

Breeding Strategy for *Pinus radiata* in Australia.

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Abstract: The Southern Tree Breeding Association Incorporated (STBA) breeding and selection program for *Pinus radiata* in Australia was developed after the amalgamation of independent member programs in 1983. The previous program documented by Boomsma (1997) and White *et al.* (1992) has been revised and changes implemented over recent years.

The STBA has modified its tree improvement strategy for *Pinus radiata* as a result of significant developments in four main areas – (i) economic breeding objectives, (ii) management of data and information, (iii) TREEPLAN® genetic evaluation and (iv) wood quality assessment. These developments have enabled the use of more flexible breeding and testing strategies which will lead to faster rates of genetic gain.

Significant developments include:

- adoption of a rolling front strategy with overlapping generations;
- increased selection pressure through larger population sizes;
- more crosses per cycle of breeding;
- greater focus on forward selection of progeny for breeding and deployment;
- a dynamic breeding population without a clearly defined nucleus and main population;
- more focus on pedigree control using full sib crosses and limited use of polycrosses;
- changes in breeding objective functions with greater emphasis on those growth, form and wood quality traits that impact significantly on profit; and
- production of genetic values targeting different production regions to account for genotype x environment interaction (GxE).

Research activities supporting the program are currently focusing on the continued development of: TREEPLAN® genetic evaluation software; the definition of economically derived breeding objective functions; an enhanced understanding of, and ability to use, GxE interaction in genetic evaluation and breeding and studies of the juvenile wood properties in the *Pinus radiata* breeding population.

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INTRODUCTION

The Southern Tree Breeding Association (STBA) manages the Australian cooperative genetic improvement program (breeding and selection) for *Pinus radiata*. This program was developed after the formal amalgamation of independent member programs in 1983.

The early history of the STBA program was documented by Boomsma (1997), who reported on the strategy revisions done through the 1980s and the early 1990s (White *et al.* 1992a, 1992b). The basic elements of the program and significant dates are listed in Table 1.

Significant events in the implementation of the STBA radiata pine breeding program, up until 1997 included: 1983 - 2 stage selection adopted; 1984/1985 - sublining of the breeding population; 1986/1987 - single pair mating with combined index selection; 1987/1988 – proposed single-cross nucleus with open-pollinated main; 1988/1989 - expanded membership (other traits) and adopted nucleus strategy; 1990 - definition of breeding objectives and allocation of material to sublines and the nucleus; 1991/1992 - multiple population subline strategy and breeding value calculation; 1992 - breeding work commenced; 1993 - New Zealand implications caused changes (two sublines to manage inbreeding, deployment crosses between lines); 1993/1994 - crossing work completed; - 1996/1997 - third generation progeny trials planted (460+ families, ~30 progeny tests).

The essential elements of the past strategy as developed and eventually adopted for the crossing effort from 1992 to 1994 were (Figure 1):

- Nucleus and Main breeding populations (open nucleus of 40, main of 300)
- Two unrelated sub-lines each comprised of three trait-based sub-populations (MP – Multi Purpose; DG – Density and Growth; GP – Growth and *Phytophthora*).
- Two-way transfer between the Nucleus and the Main.
- Crossing for deployment between unrelated sub-lines.

Over recent years, and particularly in relation to the development of data management and analytical tools, and the assessment of the 1996 and 1997 series of progeny trials, it was recognised that changes were needed to ensure rapid genetic progress and delivery of benefits to the STBA Membership. The STBA felt there was little value in continuing to breed and collect data without the tools to process and analyse the information (Kerr *et al.* 2001, 2002, McRae *et al.* 2003, 2004 this conference). As will be discussed below some of these have now been implemented.

The STBA is currently modifying its tree improvement strategy for *Pinus radiata* as a result of significant developments in four main areas - (i) economic breeding objectives, (ii) management of data and information, (iii) TREEPLAN® genetic evaluation and (iv) wood quality assessment. These developments have enabled the use of more flexible breeding and testing strategies which will lead to faster rates of genetic gain.

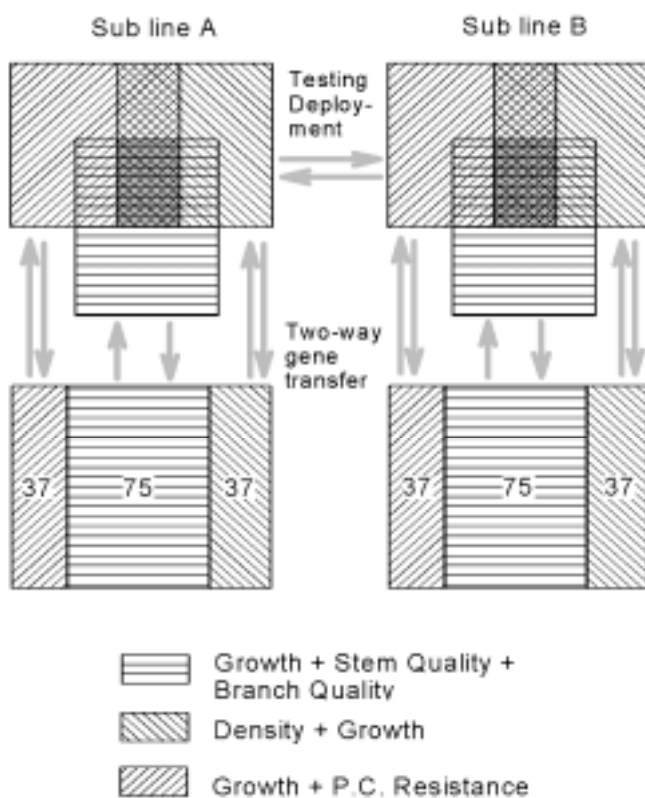


Figure 1. The 1993 STBA radiata pine breeding population structure

ECONOMIC BREEDING OBJECTIVES

Clearly defined breeding objectives are the first and most important stage in a tree improvement program as traits in the breeding objective are the traits that determine profit. Selection criteria, which are the traits measured in the breeding and selection programs, may or may not influence profitability of an enterprise. The definition of an objective is basically an economic, not a genetic issue.

In the past, breeding objectives for *P. radiata* were loosely defined and usually developed without the use of economic information. The selection criteria used historically by the STBA in the different selection indices for *P. radiata* (multi-purpose; high growth and density; growth and *Phytophthora* resistance) included stem diameter, branch quality, stem straightness, *Phytophthora* resistance, *Dothistroma* resistance and wood density.

After preliminary work done in 2000 the STBA in partnership with The Forest and Wood Products Research and Development Corporation (FWPRDC), and CSIRO Forest and Forest Products (CSIRO) initiated a research project to more clearly define the objective in economic terms for solid-wood products in *Pinus radiata*.

There are two main components in this project: (i) deriving economic weights for breeding objective traits and using them to develop selection indices and, (ii) estimating genetic parameters and their variance and covariance matrices for key breeding objective (rotation age) and selection traits (younger age).

To date two interim reports (Wu *et al.* 2004, Ivkovich *et al.* 2004) have been produced that provide data for use in the STBA *P. radiata* breeding program.

Wu *et al.* (2004) reported estimates of genetic variance and covariance matrices between breeding objective and early selection criterion traits. This work was based on the results from a significant number of wood disks, bark-to-pith wood strips, and bark-to-bark through pith billets from genetic trials nearing rotation-age. Wood strip samples were processed through Silviscan® and the data were used for estimating genetic parameters for growth and wood property traits from early to rotation ages. The other wood samples were processed in various ways to help in determining correlations between traits and to also develop more effective methods of field sampling for wood properties. Growth (DBH), stem straightness and branch size were also jointly studied with wood quality traits for the three sites.

Based on this work, genetic correlations, heritabilities, and genetic and phenotypic variance-covariance matrices were constructed by combining information for genetic parameters across the sites involved, and using information reported in the literature. These matrices will be used for deriving selection indices and for genetic evaluation purposes. The findings are currently being incorporated into the national breeding program for *P. radiata* by the STBA.

Ivkovich *et al.* (2004) reported results for a detailed study of the production systems of a number of STBA members involved in the growing and processing of radiata pine for solid wood products. They developed bio-economic models linking four breeding objective traits (MAI - mean annual increment, SWE - stem straightness or sweep, BRS - branch size, and MOE - modulus of elasticity) with the different production systems. Both industry confidential and published data were used. Economic impacts of the four breeding objective traits on the production systems were examined through the bio-economic model. Economic weights for the breeding objective traits were determined by estimating the improvement in overall profitability of various production systems as a result of changing trait values.

The economic weights developed in this study are now being incorporated into the STBA radiata pine breeding strategy through the development of selection indices. This will help industry to maximise profitability via increased genetic gain targeting solid wood production.

In addition to using the information in the breeding program, it will also be used by the STBA to develop deployment objectives specific for each members' business. This will permit members to maximise their individual benefit from tree improvement by making optimal use of the genetic information and genetic material available through the STBA.

MANAGEMENT OF DATA AND INFORMATION

The STBA currently has access to tree breeding data for about 500,000 trees and associated genetic material that has been developed over a sixty year period by members and the STBA. Data is also being captured on thousands of trees growing in current *P. radiata* trials planted by the STBA. This data is a valuable and a strategic asset that needs to be securely stored and yet made accessible for use by STBA staff and members alike. Prior to 2000 much of the data held by the STBA and/or its members was held in numerous *ad hoc* files. This made it difficult for staff to source and compile information for many purposes - statistical analysis, prediction of breeding values, management of germplasm and delivery of information to members.

The design for a data management system was developed over a period from the mid-1990s resulting in a document in 1998 that described in detail the specifications of an integrated software system to satisfy the STBA's requirements for data management.

Over the last five years the STBA has developed this system – the STBA-DMS™. This employs a web-based interface that makes access from multiple and diverse sites simple and flexible. The system has allowed the rapid update of genetic values and their dissemination to users. Use of these values within the operational programs to ensure best information is used as soon as possible after it becomes available.

The STBA-DMS™ not only stores performance data but also manages the pedigree for genetic evaluation purposes and is fully integrated with the STBA's TREEPLAN® genetic evaluation system.

Once information is stored in the STBA-DMS™ it continues to be available for future genetic evaluations (TREEPLAN®) ensuring that the STBA maximises the use of all information at hand. All trees that are stored in the system are allocated a unique genotype identity. This gives the ability to track pedigree both for use in genetic evaluation but also for general purpose use in crossing programs, trial measure and assessment *etc.*

TREEPLAN® GENETIC EVALUATION

The accurate prediction of genetic (breeding) values for trees in the breeding population is fundamental for the success of the STBA tree improvement program for *Pinus radiata*. The optimal statistical method for breeding value prediction is Best Linear Unbiased Prediction (BLUP). BLUP was first introduced to tree improvement in the STBA by Jarvis *et al.*, (1995) and is the preferred analytical method. In the past, consultants contracted by the STBA have used other methods, such as Best Linear Prediction (BLP), for the evaluation of *P. radiata* data and prediction of breeding values (White *et al.*, 1992ab). Although appropriate at the time, given the capabilities of computer hardware and software for processing the large amounts of data available, the use of BLUP will advance capabilities in genetic evaluation and help to maximise the use of genetic material within the STBA cooperative.

The STBA, in partnership with The Animal Genetics and Breeding Unit (AGBU) at the University of New England, has developed the TREEPLAN® genetic evaluation system that is customised for forest tree improvement and has been using it routinely in STBA breeding programs for over three years (Kerr *et al.* 2001, 2002; McRae *et al.* 2003). The software allows the STBA to meet its obligations to members and is a vital tool in helping to deliver maximum genetic gain in plantations per unit of time and cost.

Powell *et al.* (2004) summarise the procedures and methods used in estimating genetic values for trees in the national *P. radiata* database. They also list Breeding Objective (BO) breeding values for each genotype (tree) in the database for traits of commercial interest. The breeding values for volume (VOLUME), density (DENSITY), branching (BRANCH) and stem straightness (STEMST) are combined into an index with different weights applied to each trait. As discussed, there is currently limited economic information that allows a reliable set of economic weights to be used in the construction of selection indices. As an interim position an alternative approach was used, based on maximising the correlation with an earlier BLP-based index. The current breeding objectives project (Ivkovich 2004) has substantially improved our position in this area. The estimated breeding values for individual traits along with the combined index are important for choosing elite parents for population improvement in the breeding program and for propagation. The results are of commercial importance in choosing elite parents for grafting into seed orchards aimed at commercial deployment or crossing to produce smaller amounts of seed for use in the development of cuttings for deployment. The TREEPLAN® results can also be used to cull orchards and/or differentiate among seedlots that are currently available in the marketplace.

The TREEPLAN® system updates breeding values for genotypes in the national database on a regular basis as new data accumulates. Continually updated genetic values are readily available via the internet through the STBA-DMS™.



Figure 3. Regions used in defining *P. radiata* Selection Criteria used in TREEPLAN®

Genetic (breeding) values have been predicted for some 134,767 genotypes (different trees) in the population in 2004 and 21 selection criteria which are based on regionalised target production zones in Australia (Figure 3.). This included genetic values for genetic groups, first-generation (progeny of founder trees), second-generation and third-generation trees (progeny of crosses among selected parents).

Data was included from 78 trials including eleven STBA third-generation fullsib/polymix progeny trials. The flexibility of TREEPLAN® will enable genetic values to be region specific and to take account of any Gx E interactions.

As larger volumes of data accumulate from progeny trials over the coming years these numbers are expected to increase rapidly. This will greatly improve the accuracy and precision of breeding values for genotypes in the population. The opportunity will be

available for STBA members to realise greater genetic gain and increased profits in their commercial plantations.

The TREEPLAN® and STBA-DMS™ systems in combination will result in an ability to deliver marked improvements in the efficiencies of the STBA's breeding programs. Rapidly assessing the genetic merit of larger numbers of trees and then using the information to enhance crossing and progeny testing programs will enable real increases in selection intensity.

WOOD QUALITY ASSESSMENT

In the past, STBA breeding methods for radiata pine have concentrated on tree volume and form on the assumption that this would lead to more fibre and more profits. The first two cycles of selection in the STBA breeding population have increased growth significantly, while plantation owners have reduced rotation age from 40-45 years to about 30 years for commercial reasons. As a consequence, the proportion of juvenile wood in the harvested tree has increased to about 1/3 to 1/2 of total volume, a trend that is likely to continue. Hence the improvement of juvenile wood in terms of both quantity and quality is critical for the future of the softwood industry in Australia.

The STBA in conjunction with FWPRDC and CSIRO are funding activities which are focused on improving juvenile wood in radiata pine by combining information from broad-scale approaches in both quantitative and molecular genetics. Through acoustic testing of young standing trees and assessment of axial clearwood specimens together with collection and analysis of data from Silviscan® and WinDENDRO® X-ray scanning

systems the aim is to develop methods to accurately and economically predict MoE or wood stiffness on young trees.

Quantitative genetic analysis of the data being collected will estimate heritabilities, genetic correlations and breeding values for juvenile wood traits (density, microfibril angle (MfA), etc.) and the transition point between juvenile and mature wood. This information will be used to derive a juvenile wood stiffness index to be compared with acoustic results for prediction of MoE of young trees.

Molecular information is also being collected as part of the project (identification of candidate genes and quantitative trait loci (QTLs) for various juvenile wood traits). The aim is to combine quantitative and molecular information to predict breeding values and then select advanced material for breeding and deployment from STBA breeding trials planted widely across south-eastern Australia.

This recognition of the important role of wood properties is integral to the future success of the STBA radiata pine breeding program.

RADIATA BREEDING STRATEGY

The current breeding strategy for *P. radiata* (White *et al.*, 1999) was developed during the 1980s and implemented in the early 1990s. The strategy is continually under review. Although appropriate for the time, the focus was on theoretical aspects of the breeding strategy rather than the effective and efficient implementation of the program. New strategies will focus on total tree improvement, rather than just breeding, by integrating deployment with the breeding program. More effort is being directed towards the delivery of genetic gain per unit time. To achieve this, the generation interval must be reduced and selection pressure increased. Despite its strengths, there are several areas where the current strategy could be improved.

Revising the strategy

The STBA decided to review the strategy as part of an ongoing process. Strategically the STBA felt it was necessary to build some fundamental tools for data management and genetic evaluation as this would facilitate flexibility in the strategy and give an ability to quickly adapt to changes and enhancements.

Breeding objectives must be defined clearly and derived with the use of economic information. The selection criteria used in the different selection indices for *P. radiata* include stem diameter, branch quality, stem straightness, Phytophthora resistance, Dothistroma resistance and wood density. Clearly defined breeding objectives will ultimately determine the optimal suite of selection criteria for use in assessment and selection. In future, there will be increased emphasis on wood quality traits (juvenile wood, density, stiffness and spiral grain) and strategies for resistance to *Cyclaneusma* spring needlecast, Pitch Canker and Essigella aphid may be needed. The STBA is involved in projects that are starting to provide information on the breeding objective traits that will be crucial to the future success and relevance of the breeding effort.

BLUP is now the preferred analytical method for prediction of breeding values in trees and must be routinely applied. BLUP technology can overcome many of the statistical problems usually faced with 'rolling front' breeding and selection strategies and traditional methods of analysis. The STBA developed TREEPLAN® to ensure it has the analytical capability necessary for the effective use of BLUP.

A feature of the past strategy was a nucleus breeding system (Cotterill *et al.* 1988 and Cotterill 1989). A discrete nucleus and main breeding population is not retained in the new strategy. Nucleus breeding was largely adopted in animal breeding for situations (limited reproductive rates in females) not found in *P. radiata*. Although nucleus breeding has other advantages in management of inbreeding, there are now better and more efficient selection and mate allocation methods available.

Discrete generation breeding, a major component of the past strategy, leads to an inefficient use of resources. Annual budgets and work programs fluctuate widely with 'peaks and troughs' of activity. A 'rolling front' strategy with overlapping generations has been adopted. The advantages include: operational efficiencies, more stable budgets across years, increased selection pressure with a larger program, earlier adoption and use of elite genotypes for breeding, ease of infusing new material, reduced risk, better account of GxE and enhanced gains. A rolling front strategy also facilitates a more rapid response to changes in breeding objectives. The development of TREEPLAN® has been essential for the successful adoption of a 'rolling front'. Better selection and mate allocation tools will be needed for management of pedigree and inbreeding.

The use of pollen mix crosses for general combining ability (GCA) testing is not essential. In the revised strategy, there is no longer a discrete 'main' population. Parents will be used in several crosses (minimum 3 to 4) and more effective use of pedigree is made using BLUP evaluation.

The current practice of using independent breeding sub-lines will continue, at least in the short-term. This aids in managing inbreeding and ensures deployment populations are outcrossed. The size of the breeding population (340) can be reduced and effective population size managed with selection and mate allocation genetic algorithms.

Where required (subject to the findings of the breeding objectives project) there may be multiple breeding sub-populations with 30-40 parents targeting different breeding objectives. Separate deployment 'breeds' will be developed with two sub-lines per breed. In the interim, three breeds (Multi-purpose, High-Growth and Phytophthora resistant) will continue to be used for breeding.

The revised strategy will review the importance of clonal propagation in progeny testing and its role in deployment. Presently in Australia, clones or cuttings are used as a propagation method largely to facilitate family forestry. Technologies (tissue culture and somatic embryogenesis) are now available to facilitate the use of clonal systems. Selection pressure can be greatly enhanced and both additive and non-additive genetic variance exploited with the deployment of tested clones.

Integrated breeding facilities must also be established by the STBA to better manage the breeding population. The population of parent trees must be available on a central location to facilitate cross-pollination activities on an annual basis. Flowering success and synchrony are necessary to increase selection pressure and optimise parental combinations, and ensure that the STBA pursues gain per unit time.

The Future

The STBA has access to a highly valuable genetic resource and is capitalising on this member investment by focusing on genetic gain per year and delivery of profit to its stakeholders. Third generation progeny trials are currently being assessed and consolidation of the program is critical. The strategy review process will be dynamic with changes adopted as opportunities are identified. Projects which target key areas have been developed and are now largely in place (breeding objectives, genetic evaluation, data management, integrated breeding facilities, clonal deployment, and selection and mate allocation). These will strengthen the breeding strategy and result in enhanced genetic gain. Future revision of the tree improvement strategy for *P. radiata* will focus on implementation issues and optimising gain via an appropriate and relevant breeding objective.

The breeding strategy is being revised in stages. The STBA had to first develop the tools needed to overcome well known technical difficulties that have plagued many breeding programs in the past. There is little point in having an optimal strategy with inherent weaknesses. The most important of these is the management of pedigree and performance information. The development of the STBA-DMS™ has overcome this limitation and the development of TREEPLAN® has given the STBA the analytical tool it needs to produce accurate breeding values efficiently and on a regular basis. Multi-site, multi-trait, multi-age and multi-generation analyses can be done, thus allowing the adoption of more efficient rolling front strategies. The last major deficiency (or opportunity) is then efficient selection and mate allocation. The STBA has proposals in train that will address this issue and enable the management of inbreeding more efficiently than by the use of unrelated sub-lines.

Research activities supporting the program are currently focusing on the continued development of: TREEPLAN® genetic evaluation software; the definition of economically derived breeding objective functions; an enhanced understanding of, and ability to use, GxE interaction in genetic evaluation; studies of the juvenile wood properties in the *Pinus radiata* breeding population.

CONCLUSION

The STBA is putting in place the key tools needed to effectively manage large scale breeding programs on behalf of its members. In relation to the radiata pine breeding program the switch to an annualised, rolling front program of crossing and progeny trial establishment, in combination with better ways to manage key genetic and resource information will see major benefits coming to fruition over the next few years. The result will be

the deployment of highly genetically improved material in members plantations and the ability to advance the breeding population with a speed and a simplicity not contemplated a few years ago.

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