white spruce seed orchards was investigated in 1994. The purpose was to begin to develop production information to be used along with orchard management practises to capture genetic gain more efficiently and improve clonal balance in orchard seedlots. Bulk cone collections were made from ten randomly selected ramets for each of 48 clones of white spruce and 60 clones of black spruce. The average number of cones per tree was determined for each clone. Seeds were extracted and average seeds per tree, seeds per cone and seeds per gram were determined for each clone. Clones were ranked by cone and seed production and considerable variation was found. For white spruce, a strong correlation was found between number of cones per tree and number of seeds per tree. However, the same was not found for black spruce, largely because some of the heaviest cone bearing clones produced lower amounts of sound seed. For both species, clones which produced more cones per tree tended to produce less seeds per cone and the seed was smaller than clones with more moderate levels of cone production. Clonal balance in cone and seed production is discussed with respect to practical orchard management options.

SPATIAL AND TEMPORAL DISTRIBUTION OF SEED CONES IN ORCHARD TREES: A KEY TOWARDS DEVELOPING PROPER CONE–CROP FORECASTING AND ASSESSMENT TOOLS

Guy–Étienne Caron

École de sciences forestières
Université de Moncton, Edmundston Campus
165 Boulevard Hébert
Edmundston, New Brunswick
Canada E3V 2S8

Seed–cone crop forecasting and assessment are important tools used by seed orchard managers to plan various orchard operations to maximize seed yield. Cone–crop forecasting is normally conducted in the fall preceding the crop year. Cone–crop assessment is a quantitative evaluation conducted in the spring of the crop year. Once used for mature stands, cone–crop forecasting and assessment methods are now being modified/developed for orchard situations. To develop proper forecasting and assessment tools for orchard situations, one must gain better knowledge of tree' bearing capacity, of annual fluctuations in production, and of the integrated relationships between crop size, tree age, and sexual zonation in the tree' crown. Factors affecting sexual determination are briefly introduced. Then, quantitative information of young black spruce (*Picea mariana* (Mill.) B.S.P.) annual seed–cone production and potential yearly fluctuations are presented, and the effect of such production on seed–cone zonation in the tree' crown examined. Finally, two new forecasting and assessment tools developed for black spruce orchards are presented.

THE BIOLOGY AND MANAGEMENT OF FOREST SEEDS: GENETIC PERSPECTIVES

D.G.W. Edwards[1] and Y.A. El-Kassaby[2]

[1] Canadian Forest Service
Pacific Forestry Centre
Victoria, B. C.

[2]Pacific Forest Products Ltd.
Saanich Forestry Centre
Saanichton, B.C.

The biological impact of forest tree reproductive processes and the extent of management practices of seed orchards and nurseries on the genetic representation of new forests, are discussed in a holistic fashion. It is emphasized that all the steps in tree reproduction, their timing and duration, and how seeds are used to produce new plants for artificial regeneration, especially in containerized nurseries, are intimately related, cumulative, and bear directly on the genetic composition of the seedling crop. These processes and their interactions need to be more fully understood to maximize genetic gain, and for the sustainability of genetic diversity.

SYNTHESIS, CONCLUSION AND OUTSTANDING PROBLEMS IN ADVANCED GENERATION BREEDING IN FOREST TREES

CURRENT ISSUES IN TREE BREEDING

M.J. Carson

New Zealand Forest Research Institute
Rotorua, New Zealand

From contributions to meetings of this kind one can get a feel for the areas of research that people are working and interested in. From my breakdown of the various paper and poster contributions, it seems that quantitative genetics got about a quarter of the emphasis, forest genetics formed a significant component, while breeding strategies (the theme of the meeting) also represented a major contribution.

(See Diagram 1. - Current issues in tree breeding.)

Surely a good sign for tree breeders are the important contributions seeking applications of molecular genetics, while the session on seed production systems has reminded us of the critical dependence of breeding on these and vegetative propagation methods for optimal deployment of our improved stock. We have been well-served at this meeting with many thoughtful presentations of a consistent high standard. In addition, however, the conference organisers have encouraged us to stand back and examine the direction in which our tree breeding strategies are evolving, and have asked us to redefine those strategies in the light of parallel efforts in different crops, and with the availability of much more sophisticated research tools. It is probably fair to state that we are at, or near, a crossroad which will see us reviewing our uses of quantitative, population genetic theory against a flood of information provided through molecular genetic techniques. New directions and breeding strategies will surely result! I would like now to make some general comments relating to trends that might be identified from this meeting.

First, there appears to be an increasing trend towards integrating tree breeding research with research in related disciplines. Perhaps this trend results from a recognition that science is becoming increasingly specialised, that generalists can no longer cope with assimilating the information and options available, and that specialists must learn to collaborate to obtain full advantage of the available benefits. Recent examples of these cross-discipline collaborations include forest mensurationist input to modelling of genetic gains, and forest pathologist inputs to breeding for disease resistance. At this meeting, we have had illustrations of the benefits of working with molecular biologists, as well as some good examples (of which Tony Shelbourne's presentation was a highlight) of collaboration with timber and pulp and paper engineers.

If tree breeding is to continue to make genetic gains, integration of research across disciplines is a necessity. Molecular genetics applications are about to create a new revolution in plant and animal breeding! Genetic fingerprinting techniques are already in use in forest trees, and (as illustrated so well by Toby Bradshaw) QTLs are available for practising marker-assisted selection in hybrid poplar, and can confidently be expected to be identified for important traits in other tree species. Sue Carson and others have referred to opportunities to apply DNA markers to studies of genetic diversity, and to address and test some of the fundamental assumptions of our quantitative genetic models (delightfully referred to by Stu Barker as ' Fisher's infinitesimal model' !).

C-DNA sequencing studies are being carried out by both Ron Sederoffs and Dave Neale's research groups, and Claire Kinlaw has reported to us strong evidence for synteny among the conifers (and hence, the advantages of pooling and sharing molecular data). Claire William's work on recombination rate ('the other half of synteny') promises opportunities to control genetic variation. Gene transformation has received less attention at this meeting, but is clearly also an important part of our future. Arguably, the challenges represented by transformed trees will focus initially on issues of addressing public concerns, and of screening for stable expressions across environments, and against varying genetic backgrounds.

The session on quantitative genetics, ably introduced by Stu Barker, provided us with an excellent perspective on our past and present uses of the quantitative theory, while also illustrating that a deeper understanding of its implications can provide opportunities for further genetic gains. Stu reminded us that our approach has been more statistical than genetic, in that we know nothing about the genes exposed to selection. He outlined challenges to the validity of our assumptions about gene action and number, and foreshadowed an opening of the 'black box' surrounding the relationship between genotype and phenotype.. Gene Namkoong questioned the paradigm that assumes that naturally evolved genetic variation, when exposed to selection, is implicitly subject to erosion. He offered an alternative, dynamic view that would allow breeders to manage for simultaneous increases in both genetic gain and genetic variance. Rowland Burdon canvassed a broad spectrum of issues of managing the genetic resource for both gain and diversity, providing numerous thoughtful insights (for example, for applications of biotechnology to breeding ' there are no substitutes for field testing, which is the final court of appeal ').

Arising from this session on quantitative genetics, various speakers contributed observations that have potential application to breeding population structures. Perhaps these could be useful take-home messages for practical tree breeders. They include:

- genes initially at intermediate frequencies will be major contributors to genetic gains in the first 5-10 generations of breeding.
- genes initially at low frequency will become the main source of variation beyond 10 generations (by when those at higher frequency will have been fixed) .
- mutations will begin to make important contributions beyond 10 generations, and may be the main reason for the persistence of genetic variance under long-term directional selection.
- even in relatively small populations (i.e. n=20-40), weak levels of inbreeding can be effectively offset by selection.
- divergent selection (as for multiple populations) increases total genetic variance.
- small populations may be exposed to losses of fitness for unselected traits, due to loss of heterozygosity (at the rate of 1/(2Ne) per generation) as a result of genetic drift.

These issues are all critical in understanding the impact of long-term recurrent selection strategies. I cannot resist the comment, however, that tree breeding practitioners may well ask whether selection responses greater than 10 generations from the present are truly relevant? There may be immense changes in gene technologies in that space of time?

The concepts, or paradigms, that we use in tree breeding are of great utility in designing breeding strategies. However, we must subject them to constant review, since they can also trap our thinking, leading us to avoid innovation, and (often) embrace conservatism. Stu Barker has outlined some of the pitfalls in believing too much in statistical models, warning us against overuse of Fisher's model, and reminding us that gene action at the level of the gene has little to do with gene action at the population level (at least, with respect to epistasis and dominance). We should keep this closely in mind, particularly in our discussions with molecular biologists. Significant criticisms of the model are coming from researchers in applications of molecular biology. Toby Bradshaw has commented that the infinitesimal model has never been particularly useful to hybrid breeders, and Sue Carson has challenged the validity of our assumption of polygenic, versus oligogenic gene action. Of course, we are not yet ready to throw the model out the window! But it will be difficult to make breakthroughs in breeding gains if we remain wedded to outdated concepts. We need to re-examine this model using newly-available information and tools, and to be prepared to change it to reflect a new understanding of genes and gene action.

(See Diagram 2. Current Paradigm - The Breeding Triangle)

Another concept, discussed at the meeting by Rowland Burdon, is that of the 'Hierarchy of Populations'. This concept usefully illustrates the trade-off between genetic gain and diversity under selection, for specified breeding goals, and in a single breeding generation. But if we wish to consider, say, deployment strategies for managing genetic diversity, or perhaps, selection for diverse breeding goals using multiple populations, or even the contrasting genetic diversity of closed breeding populations versus those involving introgression, then we may want to develop some different conceptual models.

(See Diagram 3. Closed Breeding Population)

Well, I've had a crack at drawing some of these! This is what I thought could happen for a closed breeding population over several breeding generations. Would Rowland's triangle become larger, while showing a trend to decreasing diversity in the breeding population? That seems reasonable. Should the triangle have a flat top, as a response to potential concerns about monocultures?

I changed the shape of the triangle (and took Rowland's name in vain) to introduce a role for introgression.

(See Diagram 4. Introgressive Breeding Population)

This might be what would happen from, say, parallel breeding populations representing different gene pools. How would we envisage Gene Namkoong's multiple population approach?

(See Diagram 5. Selection for Diverse Breeding Goals)

My drawings may have no particular value. My point is that we must keep open minds, and be prepared to utilise new paradigms. Alvin Yanchuk has speculated about the possible merits of combining the gene resource population with the breeding population, and partitioning the whole into several multiple populations. Is this essentially what has already happened with breeding populations of sheep and cattle, for which the unimproved, originating populations are essentially lost in the distant past? Are tree breeders becoming too attached to the concept of maintaining genetic diversity through retaining access to their unimproved native resources, instead of placing greater emphasis on making tangible gains through efficient use of selection?

The importance of multiple populations has been reinforced by a number of speakers at this meeting. A large number of questions can be raised in applying the multiple population concept.

(See Diagram 6. Structuring Advanced - Generation Breeding Populations)

In this example, multiple populations can be equated with "elites" (as we currently do in New Zealand). These would be directed either at different sets of traits, or different extremes of the same trait (as in this example), which distinguished them from sublines, which would be different populations directed at a similar breeding goal. In my example, one can imagine that a 'main' population (which would be selected for a mix of breeding goals, including wood density) would track around an average, or moderately above-average wood density, while one or more elites would be directed at more intensive selection for high or low values of this trait. A number of questions arise, including:

- should the elites be discrete from the main (i.e. non-overlapping) at their formation?
- how large should they be, and should there be more than one directed at each breeding goal? (speakers at this meeting have suggested that 20 genotypes per elite may be sufficient).
- should the elites be closed, or should there be gene flow from the elites to the main, and the converse?
- is sublining necessary when using elites? (two may be sufficient if controlled-pollination is used to produce the production population).
- how quickly can relatedness, and inbreeding, be approached? (the NC State breeding plan would lead to earlier inbreeding than would most others).

Efficient handling of our multiple populations provides tree breeders with interesting challenges, which will need to be addressed with our relatively new tools of computer simulation, and molecular markers.

Often it is the tools we already have available to us, or potential availability of new tools, that shapes our thinking in designing tree breeding strategies.

(See Diagram 7. Mature Tree Breeding Technologies - 'The Butt Log')

From an early emphasis on plus-tree selection, and propagation of clones for archive and seed orchard establishment, most programmes have advanced through a progeny testing phase to a stage in which design of the advanced-generation breeding population has become of critical importance. Most of the techniques involved have received some emphasis (ranging from adequate to a total 'overkill' in some cases), and may not warrant a great deal of further research (although there may be ample scope for further applications). We do, however, need to come up with new research and new techniques to provide increased genetic gains from what we expect to get simply by repeating the first cycle.

(See Diagram 8. New Tree Breeding Technologies - 'The Second Log')

There are numerous candidate technologies. For example, Toby Bradshaw and Reinhard Stettler have conducted quantitative trait dissection on poplar. There is plenty of scope for such studies to open the 'black box' referred to by Stu Barker. I have also listed other topics of mainline research interest (for example, marker-aided selection), as well as some that are extremely speculative (for example, gene therapy of forest trees!). Our area of work promises to be both exciting and rewarding over the next few decades. As exemplified by Gary Larson's cartoon series on the subject, let us ensure that this conference has stimulated 'another great moment in the history of evolution (...of tree breeding strategies)'!

Diagram 1.

CURRENT ISSUES IN TREE BREEDING
CTIA/WFGA 1995 CONFERENCE

Themes	Sub-themes	Papers/poster presentations
Quantitative Genetics	Inbreeding depression Genetic gain vs diversity Disease management	20
Molecular genetics	Genetics of species QTL/MAS studies Population variance/diversity Disease genetics	23
Breeding strategies	Population structure Early selection Gain simulation	18
Forest genetics	Propagation Wood quality Reproductive development	13
Seed production systems	Seed yield and quality Fingerprinting	7
		81

MANAGING GENETIC DIVERSITY
CURRENT PARADIGM
"THE BREEDING TRIANGLE"

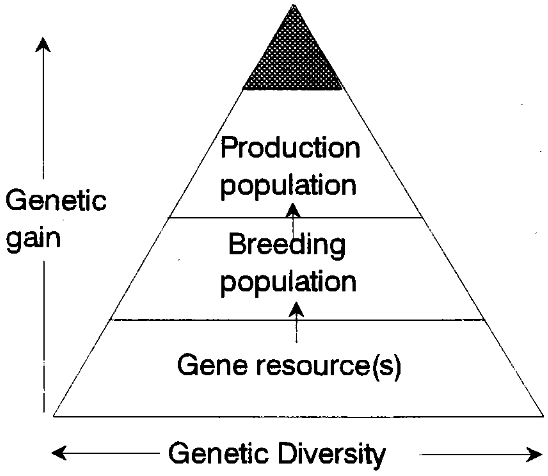


Diagram 2.

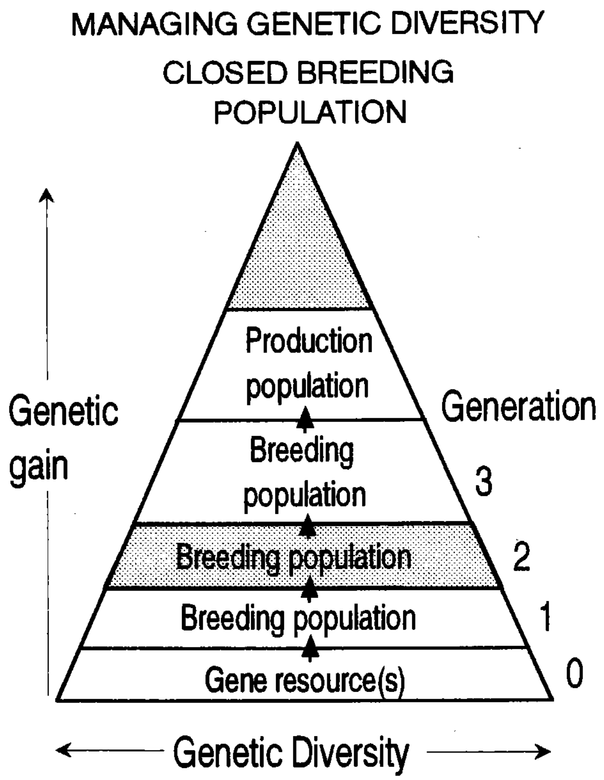


Diagram 3.

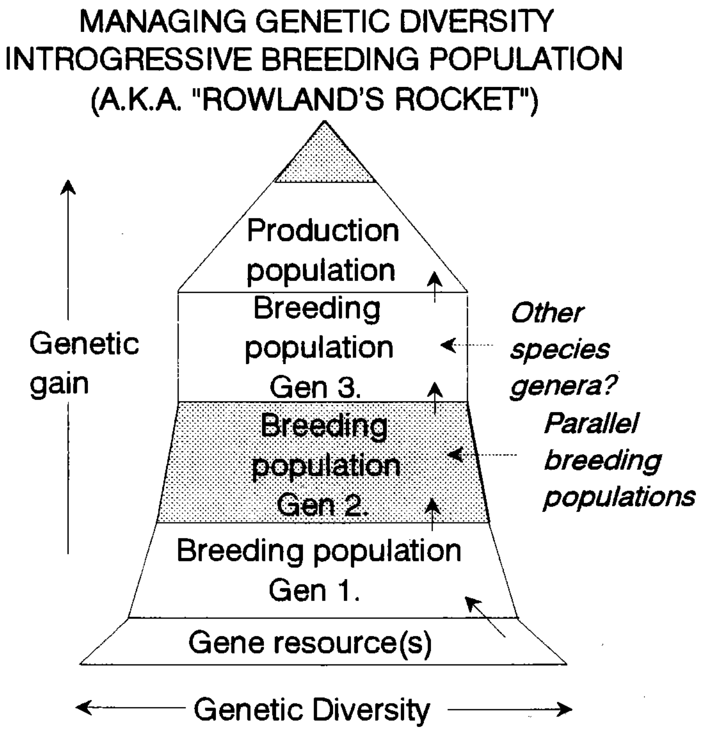


Diagram 4.

Diagram 5.

MANAGING GENETIC DIVERSITY SELECTION FOR DIVERSE BREEDING GOALS (A.K.A. "GENES CROWN"?)

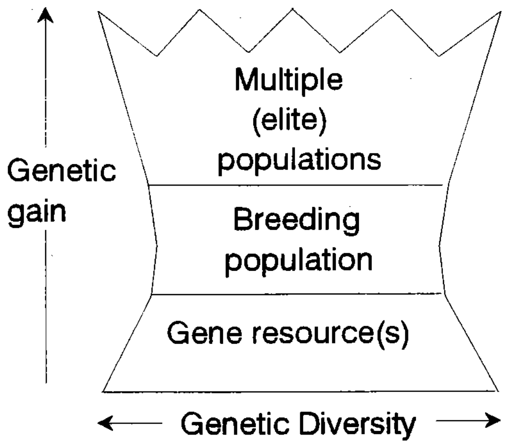
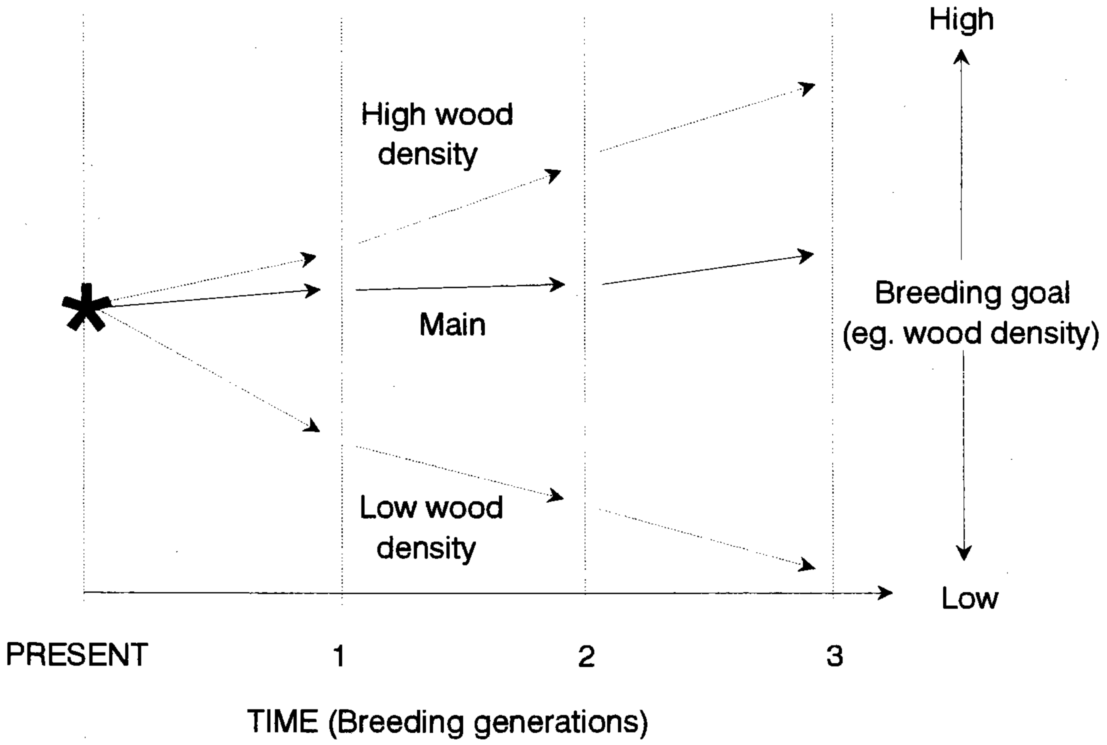


Diagram 6.

STRUCTURING ADVANCED-GENERATION BREEDING POPULATIONS

MULTIPLE POPULATIONS



Issues (a sample!)

- Should Elites be discrete sub-sets at initiation?
- How large should Elites be?
- Should genes flow from Main➤Elite, Elite➤Main?
- How many sublines for each Elite?
- Should relatedness (inbreeding) be approached rapidly?

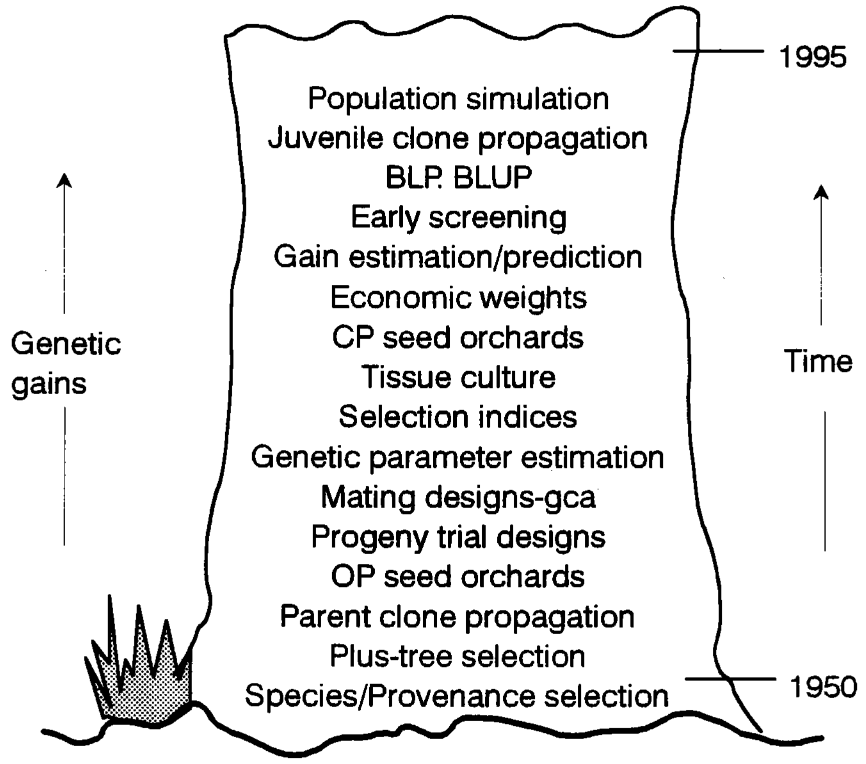


Diagram 7.

Mature Tree Breeding Technologies
'The Butt Log'

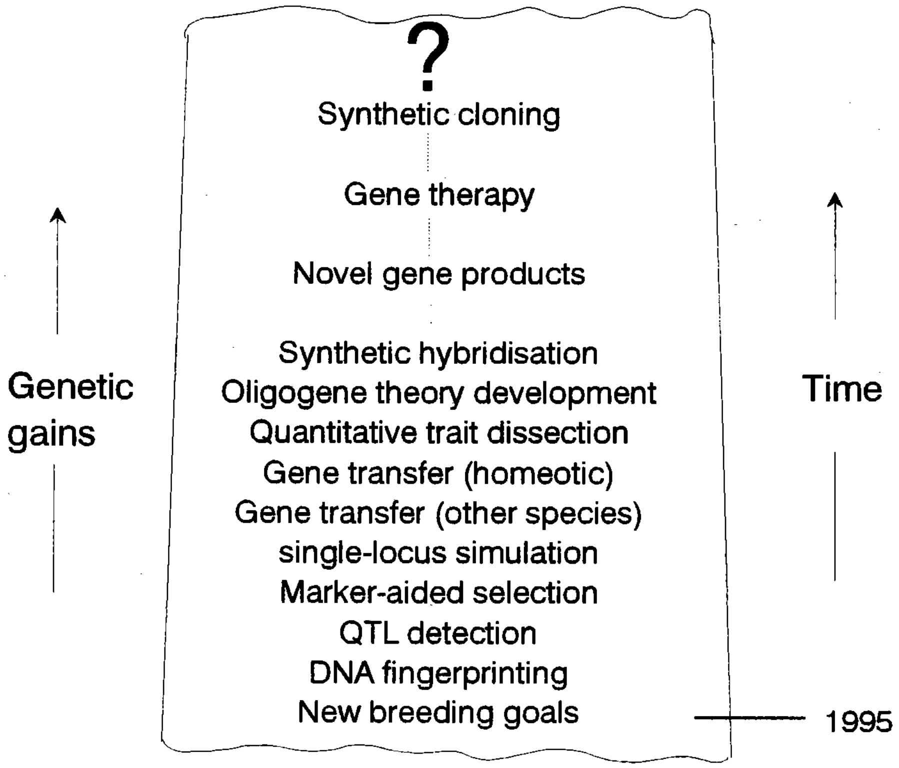


Diagram 8.

New Tree Breeding Technologies
'The Second Log'

ABSTRACTS OF

POSTER EXHIBITS

THE APPLICATION OF MOLECULAR METHODS TO THE GENETIC IMPROVEMENT OF COMMERCIAL EUCALYPTUS

S.J. Black, D. Carter & M.J. Maunders

Advanced Technologies (Cambridge) Limited
Cambridge Science Park
Cambridge CB4 4WA
United Kingdom

Molecular marker and transformation technologies are currently being employed in the improvement of commercial Eucalyptus species. By use of DNA marker analysis on an established breeding programme of *E. grandis*, individual trees have been selected as suitable parental genotypes and a third-generation population of 190 trees has been obtained for the purpose of genomic mapping. A preliminary map has been produced using RAPD and RFLP markers, and a small number of loci relating to propagation ability have been identified. This map will be expanded and transferred to related populations currently undergoing commercial evaluation. Transformation technology has also been developed to enable the direct genetic improvement of Eucalyptus. The technology is most efficient when applied to seedling-derived material, though certain mature genotypes are also amenable to some extent. The first of several field trials of transgenic Eucalyptus has now been planted and will be evaluated over the next few years.

QUANTITATIVE GENETICS ON A SPREADSHEET

J. L. Brewbaker

University of Hawaii
Honolulu, HI, USA

The evolution of personal computers and "user-friendly" software has revolutionized statistics and quantitative biology. The third in my series of "books on a disk", Quantitative Genetics on a Spreadsheet is a manual consisting of 54 problems (159 pp.) in 13 chapters, with corresponding disk (1140 kb, 3.5 inch, DOS) written in spreadsheet program QuattroPro 5.0. The hard copy tells most of the story but the disk has most of the fun. This is an interactive book that is meant to be used as well as read, as this demonstration will seek to portray. Most problems can be translated with ease into other programs, although the graphics often don't translate with ease. Spreadsheets provide our calculations a quantum leap from calculators and a solid basis for use of programs like SAS, at the same time teaching what is actually going on.

Quantitative genetics is easily (and unfairly) conveyed as a formidable science, one that weds the most mysterious models of statistics with the most mysterious and elusive genes in living organisms. Spreadsheets allow us to interact with these models in "sensitivity analyses" that can display the fragility of our models, if nothing else. Examples will be given treating QTLs, covariance methods, factorials, diallels and generation mean analysis.

ASEXUAL PROPAGATION OF JACK PINE: REGENERATION OF PLANTS FROM *IN VITRO* CULTURED COTYLEDONS

R.D. Browne and C.G. Davidson

Agriculture and Agri-Food Canada
Morden Research Centre
Morden, MB, Canada

This study reports improvements in development of *in vitro* propagation methodology for jack pine which has long been considered difficult to propagate (Harry and Thorpe 1994). Adventious buds and shoots were successfully induced from cotyledon explants when cultured on Gresshoff and Doy (Mehra-Palta et. al. 1978) medium supplemented with 1% sucrose, 0.7% agar and 22.1 M benzyladenine. Mean shoot production rate was 12.3 per cotyledon over a one year period. Based on the mean number of cotyledons per seed (4.3), an increase of approximately 50 shoots per seed could be obtained. Individual shoots were successfully excised from the cotyledons and maintained in culture for 6 months. Overall survival at the end of this period was 76.4%. Spontaneous rooting occurred in 15.5% of the shoots. Rooted shoots were successfully transferred to peat based medium in a greenhouse. Preliminary studies indicated that rooting of shoots was enhanced with an 8 day pulse treatment of 54 M naphthalene acetic acid. Further studies are targeted at improving rooting frequency and the level of shoot multiplication.

ASEXUAL PROPAGATION OF JACK PINE: PROCEDURES FOR ROOTING CUTTINGS FROM SEEDLING DONOR PLANTS

C.G. Davidson and R.D. Browne

Agriculture and Agri-Food Canada
Morden Research Centre
Morden, MB, Canada

An operational method of rooting jack pine cuttings from seedlings has been developed. This procedure uses proliferated dwarf shoots produced from pruning treatments. Rooting frequencies of 80-100% have been obtained routinely. Environmental factors influencing donor plants and cuttings have been investigated. Donor plants are best grown under standard greenhouse conditions (25 - 30 C day/15 - 20 C night; 16 hour photoperiod) with limited fertilization. A series of at least 3 pruning treatments could be applied over one growing season. Multiplication rates without serial propagation were between 15 and 20 cuttings per seedling. Cuttings (5-10 cm length with basal needles removed) were dipped in 5.4 M NAA, inserted in a peat based medium and placed in a misting regime to maintain high humidity (10 sec bursts every 10 min). Visible rooting occurred by 6-8 weeks with a mean of 5 roots per cutting. The majority of roots were greater than 8 cm in length at 8 weeks. Rooted cuttings have been established in both greenhouse and field plantings. Performance evaluations are underway in Saskatchewan (Weyerhaeuser Canada Ltd.) and Manitoba (CFS).

KARYOTYPING SPRUCE AND FIR BY *IN SITU* HYBRIDIZATION

J.E. Carlson, G.R. Brown, and V. Amarasinghe

Biotechnology Laboratory and Department of Forest Sciences
University of British Columbia
Vancouver, B.C. Canada V6T 1Z3

We have developed a fluorescence *in situ* hybridization (FISH) technique for determining the distribution of repetitive DNA elements in chromosomes of white spruce (*Picea glauca)* (2n = 24), Sitka spruce (*Picea sitchensis)* and Douglas-fir. Results will be presented on the chromosomal location of several repetitive DNAs, including the 5S ribosomal RNA genes, the nucleolar organizer regions (18S/5.8S/26S rRNA genes), rDNA intergenic spacer sequences, centromere sequences, telomere sequences, and total genomic DNA probes. These repetitive DNAs reveal a structural organization of chromosomes unique to conifers. The FISH technique has a powerful application in forest genetics for karyotyping conifers and for determining the chromosomal constitution of suspected hybrids, either from introgression zones or from controlled crosses. Using the FISH technique it is possible to accurately identify all of the chromosomes of a species and to distinguish homologous chromosomes between species.

USE OF MICROSATELLITE DNA MARKERS TO ASSESS GENETIC DIVERSITY WITHIN AND AMONG POPULATIONS OF PLANTED AND NATURALLY OCCURRING WHITE SPRUCE (*PICEA GLAUCA* [MOENCH] VOSS)

H. Cobban[1], A. Good[2], C. Strobeck[2], B. Dancik[1]

[1] Department of Renewable Resources;
[2] Department of Biological Sciences
University of Alberta
Edmonton, AB., Canada

Genetic diversity within and among white spruce populations can be examined directly at the DNA level through comparison of highly variable, repetitive sequences known as microsatellites. In order to provide a useful assessment of genetic diversity, genetic markers should be highly variable, neutral, easily interpreted, and ubiquitous among differing populations. Microsatellite markers appear to offer these advantages. The codominant nature of microsatellite markers allows for clear, unambiguous scoring of heterozygotes as well as homozygotes among samples. Heterozygosity, along with other gene diversity indices, can be easily evaluated. A further bonus associated with microsatellite markers is that they can be generated by means of the polymerase chain reaction which facilitates fast results.

APPLICATION OF SOMATIC EMBRYOGENESIS TO THE DEVELOPMENT OF A CLONAL SELECTION PROGRAM FOR INTERIOR SPRUCE (*PICEA GLAUCA-ENGELMANNI* COMPLEX)

D. Cyr[1], S. Binnie[1], S. Fan[1], S. Grimes[1], S. Grossnickle[1], C. Hawkins[2], G. Kiss[3], I. Loyola[1], G. Quan[1], J. Russell[4] and A. Yanchuk[5]

B.C. Research Inc., Vancouver, BC, Canada[1]
BCMoF, Red Rock Res. Stn., Prince George[2]
Kalamalka Res. Stn., Vernon[3]
Cowichan L. Res. Stn., Mesachie L.[4]
Research Branch, Victoria, BC, Canada[5]

The development of SE for interior spruce has evolved to the implementation of a clonal selection program using 1,000 lines from up to 30 full-sib crosses (partial diallel). Parents from a breeding arboretum (174 trees) were selected by the BCMoF for height and weevil resistance using 4 OP progeny tests. During 1993/94, 655 embryogenic lines were induced from 18 families (9 parents) and subsequently cryopreserved. Greater than 50 % of these produced sufficient numbers of somatic embryos for plant production. During 1994 approximately 400 lines (11 families) were matured with a conversion frequency to somatic seedlings of 44 to 75 %. This facilitated transplanting of plants from 309 lines to the nursery in 1995. Currently, 190 growth-ranked lines are in field trials, with a delivery of the new material expected during 1996. Subsets of both are being assessed for phenotypic variation (physiological and morphological). For the clonal trials, 6 ramets will be planted on sites selected for high growth potential and susceptibility to weevil attack. Embryogenic lines will be produced from new families for trials during 1997 and 1998. Using a 5 % selection criterion, approximately 50 lines will be chosen for operational implementation. It is anticipated that this may occur within 5 to 6 years based on early assessments.

PHENOTYPIC VARIATION IN AN ABSCISSIC ACID-INDEPENDENT LINE OF HYBRID LARCH (*LARIX* X *EUROLEPIS*)

E. Irene Hay and P. von Aderkas

Centre for Forest Biology
University of Victoria
Victoria, B.C.

Phenotypic variation in the embryogenic line #2086 of *Larix* x *eurolepis* is found in all stages of development, from the embryonal mass plus suspensor stage to the mature, rooted embryo. In the embryonal mass plus suspensor stage, there are six phenotypes. These phenotypes can be found in the same tissue aggregate and can proliferate and give rise to one another. The development of an embryonal mass plus suspensor into a mature embryo can take one of ten different routes. The phenotype of the embryonal mass does not appear to determine the final phenotype of the mature embryo. All roots formed are normal in appearance regardless of the phenotype of the shoot. Abnormal phenotypes can form normal shoot apices and epicotyls. In this study, the reasons for the phenotypic plasticity of this line and the failure of the majority of embryonal masses to mature are explored.

THE DOUGLAS FIR LUMINAL BINDING PROTEIN AS A MOLECULAR MARKER OF STRESS AND DEVELOPMENT

Benjamin S. Forward, Timothy J. Tranbarger, Santosh Misra

Department of Biochemistry and Microbiology
University of Victoria
Victoria, B.C.

The luminal binding protein (BiP) is a molecular chaperone and member of the heat shock protein 70 family of proteins which is localized within the endoplasmic reticulum (ER) of plant, animal, and yeast cells. It is responsible for the proper folding and assembly of newly synthesized proteins in the lumen of the ER.

DNA sequence analysis of full length Douglas fir BiP cDNAs revealed that it is encoded by a multigene family. The inferred amino acid sequence of the Douglas fir BiP is highly similar to other plant BiPs, however, it does contain a novel ER retention signal sequence at the carboxy terminus. The level of Douglas fir BiP mRNA expression increases during germination with maximal levels occurring in 4 day old seedlings. The level of BiP protein, however, increases steadily from mature seed to 14 day old seedlings. The level of BiP protein increases in response to cold treatment in 14 day old seedlings. These observations suggest that BiP mRNA and protein levels may be used as molecular markers to gauge the progression of these events in Douglas fir. Furthermore, due to the chaperoning nature of this protein, other proteins with more direct roles in development and cold acclimation can be isolated through co-immunoprecipitation and characterized to gain a better understanding of these processes in Douglas fir.

EFFECT OF TEMPORAL INBREEDING ON FIELD PERFORMANCE AND HEIGHT HERITABILITY OF WESTERN WHITE PINE FAMILIES FROM A SINGLE STAND

M. D. Meagher

Canadian Forest Service
Pacific Forestry Centre
Victoria, B.C., Canada

Differences in temporal inbreeding among seed parents in a single stand were found by El-Kassaby et al., 1993. Progeny of 15 parents, each represented by 3 seed collections, were planted in a single site in an RCB design. The influence of inbreeding estimate per seed crop in parent will be determined for 5-year height, and family rank and heritability will be calculated for each seed year.

EVALUATION OF SUPPLEMENTAL MASS POLLINATION SUCCESS USING RAPD MARKERS

Yong-Bi Fu, John E. Carlson, Joe Webber, and Michael Stoehr

Biotechnology Lab/Department of Forest Sciences
University of British Columbia
Vancouver, B.C.

and

British Columbia Ministry of Forests
Research Branch
Victoria, B.C.

Determining the success of supplemental mass pollination (SMP) is crucial to developing efficient SMP techniques in a seed orchard management. Previous studies using phenotypic markers, rust resistance, dyed pollens, and isozyme variation, indicate that SMP success rate ranged from 4 to 69%. Because of the lack of genetic markers, most of these studies examined only one pollen donor with a unique genotype and the methods used cannot be extended to a relatively large scale. In this study, we explore the potential of using RAPD markers to evaluate the SMP success from 4 and 12 mixed pollen donors in a white spruce seed orchard. By genotyping seeds as well as maternal and specific potential paternal trees and implementing a maximum likelihood method, we aim at determining with more confidence the proportion of seeds of a maternal tree from each of 12 mixed pollen donors.

RANDOM AMPLIFIED POLYMORPHIC DNA (RAPD) VARIATION IN INTERIOR SPRUCE

P. D. Khasa and B.P. Dancik

Department of Renewable Resources
Faculty of Agriculture, Forestry, and Home Economics
University of Alberta
Edmonton, Alberta, Canada T6G 2H1.

Interior spruce [*Picea glauca* (Moench) Voss (white spruce)-*Picea engelmannii* Parry (Engelmann spruce) complex] is important economically, ecologically, and aesthetically in B.C. Random amplified polymorphic DNA (RAPD) genetic fingerprints have been developed in interior spruce. These genetic markers are now currently being used to quickly identify both species and their hybrid swarms at any stage in the sporophyte phase of the life cycle, to validate parents being used in controlled crosses in breeding programs for interior spruce, to examine the variation within and among populations and determine the occurrence and the magnitude of introgressive hybridization in an overlap zone between the two species. The extent and evolutionary significance of interspecific introgression and implications for conservation of interior spruce are discussed.

GENETIC VARIATION OF INTERNAL NUTRIENT RETRANSLOCATION IN DOUGLAS-FIR (*PSEUDOTSUGA MENZIESII* (MIRB.) FRANCO)

S.B.R. Kiiskila and R. van den Driessche

Centre for Forest Biology
University of Victoria
Victoria, B.C., Canada

Research Branch
B.C. Ministry of Forests
Victoria, B.C., Canada

Internal retranslocation within plant organs of nitrogen, phosphorus, potassium and magnesium may supply a significant proportion of a conifer's nutritional requirements. Nutrient retranslocation in the foliage is generally associated with leaf senescence in the fall, however it also occurs in the spring from one year old and older needles to newly forming needles. It is not known whether there are significant genetic differences in internal nutrient retranslocation. Twelve families of Douglas-fir (*Pseudotsuga menziesii* (Mirb.) Franco) with varying growth rates, grown at three sites of differing productivity, have been sampled to determine whether efficient retranslocation is genetically determined, or associated with increased productivity, as measured by stem volume and foliar biomass. Foliage (current, one and two years old), foliar leachate, and needle litter were collected over a one year period for mineral nutrient analysis. There are some consistent internal nutrient retranslocation patterns within families, but apparently little relationship between retranslocation and productivity. Nutrient retranslocation is also affected by site productivity, and varies by element.

HERITABILITY OF REDUCED SPOTTING AND GAIN VS. BLISTER RUST IN WHITE PINE FOR BRITISH COLUMBIA

M.D. Meagher and R.S. Hunt

Canadian Forest Service
Pacific Forestry Centre
Victoria, B.C., Canada

Data of mean rust spots per seedling following artificial inoculation with blister rust of 215 families in 4 separate years of inoculations were analysed. Family heritability ranged from 18% to 87%, averaging 62.5% among years. Families inoculated more than once showed strong rank stability. A good trend of reduced cankering 18 months after inoculation with reduced spotting was found. Phenotypic gain via collection of open-pollinated seeds from Parent Trees (PTs) should reduce mean spots per seedling by 21% vs. woods-run parents, while collecting OP seeds from the best PTs after screening should reduce mean spots by a further 19%.

GENETIC VARIATION OF TEN NATURAL POPULATIONS OF *PINUS DENSIFLORA* IN KOREA BASED ON RAPD MARKER ANALYSIS

Yong-Yul Kim[1], Jung Oh Hyun[1], Kyung-Nak Hong[1], Tae-Bong Choi[1]
and Kyu-Shick Kim[2]

[1]Department of Forest Resources
Seoul National University, Korea
[2]Forest Genetics Research Institute, Korea

From ten populations of *Pinus densiflora* in Korea, 17 to 31 trees per population were sampled. Each megagametophytic DNAs were extracted from seven seeds per tree and used for genotyping of the seed tree by RAPD PCR. From the 1-to-1 segregation tests, 30 RAPD bands were confirmed to be diallelic loci and one was a monomorphic locus. High levels of genetic variation were indicated by expected heterozygosity(0.395) and percentage of polymorphic loci(95.2%). The value of F_{IS} and F_{IT} was -0.353 and -0.177, respectively, indicating the species may suffer from an inbreeding depression. The value of F_{ST} and G_{ST} was 0.130 and 0.129 which were much higher value compared to those from isozyme studies. The Nm value (1.667) indicated a significant migration taking place between populations through a long distance of pollen dispersal. However, it was not in agreement with the implication made from the F_{ST} and G_{ST} values estimated. The mean value of Nei's genetic distances was 0.095 and the dendrogram by the UPGMA method indicated some geographic patterning.

SELECTION FROM BASE POPULATIONS OF *EUCALYPTUS*: A CASE STUDY

B.M. Potts and N.M.G. Borralho

Cooperative Research Centre for Temperate Hardwood Forestry
Department of Plant Science
University of Tasmania
GPO Box 252C, Hobart, Tasmania

An evaluation of family trials derived from a range-wide collection of open-pollinated seed of *Eucalyptus globulus* ssp. *globulus* from native stands in Australia is currently being undertaken. A racial classification based on geography, taxonomy and growth responses on five sites (Jordan *et al.*, Silvae Genetica, in press) was used to define genetic groups. Each parent was also assigned a stand classification to account for different levels of inbreeding amongst parents due to vary degrees of isolation in native stands (Borralho and Potts, New Forests in press). Genetic parameters and breeding values for both parents and progeny were estimated using individual tree mixed model procedures, incorporating the racial (random) and stand (fixed) classifications. Overall breeding values were estimated as the sum of the race effect and BV within race.

THE ACCURACY OF GENETIC PARAMETERS AND BREEDING VALUES ESTIMATED FROM OPEN-POLLINATED PROGENIES OF *EUCALYPTUS GLOBULUS*

B.M. Potts, G.R. Hodge, P.W. Volker and C.M. Hardner

Cooperative Research Centre for Temperate Hardwood Forestry
Department of Plant Science
University of Tasmania
GPO Box 252C, Hobart, Tasmania

Eucalyptus globulus ssp. *globulus* has a mixed mating system with trees exhibiting varying levels of self-incompatibility and marked inbreeding depression for growth (Hardner and Potts 1995, Silvae Genetica 44: 46-54). Base populations derived from collection of open-pollinated seed from native stands in Australia are currently being exploited for breeding purposes in many countries. Genetic parameters and parental breeding values estimated from families derived from open-pollinated seed collected from native stands were compared with those obtained using pollen of the same 26 base parents in a factorial mating design. Narrow-sense heritabilities for two-year growth were highly inflated, site x genotype interactions underestimated, and breeding values poorly correlated with controlled cross estimates (Hodge *et al.* 1995, TAG in press).

EARLY SELECTION OF BLACK SPRUCE (*PICEA MARIANA*) CONTROLLED CROSSES IN NURSERY TESTS

Michel Villeneuve

Ministère des Ressources naturelles du Québec
Direction de la recherche forestière
Sainte-Foy, Québec, Canada

A multifamilial variety of black spruce developed for the Gaspé breeding zone is tested in three disconnected groups of 44 to 76 full-sib families (on a nursery site for early testing and on three field sites for long-term testing). Seven standard provenances are used as control lots. The nursery test is used to rapidly eliminate the slow-growing families (in 5 years or less). Only the seed from the better crosses will be sent to the « bouturathèque » for vegetative propagation (rooted cuttings).

Family heritabilities for height are high (from 0.76 to 0.89 after 3 to 5 years of measurements). The observed superiority of the families over the control lots are substantial (15% to 19%) and comparable between groups. The validity of these nursery tests will be assessed by comparing the results with those obtained from the standard long-term tests.

RESULTS FROM USDA FOREST SERVICE SCREENING FOR BLISTER RUST RESISTANCE IN SUGAR PINE AND WESTERN WHITE PINE AT DORENA GENETIC RESOURCE CENTER

R.A. Sniezko and J. Danielson

USDA Forest Service
Dorena Genetic Resource Center
Cottage Grove, OR, USA

The USFS has been screening sugar pine (*Pinus lambertiana*) and western white pine (*Pinus monticola*) trees for resistance to blister rust (*Cronartium ribicola*) for nearly 40 years in the Pacific Northwest Region (Oregon and Washington). The rust resistance program at Dorena has been patterned after work done at the USFS Intermountain Research Station. Progeny of over 10,000 selections from a range of land ownerships have been evaluated for this exotic disease. Generally, over 95 percent of seedlings develop needle lesions, and seedlings are examined for up to five years after inoculation for a variety of resistance mechanisms. Species diversity issues and timber value have stimulated high interest in rust-resistant seed. Seed orchards have been established by the USFS and/or the BLM for many breeding zones; seed is now available for some zones. Validation runs indicate a dramatic increase in resistance in one generation, particularly in the percentage of trees free of cankers. Also, the first field validation plantings indicate good progress has been made in increasing resistance. To facilitate advanced-generation planning, further information is needed on the inheritance of the resistance mechanisms, the frequency of occurrence of these mechanisms within breeding zones, and strategies for seed deployment.

FARM-FIELD TESTING FOR EARLY SELECTION OF *PINUS RADIATA*

G. Stovold, P. Jefferson and M. Carson

New Zealand Forest Research Institute Ltd.
Private Bag 3020
Rotorua, New Zealand

Farm-field tests of improved clones or families offer the potential for early selection of plantation conifers. A farm-field trial consisting of 120 seedling families of *P. radiata* was established at two 'farm' sites and one forest site in 1992, to a single-tree plot family sets-in-replications design. The families represented open-pollinated seed orchard progenies of plus-trees first selected in the 1980s. Open-pollinated progenies from the plus-tree ortets had previously (in 1982 and 1984) been planted in two series of forest trials (of two and four sites, respectively) and assessed for growth, form and disease resistance traits at various ages to age 11.

This paper relates results of assessments of growth and form traits at ages 1, 2 and 3 in the farm-field trials to older-age assessment results from the forest trial series. Conclusions address the utility of farm-field testing for the New Zealand *P. radiata* breeding programme, and future research plans.

TOWARD IMPROVING THE QUALITY OF SOMATIC EMBRYOS OF WHITE SPRUCE (*PICEA GLAUCA*)

Martin S. Lapp and David I. Dunstan

Plant Biotechnology Institute
National Research Council
Saskatoon, Saskatchewan, Canada, S7N OW9
Phone (306) 975-5250; Fax (306) 975-4839

Somatic embryogenesis is a biotechnological technique which can be used for the rapid deployment of select genotypes for reforestation. In the 10 years since the first demonstration of somatic embryogenesis in conifers, there has been sufficient progress that the technology can start to be used in applied forestry. However work is continuing in order to improve the technology in a number of areas, two of which will be discussed here. The goal is to eliminate bottlenecks to the production of superior quality somatic embryos. The effect of different carbohydrate sources on growth in shake flasks and in bioreactors is being tested for both short term (dry weight, number of stage 1 embryos) and longer term measures (maturation, adequate lipid content to ensure successful drying and germinability of the dried somatic embryo). At the present only 10% to 20% of spruce genotypes will consistently form mature somatic embryos. The use of a variety of ABA treatments to widen the potential for more genotypes to successfully mature is being investigated. Efforts are being made to find a practical indicator of genotype response to ABA treatment.

PHYSIOLOGICAL MECHANISMS RELATED TO G X E IN GROWTH RATE IN MATURE BLACK SPRUCE TREES

John E. Major[1], Kurt H. Johnsen[1] and Lawrence B. Flanagan[2]

[1]Canadian Forest Service
Petawawa National Forestry Institute
Chalk River, Ontario, Canada, K0J 1J0
[2]Dept. of Biology
Carleton University
Ottawa, Ontario, Canada, K1S 5B6

Mature trees from four full-sib families of a 7 x 7 diallel on three sites at the Petawawa National Forestry Institute (46° Lat. 77° 30' Long.) exhibited a genetic x environment interaction in growth characteristics. These families constitute a 2 female parent x 2 male parent factorial breeding structure. Progeny from female parent 59 displayed high growth rates on all three out planting sites. Progeny from female 63 displayed high productivity on two of the sites (1 and 3) but not on the third least productive site (2). To understand the physiological mechanisms related to the G x E, we first wanted to identify the E factor(s) contributing to the interaction. Second, we wanted to clearly establish that the selected subset of G entries were interacting with site. Third we wanted to identify mechanisms, interpretable with regard to the environmental response, that are under genetic control. To assess the above, measurements of stable carbon isotope, gas exchange, water potential, and water relations of leaf tissues were made from 1, 2, or 3 sites during the 1991, 1992, and 1993 growing seasons. Summary results from completed analysis and observations on repeatability will be reported.

A SECOND GENERATION BREEDING PLAN FOR JACK PINE IN MANITOBA

Jerome I. Klein and John Dojack

Natural Resources Canada
Canadian Forest Service
Winnipeg, MB

Manitoba Natural Resources
Forestry Branch
Winnipeg, MB

A plan is described for a second generation breeding program with jack pine for southeastern Manitoba. Parent trees for the second generation were selected in first generation family test plantations by Best Linear Prediction of breeding values for height, diameter, stem quality, rust resistance, and wood density, using measurements at 20-21 years from planting. Controlled matings to produce second generation full-sib progenies will be performed on grafts of 116 parent trees. Parent clones will be paired assortatively, and most or all will be used in two matings. About 116 full-sib progenies will be planted to constitute the second generation breeding population. Within-family phenotypic selection of ortets or parents for a second generation seed orchard will be carried out within the best unrelated progenies at about 10 years from planting. Polycross testing of parent trees may be initiated after completion of the matings which produced the second generation progenies, to indicate useful remedial matings. Within-family progeny testing may be performed on promising trees of the original second generation families, but selection within remedial families will probably need to be phenotypic, to avoid excessive generation length.

STOCHASTIC SIMULATION APPROACH TO MANAGEMENT OF TREE POPULATIONS FOR GENETIC GAIN AND DIVERSITY

T.J. Mullin
Genesis Forest Science Canada, Inc.
P.O. Box 1321
Truro, NS, Canada

Y.S. Park and J.D. Simpson
Canadian Forest Service
Maritimes Region
P.O. Box 4000
Fredericton, NB, Canada

Predicting genetic gains from a single round of breeding and selection may be relatively straightforward; however, resulting changes in population structure are less readily described, yet these have a profound impact on the outcome of future breeding and selection in multi-generation breeding programs. Algebraic calculations of such sequences of breeding and selection are, if not impossible, extremely complex. Simulation techniques can make such complex multi-generation tree breeding issues relatively straightforward. This presentation demonstrates the application of a decision-support software tool, called POPSIM, for the simulation and analysis of tree breeding population management strategies on modern personal computers.

Genetic effects are simulated according to an additive-dominance-epistasis model for a base population of trees that are mated according to a user defined breeding plan. Individual progeny from the breeding plan are generated, simulating the recombination of genetic effects and the random environmental effects during field testing. Selection is carried out among the test progeny to assemble various kinds of production populations for deployment of improved stock, and to advance the breeding population to the next cycle of breeding. Genetic variances are calculated directly from the simulated data and reported for each generation, together with the average level of inbreeding, effective population size, and cumulative genetic gain realized by each type of production population.

MONITORING SEED ORCHARD PERFORMANCE USING CHLOROPLAST DNA

C. Newton[1], T. Vo[1], R. Gawley[1], M. Stoehr[2], B. Sutton[1]

B.C. Research Inc.
Vancouver, BC, Canada [1]

Research Branch
Ministry of Forests
Victoria, BC, Canada[2]

The effectiveness of tree improvement programs is presently dependent on how well superior parental genotypes in seed orchards are captured in the annual seed crop. Due to wind pollination, the single greatest variable is the paternal pollen source. In the absence of controlled pollination, significant losses of genetic gain and diversity can result from pollen contamination, self-fertilization, and differential pollen fertility. Here we describe a method to assess pollen contribution using polymorphism in the chloroplast (cp) DNA of Douglas fir (*Pseudotsuga menziesii*). Paternity tests based on cpDNA have the advantages of paternal inheritance, lack of recombination and heterozygosity, and high copy number relative to nuclear DNA. Using PCR amplification of a single highly polymorphic locus and high-resolution gel electrophoresis, a large proportion (0.7) of seed orchard parents in an experimental meadow orchard can be distinguished from each other and potential contaminating pollen sources. We apply this system to estimate the efficacy of supplemental mass pollination, self-fertilization rates, differential pollen fertility, and pollen contamination. Using a multilocus approach, we describe experiments aimed at developing equivalent levels of pollen discrimination in all conifer species for which advanced breeding programs and operational seed orchards exist. We propose this system as a method to monitor seed orchard performance and assess management practices.

PARTICIPANTS

OF THE

25th CTIA/ACAA

PARTICIPANTS OF THE TWENTY-FIFTH MEETING - VICTORIA, B.C., AUGUST 1995

Greg Adams
J.D. Irving Ltd.
Sussex Tree Nursery
R.R. #4
Sussex, NB E0E 1P0

Dieter Adams
Rayonier New Zealand Ltd.
Level 5 49 Symonds St.
P.O. Box 9283 Newmarket
Auckland, New Zealand

Vellamo Ahola
University of Helsinki
Faculty of Agriculture & Forestry
Helsinki, Finland

Sally Aitken
Oregon State University
Forestry Sciences Lab 020
Corvallis, OR 97331-7501
USA

Maarten Albricht
B.C. Forest Service
3401 Reservoir Rd.
Vernon, BC V1B 2C7

John Allen
7390 Chico Way NW
Bremerton, WA 98312
USA

Val Ashley
Ministry of Forests
Kalamalka Research Station
3401 Reservoir Rd.
Vernon, BC V1B 2C7

Ian Bail
CFTT Australia
P.O. Box 406
Kew Victoria 3101
Australia

Susan Baker
Pacific Forestry Centre
506 Burnside Rd. W.
Victoria, BC V8Z 1M5

Brian Barber
Ministry of Forests
B.C. Forest Service
3rd Floor
Bastion Sq.
Victoria, BC V8W 3E7

Stuart Barker
University of New England
Department of Animal Science
Armidale NSW 2351
Australia

Jim Barnett
USDA Forest Service
2500 Shrevport Hwy.
Pineville, LA 71360
USA

Wolfgang Binder
Ministry of Forests
1320 Glyn Rd.
Victoria, BC V8W 3E7

Clark Binkley
University of British Columbia
Faculty of Forestry
270-2357 Main Mall
Vancouver, BC V6T 1Z4

Keith Bird
R.R. #1
Cowichan Bay, BC V0R 2N0

Bruce Bongarten
The University of Georgia
Warnell School of Forest Resources
Athens, GA 30602-2152
USA

Mike Bordelon
Oregon Department of Forestry
2600 Salem St.
Salem, OR 97310
USA

Susan Boyes
4312 44th Ave.
Drayton Valley, AB T7A 1G2

H.D. (Toby) Bradshaw
College of Forest Resources
University of Washington
Seattle, WA 98185
USA

James Brewbaker
Dept. of Horticulture
University of Hawaii
3190 Maile Way
Honolulu, HI 96822
USA

Floyd Bridgwater
USDA Forest Service
Texas Forest Service
Forest Sciences Laboratory
College Station, TX 77843
USA

Patti Brown
R.R. #1
Bluegrouse Site C5
Sechelt, BC V0N 3A0

Garth Brown
Biotech Laboratory
Dept. of Forest Sciences
University of BC
Vancouver, BC V6T 1Z3

Robin Browne
Agriculture & Agri-Food Canada
100-101 Route 100
Morden, MB R6M 1Y5

Rowland Burdon
Forest Research Institute
Private Bag 3020
Rotorua
New Zealand

Michael Carlson
Ministry of Forests
Research Branch
3401 Reservoir Rd.
Vernon, BC V1B 2C7

John Carlson
University of British Columbia
Biotechnology Laboratory
237-6174 University Blvd.
Vancouver, BC V6T 1Z3

Guy Caron
Université de Moncton
Edmundston Campus
165 Boulevard Herbert
Edmundston, NB E3V 2S8

Susan Carson
New Zealand Forest Research Institute
Private Bag 3020
Rotorua
New Zealand

Michael Carson
New Zealand Forest Research Institute
Private Bag 3020
Rotorua
New Zealand

Charles Cartwright
General Delivery
Mesachie Lake, BC V0R 2N0

Fiona Chandler
Ministry of Forests Research Branch
31 Bastion Square
Victoria, BC V8W 3E7

Paul Charrette
Lakehead University
955 Oliver Rd.
Thunder Bay, ON P7B 5E1

Marilyn Cherry
3176 W. 31st Ave.
Vancouver, BC V6L 2A6

Heather Cobban
307-8210 111th St.
Edmonton, AB T6G 2C7

Silvia Concheyro
New Zealand Forest Research Institute
Private Bag 3020
Rotorua
New Zealand

Thompson Conkle
USFS-PSW Research Station
2530 Nason Ave.
El Cerrito, CA 94530
USA

Louise Corriveau
Weyerhaeuser Canada
P.O. Box 1720
Prince Albert, SK S6V 5T3

Paul Cotterill
Stora-Celbi
Quinta Do Furadouro
Obidos 2510
Portugal

Keith Cox
Skimikin Seed Orchards - BCFS
Site 13
R.R. #1
Tappen, BC V0E 2X0

Dan Cress
NWTIC
848 NE 56th St.
Seattle, WA 98105
USA

Tim Crowder
Timberwest Forest Ltd.
1450 Mt. Newton Cross Rd.
Saanichton, BC V8M 1S1

Michael Crown
Forest Seed Orchard Consultants
38211 Schooner Way, R.R. #1
Pender Island, BC V0N 2M0

David Cyr
BC Research Inc.
3650 Wesbrook Mall
Vancouver, BC V6K 4E1

Bruce Dancik
University of Alberta
Dept. of Renewable Resources
Edmonton, AB T6G 2H1

Jess Daniels
Daniels and Associates Inc.
1143 W. Roanoke St.
Centralia, WA 98531
USA

Campbell Davidson
Agriculture & Agri-Food Canada
100-101 Route 100
Morden, MB R6M 1Y5

Christian De Veer
Bioforest
Edificio Prales Piso 4
Valdivia
Chile

Christine Dean
Weyerhaeuser Company
Weyerhaeuser Technology Centre
WTCIA3
Tacoma, WA 98477
USA

Laura Dewald
Northern Arizona University
Department of Forestry
Box 15018
Flagstaff, AZ 86011
USA

John Dojack
Manitoba Forestry Branch
300-530 Kenaston Blvd.
Winnipeg, MB R3N 1Z4

Denise Donelle
1102 Cascades St.
Greenfield Park, QC J4V 1C4

Dale Draper
Ministry of Forests
Research Branch
31 Bastion Sq.
Victoria, BC V8W 3E7

Nicole Dumont-Beboux
Dept. of Biology
University of Victoria
P.O. Box 1700
Victoria, BC V8W 2Y2

Glen Dunsworth
McMillan Bloedel Ltd.
65 Front St.
Nanaimo, BC V9R 5H9

Andrea Eastham
Canadian Forest Service
Site 25 C 10
R.R. #8
Prince George, BC V2N 4M6

Abul Ekramoddoullah
Paicific Forest Research
506 Burnside Road W.
Victoria, BC V8Z 1M5

Yousry El-Kassaby
Pacific Forest Products ltd.
8067 Saanich Rd.
Saanichton, BC V0S 1M0

Alan England
USDI BLM
1300 Airport LN
North Bend, OR 97420
USA

Gosta Eriksson
Dept. For. Genetics
Box 7027
Uppsala S-750-07
Sweden

Shihe Fan
Forest Biotechnology Centre
3650 Wesbrook Mall
Vancouver, BC V6S 2L2

Craig Ferguson
Ministry of Forests
P.O. Box 335
Mesachie Lake, BC V0R 2N0

Lauren Fins
Dept. of Forest Resources
University of Idaho
Moscow, ID 83843
USA

Randy Ford
Ontario Tree Improvement Board
Northeast Seed Management Assoc.
P.O. Box 329
Swastika, ON P0K 1T0

Eric Forget
Université Laval
Pav Marchand
Sainte-Foy, QC G1K 7P4

Benjamin Forward
University of Victoria
Dept. of Biochemistry and Microbiology
Box 3500
Victoria, BC V8W 3N6

David Foushee
Potlatch Corporation
805 Mill Rd.
Lewiston, ID 83501
USA

Howard Frame
Nova Scotia Dept. of Natural Resources
P.O. Box 190
Debert, NS B0M 1G0

Yong-Bi Fu
University of British Columbia
Dept. of Forest Sciences
193-2357 Main Mall
Vancouver, BC V6T 1Z4

Luis Gea
New Zealand Forest Research Institute
Sala Street
Rotorua
New Zealand

Jeff Glaubitz
Biotech. Laboratory
Dept. of Forest Sciences
University of BC
Vancouver, BC V6T 1Z3

Josefina Gonzalez
Forintek Canada Corp.
University of British Columbia
2665 E. Mall
Vancouver, BC V6P 4H3

Geoff Gooding
Oregon State University
Department of Forest Science
Forestry Sciences Lab 020
Corvallis, OR 97331-7510
USA

Matti Haapanen
Finnish Forest Research Institute
Vantaa Research Center
Box 18
Vantaa FIN-01301
Finland

Jim Hamlin
US Forest Service
Umpqua NF
P.O. Box 1008
Roseburg, OR 97470
USA

Christine Hansen
Alberta Land & Forest Services
Pine Ridge Forest Nursery
Box 750
Smoky Lake, AB T0A 3C0

Jon Hansen
Arboretum Denmark
Kirkegardsvej 3A
2970 Horsholm
Denmark

Chris Hawkins
Red Rock Research Station
R.R. #7
RMD 6
Prince George BC V2N 2J5

Irene Hay
302-1320 Purcell Pl.
Victoria, BC V8S 1Y7

Lisa Hayton
BC Forest Service
31 Bastion Sq.
Victoria, BC V8W 3E7

Chris Heaman
3640 Iona Dr.
Victoria, BC V8P 4S7

Clare Hewson
Interior Seed Orchards - BCFS
3401 Reservoir Rd.
Vernon, BC V1B 2C7

Peggy Hickey
University of New Brunswick
Faculty of Forestry and
Environmental Management
P.O. Box 4400
Fredericton, NB E3B 5A3

Christopher Hoh
2211 Sayward St.
Victoria, BC V8R 3T4

Bonnie Hooge
Red Rock Research Station
R.R. #7
RMD 6
Prince George, BC V2N 2J5

Jung-Oh Hyun
Seoul National University
Dept. of Forest Resources
103 Soedundong
Suwon Kyunggi-Do 441-744
Korea

Gunnar Jansson
Skog Forsk
Glunten
S-751 83 Uppsala
Sweden

Barry Jaquish
Ministry of Forests
3401 Reservoir Rd.
Vernon, BC V1B 2C7

Keith Jayawickrama
North Carolina State Univesity
Dept. of Forestry
Raleigh, NC 27695-8002
USA

Keith Jech
Weyerhaeuser Company
505 N. Pearl St.
Centralia, WA 98531
USA

Paul Jefferson
New Zealand Forest Research Institute
Private Bag 3020
Rotorua
New Zealand

Henning Jensen
Cowichan Lake Research Station
P.O. Box 335
Mesachie Lake, BC V0R 2N0

Jan Jensen
Royal Vet. & Agricultural University
Kirkegardsvej 3A
Hoersholm DK-2970
Denmark

Kathleen Jermstad
Institute Forest Genetics
2480 Carson Rd.
Placerville, CA 95667
USA

Sally John
SADC Tree Seed Centre Network
SADC TSCN P. Bag BW 6238
Borrowdale
Harare
Zimbabwe

Randy Johnson
US Forest Service
Forestry Services Lab.
3200 Jefferson Way
Corvallis, OR 97331-4401
USA

Dennis Joyce
Ontario Ministry of Natural Resources
1235 Queen St. E.
Sault Ste. Marie, ON
P6A 5N5

Sanna Kallio
University of Helsinki
Faculty of Agriculture & Forestry
Helsinki
Finland

Katri Karkkainen
University of Oulu
Department of Genetics
Oulu 90570
Finland

Damase Khasa
University of Alberta
Dept. of Renewable Resources
751 General Service Bldg.
Edmonton, AB T6G 2H1

John King
Ministry of Forests
31 Bastion Sq.
Victoria, BC V8W 3E7

Gyula Kiss
BC Forest Service
KFC
3401 Reservoir Rd.
Vernon, BC V1B 2C7

Jerry Klein
Canadian Forest Service
200-180 Main St.
Winnipeg, MB R3C 1A6

David Kolotelo
Ministry of Forests
18793 32nd Ave.
Surrey, BC V4P 1M5

Mathew Koshy
University of British Columbia
Faculty of Forestry
193-2357 Main Mall
Vancouver, BC V6T 1Z4

Sylvia L'Hirondelle
Ministry of Forests
Glyn Road Research Station
1320 Glyn Rd.
Victoria, BC V8W 3E7

Martin Lapp
National Research Council
Plant Biotechnology Institute
110 Gymnasium Pl.
Saskatoon, SK S7N 0W9

Bob Latta
University of Colorado
Dept. of EPO Biology
Campus Box 334
Boulder, CO 80309
USA

Joy Lavereau
Petawawa National Forestry Institute
P.O. Box 2000
Chalk River, ON K0J 1J0

Carole Lee
Prince George Tree Improvement
Station - BCFS
Site 17, Comp. 105
SS 2
Prince George, BC V2N 2K6

Bailian Li
University of Minnesota
1861 Hwy. 169 E.
Grand Rapids, MN 55744
USA

William Libby
28 Valencia Rd.
Orinda, CA 94563
USA

William Lowe
Texas Forest Service
Forest Science Lab
College Station, TX 77843
USA

Hector Magsambol
501-780 Montgomery St.
Fredericton, NB E3B 2Y1

Mary Mahalovich
USDA Forest Service: North Reg.
Forestry Science Lab
1221 S. Main St.
Moscow, ID 83843
USA

John Major
Canadian Forest Service
Maritimes Region
P.O. Box 4000
Fredericton, NB E3B 5P7

Kimberly Marshall
US Forest Service
800 Buchanan St.
Albany, CA 94710
USA

Sheila Martinson
USDA Forest Service
P.O. Box 3623
Portland, OR 97208
USA

Chuck Masters
Weyerhaeuser
505 North Pearl
Centralia, WA 98531
USA

Anne Matilainen
University of Helsinki
Faculty of Agriculture & Forestry
Helsinki
Finland

Martin Maunders
Advanced Technologies Ltd.
210 Cambridge Science Park
Milton Rd.
Cambridge CB4 4WA
UK

Barbara McCutchan
Westvaco Corporation
Westvaco Research Center
Mill Rd.
Covington, VA 24426
USA

Steve McKeand
North Carolina State Unvirsity
Box 8002
Raleigh, NC 27695-8002
USA

Michael Meagher
Canadian Forest Service
506 Burnside Rd. W.
Victoria, BC V8Z 1M5

Diane Medves
MacMillan Bloedel Ltd.
65 Front St.
Nanaimo, BC V9R 5H9

E. Morgenstern
Genetic Resource Consultants
R.R. #7
Pembroke, ON K8A 6W8

John Murphy
Ministry of Forests
3401 Reservoir Rd.
Vernon, BC V1T 8H8

Gordon Murray
Canadian Forest Service
Petawawa National Forestry Institute
P.O. Box 2000
Chalk River, ON K0J 1J0

Mary Myers
PEI Forestry Division
PEI Dept. of Agriculture, Fisheries
and Forestry
P.O. Box 2000
Charlottetown, PEI C1A 7N8

Gene Namkoong
Dept. of Forest Sciences
University of BC
MacMillan Building
189-2357 Main Mall
Vancouver, BC V6T 1Z4

David Neale
Institute of Forest Genetics
P.O. Box 245
Berkeley, CA 94701
USA

Craig Newton
3650 Wesbrook Mall
Vancouver, BC V6S 2T0

Ulrik Nielsen
Forestry Sciences Laboratory
3200 SW Jefferson Way
Corvallis, OR 97331
USA

Peter Nitschke
197 Panoramic Dr.
Sault Ste. Marie, ON P6B 6E3

Sarah Nourse
P.O. Box 105
Whately, MA 01093
USA

Greg O'Neill
Oregon State University
Dept. of Forest Science
FSL 020
Corvallis, OR 97331-7501
USA

Bob Ohrn
Bureau of Land Management
1717 Fabry Rd. SE
Salem, OR 97306
USA

Doug Ormrod
University of Guelph
Department of Horticultural Science
Guelph, ON N1G 2W1

John Owens
9310 Glenelg Ave.
R.R. #2
Sidney, BC V8L 3S1

Roger Painter
BC Ministry of Forests
Glyn Road Research Station
1320 Glyn Rd.
Victoria, BC V8W 3E7

Rungnapar Pattaavibool
Dept. of Biology
University of Victoria
Victoria, BC V8N 6K8

Martin Perron
Laval University
PC-E Marchand
St-Foy, QC G1X 7P4

Gisele Phillips
Ministry of Forests
Kalamalka Forestry Centre
3401 Reservoir Rd.
Vernon, BC V1B 2C7

Brad Potts
CRC For Temperate Hardwood
Forestry - Dept. of Plant Sci.
GPO Box 252C
Hobart, Tasmania 7001
Australia

Wendy Potts
Parks & Wildlife Tasmania
Springfield Channel Highway
Kingston, Tasmania
Australia 7050

Tapani Poykko
Foundation for Forest Tree Breeding
Karkkilante 193
Layliainen FIN-12600
Finland

Pasi Puttonen
Ministry of Forests
Research Branch
31 Bastion Sq.
Victoria, BC V8W 3E7

Grant Quan
BC Research Inc.
3650 Wesbrook Mall
Vancouver, BC V6S 2L2

Om Rajora
University of Alberta
Dept. of Renewable Resources
751 General Services Building
Edmonton, AB T6G 2H1

David Reid
Ministry of Forests
Coast Seed Orchards
7380 Puckle Rd.
Saanichton, BC V8M 1W4

Sanna Rekola
University of Helsinki
Faculty of Agriculture & Forestry
Helsinki
Finland

Kermit Ritland
Dept. of Botany
University of Toronto
Toronto, ON M5S 3B2

Jim Roberds
USDA Forest Service
North Carolina State University
Forestry Dept. Camp Box 8002
Raleigh, NC 27695
USA

John Runions
University of Victoria
Forest Biology
Biology Department
P.O. Box 1700
Victoria, BC V8W 2Y2

John Russell
Cowichan Lake Research Station
Box 335
Mesachie Lake, BC V0R 2N0

Marc Rust
University of Idaho
College of Forestry
Moscow, ID 83844-1133
USA

Deogratias Rweyongeza
University of Alberta
Dept. of Renewable Resources
Edmonton, AB T6G 2H1

Henrik Saxe
Royal Vet. & Agricultural University
Kirkegaardsvej 3A
DK-2970 Hoersholm
Denmark

Ron Schmidtling
USDA Forest Service
23332 Hwy. 67
Saucier 39574
USA

Frank Schnekenburger
Ministry of Natural Resources
1235 Queen St. E.
Sault Ste. Marie, ON P6A 5N5

Doug Shaw
Dept. of Ponology
University of California at Davis
Davis, CA 95616
USA

Tony Shelbourne
New Zealand Forest Research Institute
Private Bag 3020
Rotorua
New Zealand

Dale Simpson
Canadian Forest Service
Maritimes Region
P.O. Box 4000
Fredericton, NB E3B 5P7

Vera Sit
Ministry of Forests Research Branch
4th Floor, 31 Bastion Square
Victoria, BC V8W 3E7

Pia Smets
3806 W. 35th Ave.
Vancouver, BC V6N 2N9

Tania Smith
PF Olsen & Company (1995) Ltd.
430 Ngongotaha Rd.
P.O. Box 1127
Rotorua
New Zealand

Richard Sniezko
USDA Forest Service
Dorena Genetic Research Center
34963 Shoreview Rd.
Cottage Grove, OR 97424
USA

Reinhard Stettler
University of Washington
Forest Resources
Seattle, WA 98195
USA

Michael Stoehr
Research Branch
B.C. Ministry of Forests
1320 Glyn Rd.
Victoria, BC V8W 3E7

Roy Stonecypher
QFG Consulting
116 NE Hillside Dr.
Chehalis, WA 98532
USA

Ben Sutton
BC Research Inc.
3650 Wesbrook Mall
Vancouver, BC V6S 2L2

Cheryl Talbert
West Campus WWC - 2F2
Tacoma, WA 98477
USA

Jordan Tanz
Tree Improvement Council of BC
201-1290 Broad St.
Victoria, BC V8W 2A5

Barbara Thomas
University of Alberta
751 General Services Bldg.
Edmonton, AB T6G 2E1

Kathy Tosh
New Brunswick Department of
Natural Resources
Kingsclear Forest Nursery
R.R. #6
Fredericton, NB E3B 4X7

Vasilios Tsarouhas
University of Toronto
Earth Science Centre
Faculty of Forestry
33 Willcocks
Toronto, ON M5S 1W4

Robert Van Den Driessche
Research Branch
Ministry of Forests
1320 Glyn Rd.
Victoria, BC V8W 3E7

Martti Venalainen
The Finnish Forest Research Institute
Punkaharju Research Station
Punkaharju
Finland FIN-58450

Michel Villeneuve
Ministère des Ressources Naturelles
du Québec
2700, rue Einstein
Sainte-Foy, QC G1P 3W8

Dave Wagner
University of Kentucky
Department of Forestry
204 TP Cooper Bldg.
Lexington, KY 40546-0073
USA

Chris Walsh
Kalamalka Seed Orchards - BCFS
3401 Vernon Rd.
Vernon, BC V1B 2C7

Joe Webber
BC Ministry of Forests
Glyn Rd. Research Station
1320 Glyn Rd.
Victoria, BC V8W 3E7

Robert Westfall
Institute of Forest Genetics
USDA Forest Service
PSW P.O. Box 245
Berkeley, CA 94701
USA

Nicholas Wheeler
Weyerhaeuser Company
505 N. Pearl St.
Centralia, WA 98531
USA

Eleanor White
Canadian Forest Service
Pacific Forestry Centre
506 West Burnside Road
Victoria, BC V8Z 1M5

Claire Williams
Texas A&M University
Dept. of Forest Science
College Station, TX 77843
USA

Jack Woods
BC Forest Service
P.O. Box 335
Mesachie Lake, BC V0R 2N0

Harry Wu
306-627 MacDonald Ave.
Sault Ste. Marie, ON P6B 6A6

Chang-Yi Xie
Ministry of Forests
31 Bastion Sq.
Victoria, BC V8W 3E7

Alvin Yanchuk
Ministry of Forests
Research Branch
31 Bastion Sq.
Victoria, BC V8W 3E7

C. Yeatman
Genetics and Seed Consultant
9B Glendale Ave.
Box 721
Deep River, ON K0J 1P0

Cheng Ying
Ministry of Forests
Research Branch
31 Bastion Sq.
Victoria, BC V8W 3E7

Yuan Zhong
Faculty of Forestry &
Environmental Management
University of New Brunswick
Fredericton, NB E3B 6C2

Louis Zsuffa
University of Toronto
Faculty of Forestry
33 Willcocks St.
Toronto, ON M5S 3B3

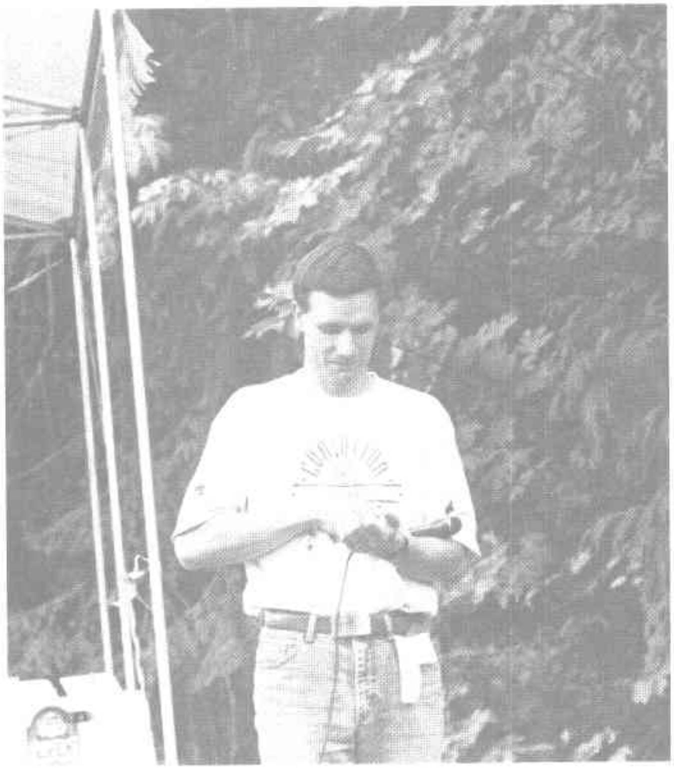
Jinghua Zuo
Oregon State University
Department of Forest Science
Forestry Sciences Lab 020
Corvallis, OR 97331-7501
USA

PHOTOS

CTIA / ACAA Sponsored Students

(left to right:)

Susan Boyes (Lakehead University); Peggy Hickey (University of New Brunswick); Denise Donelle (University of Moncton); Pia Smets (University of British Columbia); Vasilios Tsarouhas (University of Toronto); Barb Thomas (University of Alberta)
Missing: Martin Perron (Laval University)



Now.......
what was I going to say?

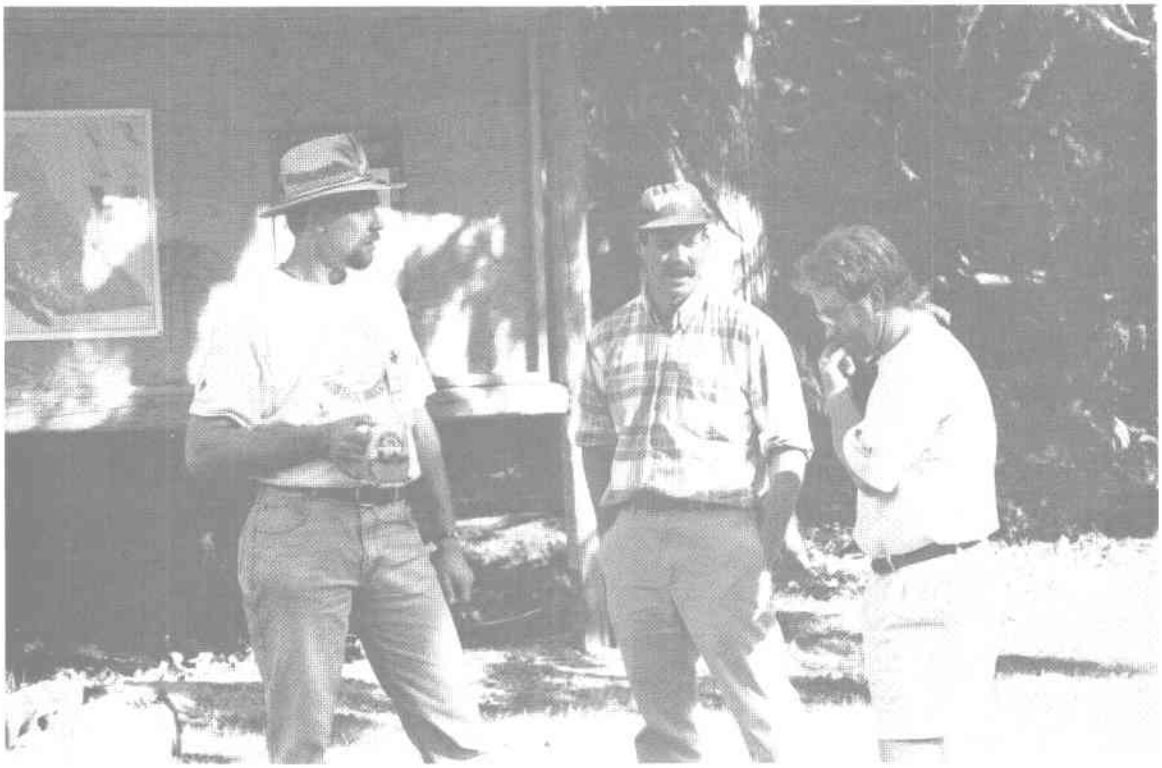
Alvin Yanchuk

♬ ♬
I did it my way!

Michael Stoehr

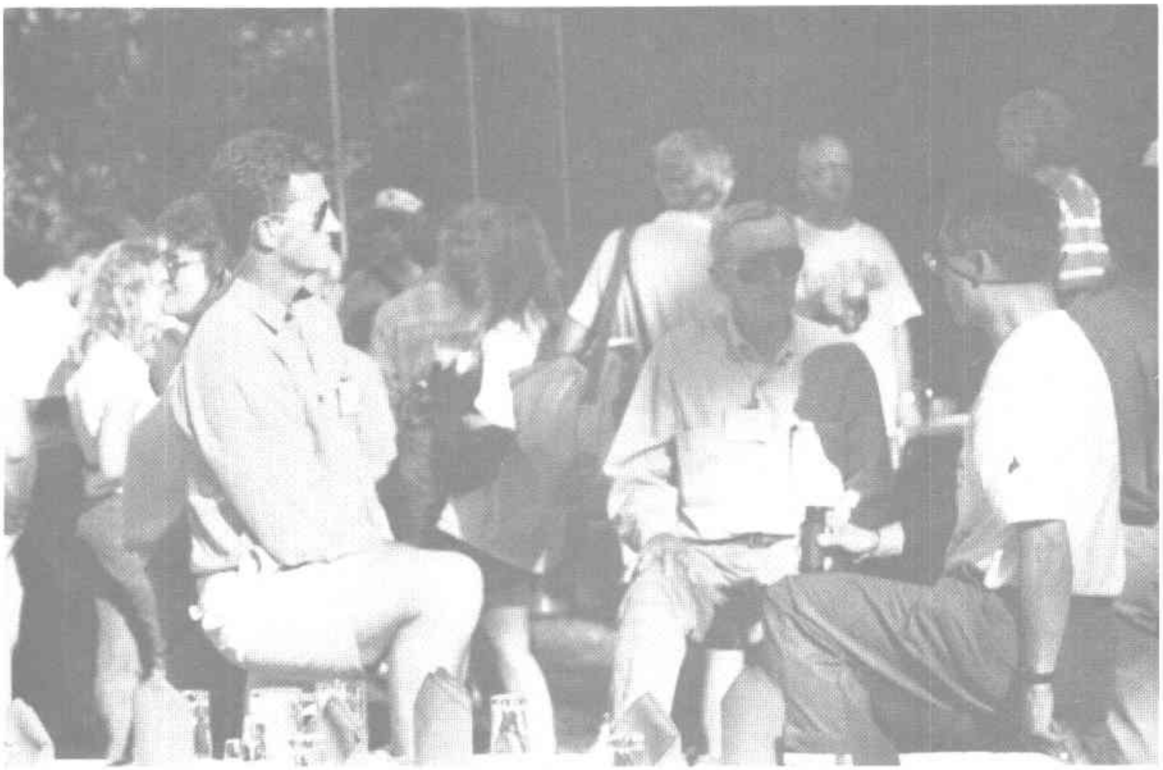
With those smiles, retirement must be good! **Chris Heaman and Kit Yeatman**

I just knew it !!!....... I'm going to win a cap! **Kris Morgenstern**

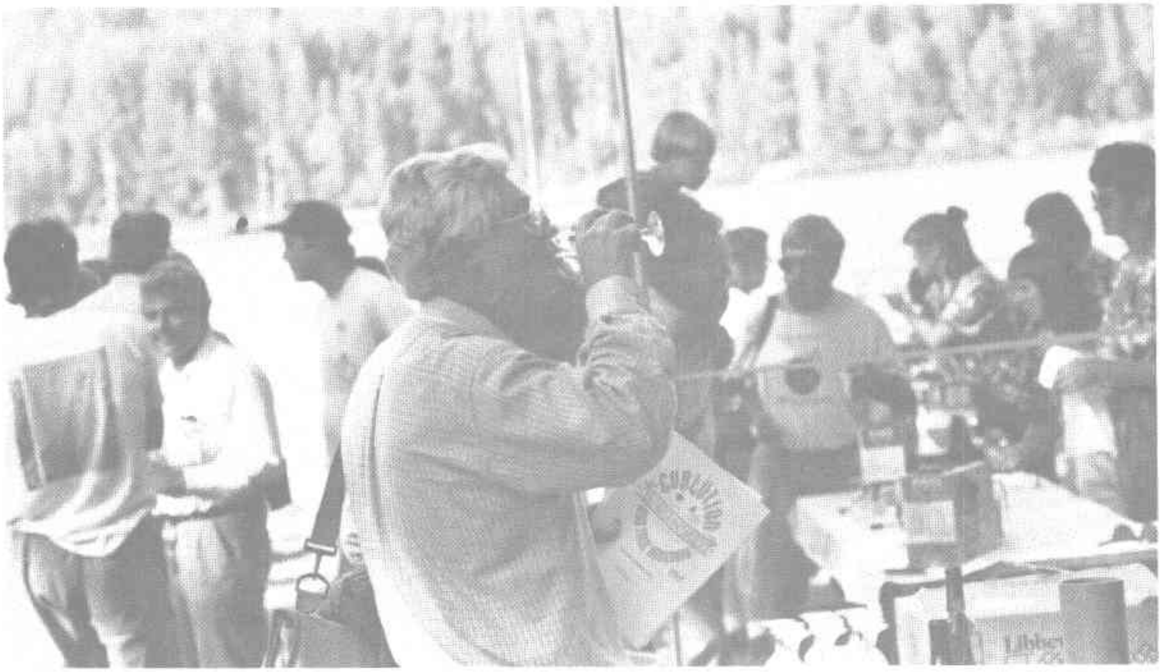


Now, where did I leave my beer mug!

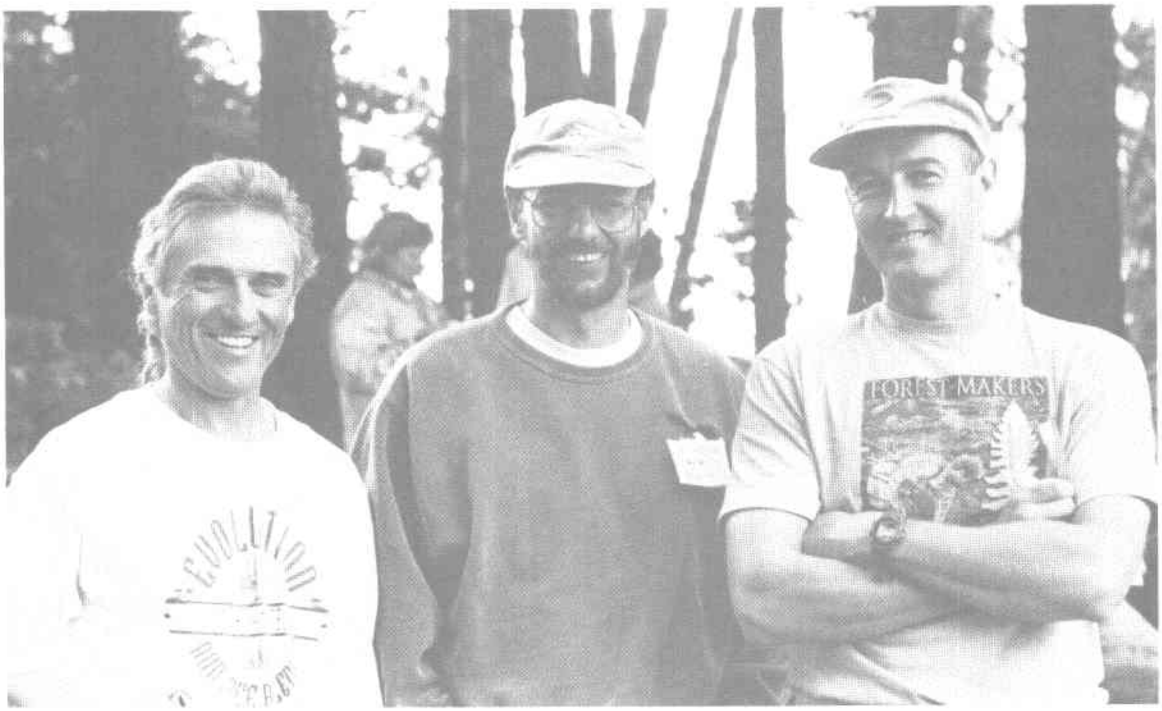
Michael Stoehr, Doug Shaw and John Russell



"Is it really a correlation between IQ and raised knees?"
Dale Draper, Stu Barker & Gene Namkoong



Conference Wine Taster   *Umh!!!!*   *Next!!!*   **Rowly Burdon**



Check out the scenery!   **John Murphy, Keith Cox and Brian Barber**

What's the matter with my legs?
Tony Shelbourne, Dieter Adams and John King

Are YOU talking to us?
Vera Sit, Lisa Hayton and Pat McGuire

With the closure of Petawawa National Forestry Institute,
(the home of the Canadian Tree Improvement Association for many years)

I'd like to thank:

Sharon Moreau,
who once again did
an excellent job of
computer manipulation
for the 25 CTIA/ACAA
proceedings .

and to all the members
of the Association, I have
enjoyed my connection
with this organization.

Joy Lavereau
CTIA/ACAA
Executive Secretary
Editor
Treasurer