Section 1: NZDFI Breeding Research Plan

Table of contents

Introduction

Until the 1970s several countries in the Southern Hemisphere tested numerous *Eucalyptus* species, aiming to find a small number of species that could be used at a large-scale in commercial forestry (Brazil, Chile and Uruguay in FAO 1981). The most promising species were selected and work began on expanding the sampling of genetic diversity and establishing breeding programmes. As an example, Brazil focused mostly on *E. urophylla* x *E. grandis* hybrids, Chile on *E. globulus* and *E. nitens*, and Uruguay on *E. globulus*. The pulp industry is the main end-user of these plantations.

New Zealand followed a similar initial trend, testing dozens of species in the 1970s, with the work done by the Ministry of Works and Development Plant Division, NZ Forest Research Institute and later Scion. Only a few of the species reached enough popularity in New Zealand to be considered for commercial forestry (most notably *E. nitens*– a pulp species). While *Eucalyptus* are popular in the farm forestry community—where farmers have planted dozens of species there has been little exploration of the genetic variability for each species.

The New Zealand Dryland Forests Initiative (NZDFI) was established in 2008 as a collaborative tree breeding and forestry research project to improve drought tolerant eucalypts that produce high quality naturally ground-durable hardwood required for New Zealand’s agricultural, transport and energy sectors as well as specialty wood products for export to international markets.

The NZDFI vision is for New Zealand to be a world-leader in breeding ground-durable eucalypts, and to be home to a valuable sustainable hardwood industry based on 100,000 hectares of eucalypt forests, by 2050.

NZDFI’s unique research focus and strategic vision will benefit future generations of New Zealanders by delivering to NZ forest growers the plants and knowledge to select and grow a eucalypt species suited to their site and to learn how to economically produce a high quality durable wood product that meets the requirements of domestic and international markets.

NZDFI is providing a new option to grow durable eucalypts that will diversify grower income and enhance environmental sustainability by combating soil erosion due to eucalypts extensive root systems that coppice following felling. These new hardwood forests can offer benefits beyond wood production and will sequester carbon while supplying bees and native fauna with nectar/pollen. The species have a low wilding risk.

NZDFI pursues a strategy that involves carefully matching sites to species and end-products, with a focus on sites that have a rainfall <1000 mm/year and regions that need new land use options that can diversify regional economic development. Therefore, rather than looking for generic species we are looking for species that complement New Zealand's main plantation species and target high-value products.

At the same time, we need growers to learn and understand how to successfully establish these new species to ensure our target of 100,000 hectares is planted to produce a sustainable resource for a new hardwood industry.

Species selection for NZDFI's breeding programme

Species testing prior to selection

In 1991 the Special Purpose Species group of the NZ Forest Research Institute included naturally durable species in its research programme (McKenzie, 1993). Existing stands were surveyed to evaluate species performance, and identify site requirements.

Further to this, in 2003, FRI planted a set of research trials of 12 species from the stringybark group.This research was focused on testing both a wider selection of durable species for survival and early growth. (McConnochie *et al*, 2008). These are classified within the monocalypt group with most being class 2 and 3 on the Australian standard. (AS 5604:2005). Some were already proven in Northland and Bay of Plenty's warm environment, and seem to be resistant to most eucalypt pests and diseases currently in NZ. They also saw well producing stable durable timber. Linked to this work was the establishment in 2004 of over 40 small trials throughout NZ by the Eucalypt Action Group of the NZ Fram Forestry Association (Gordon, 2007).

In Marlborough, between 2003 and 2006, over 80 small research trials of 25 durable eucalypt species were established in an early research joint venture between Vineyard Timbers, the Marlborough District Council, Proseed NZ Ltd and several private landowners in Marlborough. These included a significant number of species of the symphomyrtus group of which a number where there was no record of being previously tested in New Zealand. Despite some of these being palatable to eucalypt pests, several that produce class 1 durable timber have proven to be medium to fast growing, with some that are frost and drought tolerant.

Several stringybark species of interest to Scion and NZFFA were tested in the trials.

A full list of those species tested in Marlborough from 2003-2006 is shown in Appendix 1.

NZDFI tree breeding species selection

Based on this extensive species testing NZDFI has made an informed choice of species at the outset, concentrating efforts and resources on breeding a few promising species that are drought tolerant and produce very durable heartwood in cool climates. Moreover, instead of going for 'all rounders' we have made the conscious decision to tackle specific environments and durable solid wood products and markets. The five species for genetic improvement (scientific name, common name, wood durability class) are:

E. argophloia	Western white gum	*Class 1
E. bosistoana	Coast grey box	*Class 1
E. globoidea	White stringybark	*Class 2
E. quadrangulata	White-topped box gum	*Class 1-2
E. tricarpa	Red iron bark	*Class 1

*Australian Standard, AS5606-2005

All species readily coppice and this presents new opportunities for propagation and forest management. There is potential to produce hybrids to capture selected traits within the pure species, to broaden the market and increase the value of the wood products.

In addition, with the NZDFI well set up by 2011, the testing of several other durable promising species was expanded beyond the initial regional focus that had developed in Marlborough. A suite of six secondary species of interest were identified and deployed (along with the chosen breeding species) in a series of species demonstration trials in 2011 and then management trials in 2013 and 2014 from Bay of Plenty to north Canterbury. These are described in the section titled 'Species Demonstration and Management Trials' on page 20.

General principles behind the breeding strategy

Genetic gain is queen

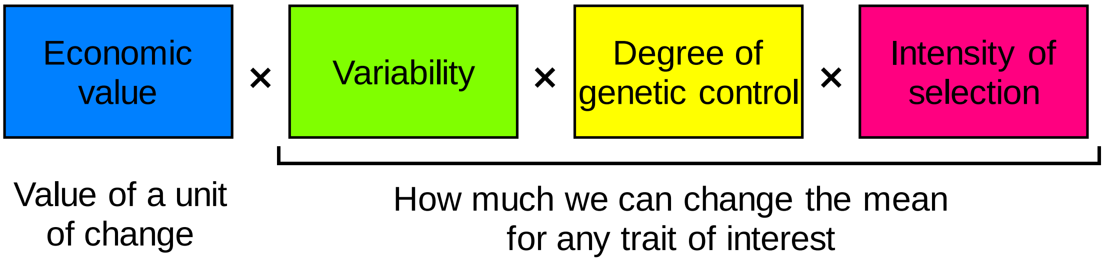
Breeding programmes should be evaluated on their ability to deliver genetic gain; that is, on the difference between the means for the traits of interest between the current generation and the next. Genetic gain from selection can be estimated using the breeder's equation:

$G = (i\ r_{IH}\ \sigma_H)/\text{time}$

where:

1. i is the selection intensity, a measure of how many trees we are selecting,
2. r_{IH} is the accuracy of selection, which is the correlation between what we are assessing when selecting and what we want to breed for. In the simplest situation (mass selection on a single selection criterion, identical to the objective trait) the heritability is the accuracy of selection. For more complex selection (for example, indirect, early selection for multiple traits) the calculation is more complex and needs estimation of the heritability for all criteria and traits, as well as the genetic correlations between them.
3. σ_H is the variability of the trait (or combination of traits) we want to improve.
4. time needed to achieve the gain.

That is, genetic gain per year is *directly proportional* to selection intensity, accuracy of selection and the genetic variability for the traits under selection. Gain per year is also inversely proportional to the time required to achieve said gain, highlighting the benefit of early selection.



As delivering genetic gain is the core function of a breeding programme, the NZDFI general work principles are directed to the breeders' equation:

1. We are aiming to capture as much variability as possible for each of the NZDFI species. Shallow species introductions (with too little genetic variability) will yield poor genetic gain. For the NZDFI species there was very little genetic material in New Zealand, which required extensive seed collections and imports from the Australian natural populations.

2. There are trade-offs between selection accuracy and reducing the timeframe for selection. Many of the selection criteria require the trees to be greater than 5 years old for accurate trait assessments. A less accurate technique will be applied if this substantially reduces the testing time and increases the number of individuals that can be sampled.

   Furthermore, there are principles that apply to eucalypts in general:

3. The performance of eucalypts is more site-specific than radiata pine for example. Therefore, testing over multiple environments is essential. The natural populations of most eucalypt species are highly variable with a propensity for hybridising and selfing. Large family collections can be rogued to remove these poor individuals and still retain enough elite individuals as the basis for the breeding programme.

4. Natural seed production of some of the species is seasonally sporadic and collections were required over several years to increase the number of families available for testing. As a result, rather than delay planting progeny trials until the target number of families were obtained, smaller trials with fewer families were established over successive years.

Exploiting variability

Every tree-improvement program is based on the exploitation of variability, for which there are three essential steps: selection, mating and testing. Selection will be based on improving productivity and wood quality traits, mating will depend on reproductive biology and economic considerations (open-pollination versus control-pollination), and testing will depend on statistical considerations.

The breeders' equation dependence on selection intensity highlights the requirement for large base breeding populations to provide: (1) a description of genetic architecture (degree of genetic control and association between traits) for each species, (2) estimates of genetic superiority for the best material, (3) selection for multiple traits and (4) superior material for the future generations.

There are two scales of variability utilised in the structure of the breeding programme: between – and within family. The targeted size of the NZDFI breeding populations is 100 families and the number of individuals tested within a family is 50 – 80 trees. Progeny trials are generally planted at three sites, giving a total of 150 -240 trees per family. This could be considered too small but can sustain appreciable genetic gain over multiple generations (White 2001). Small populations will accumulate inbreeding faster than larger programmes; NZDFI is continually looking for opportunities to increase population size, as some original seed collections have been limited by poor flowering years.

Apiolaza (2009) simulated open-pollinated populations with three traits, with a size ranging from 1000 to 6000 trees (e.g. 100 families with 60 trees each), to study how many trees are in the top 10% or 20% for each of the selection criteria. The simulation considered traits with low (e.g. growth, $h^2 = 0.25$), medium (e.g. wood stiffness, $h^2 = 0.50$) and high (e.g. basic density, $h^2 = 0.75$) genetic control under three scenarios of genetic correlation: no association between all three traits (zero correlation, independent traits), intermediate positive correlation (0.50) between all three traits and intermediate negative correlation (-0.5) between the traits.

Table 1 shows that with 1000 trees we would find between 0 and 14 trees in the top 10% for all traits depending on the assumed correlation. The number of available trees increases substantially when we reach 6000 trees in a trial.

Table 1: number of trees in the top 10% and 20% for all three traits under three scenarios of genetic correlation.

	Uncorrelated		Positive correlation		Negative correlation	
	Top 10%	Top 20%	Top 10%	Top 20%	Top 10%	Top 20%
1000	0.9	8.4	14.0	46.6	0.1	1.9
2000	2.2	15.2	26.7	91.8	0.1	3.7
3000	3.1	23.5	41.4	138.7	0.2	5.6
4000	4.0	32.4	54.4	185.8	0.3	7.3
5000	5.1	41.0	69.9	229.6	0.5	9.3
6000	5.6	49.0	83.0	275.6	0.5	11.5

In addition, it is essential to understand that *Eucalyptus* species often present highly structured spatial variability, with ‘provenances’ expressing different trait means. For example, *E. globulus* (Dutkowski and Potts 1999) and *E. nitens* (Dutkowski et al. 2001) show large variability for population means for multiple traits, which affect the profitability of the provenances (Apiolaza et al. 2005).

As an example, *Figure 1* shows variability for heartwood quantity (in pink) in a small *E. globoidea* provenance row plot stand harvested at age 5 years. Some trees have greater than 80% heartwood content while there are others that have very little heartwood, independent of disc diameter. This highlights the importance to sample broadly so as to capture the variability across the range of the species natural populations.

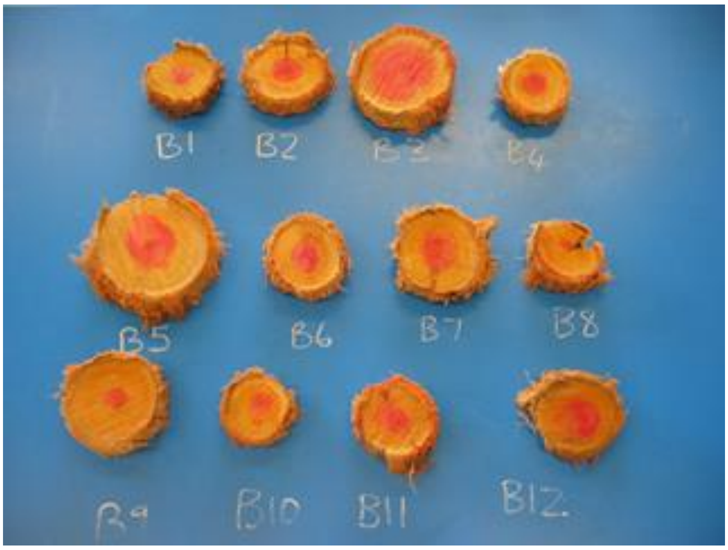


Figure 1: example of phenotypic variability for heartwood quantity (in pink) in a small durable *Eucalyptus globoidea* stand age 5 years. Some trees barely show signs of heartwood, while others reach over 90% of the stem section.

Genetic material: how much and from where?

For most of the species selected by the NZDFI for genetic improvement they have never undergone any formal domestication and there is very little genetic material in New Zealand.

Therefore individual family seedlots were purchased from seed providers in Australia where available however these were very limited. Proseed NZ Ltd contracted extensive seed collections to provide broad based genotypes of *E. bosistoana*, *E. quadrangulata* and *E. globoidea* from across the natural range of these species. As additional seed becomes available it will be included into the breeding programme. For example 25 new *E. bosistoana* families were purchased in 2016, and another 30-50 families will be collected in early 2017.

The plan for collecting seed for the NZDFI Project requires *"the collection of sufficient seed from a minimum of 100 families to establish genetic progeny trials of these eucalypt species as well as management trials needed to evaluate post, pole and timber production systems. These trials will form the base populations for our future tree breeding programme."* (NZDFI Strategy 2008)

Species collections

1. *Eucalyptus bosistoana* (Coast Grey Box)

Natural Range: Coastal Victoria/NSW

E. bosistoana is found along the coast of eastern Victoria and up into NSW, just south of Sydney. It is a Class 1 durable species and historically was used for poles, sleepers and fences in Australia. The species was almost cut to exhaustion in the 1920s because of its high durability and stability. There has been no previous structured seed collection or breeding programme for this species.

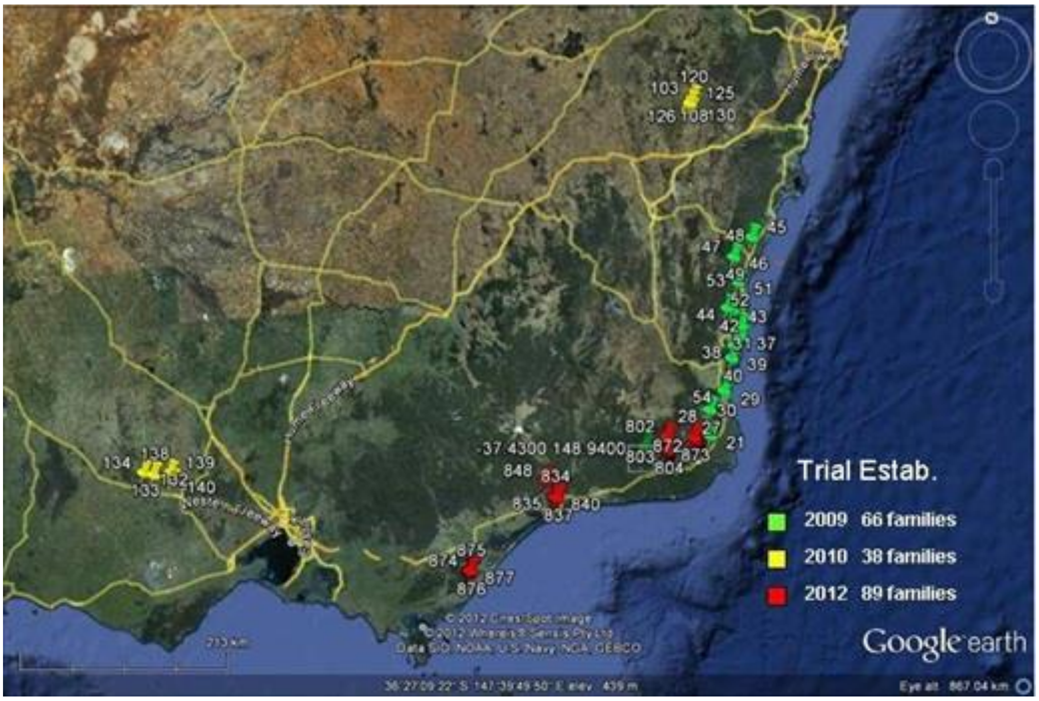


Figure 2: Location of NZDFI *E.bosistoana* seed collections

Eucalyptus globoidea (White Stringybark)

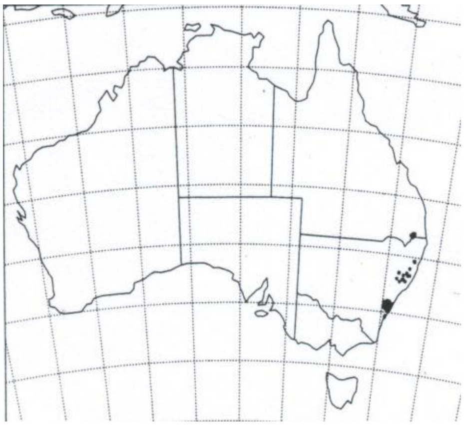
Natural Range: Central NSW and Coastal Victoria/NSW

E. globoidea occupies much of the same coastal range as *E. bosistoana* along coastal eastern Victoria and southern NSW. It is found on a variety of sites including gravelly loams, clays and skeletal soils. *E. globoidea* is rated as a class 2 durable hardwood with densities around 900 kg/m^3 in mature trees. There are plantings of *E. globoidea* throughout the North Island and NZ plantation material is reported to have sawn well with densities ranging from 527 to 623 kg/m^3 with modulus of elasticity of around 14 GPA. There is a small breeding programme in NSW.

Figure 3: Location of NZDFI *E.globoidea* seed collections across natural range

3. *Eucalyptus quadrangulata* (White-topped box)

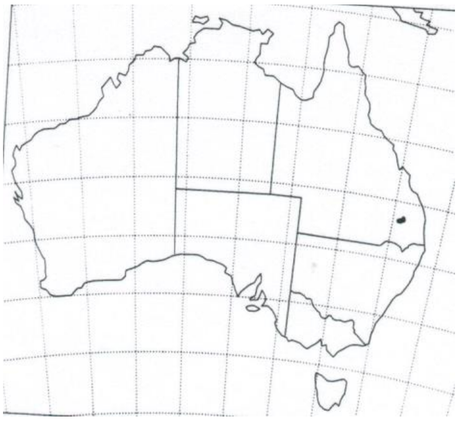
Natural Range: Southern Queensland



E. quadrangulata is found on the better soils of the coast and adjacent ranges in New South Wales and southern Queensland. It occurs in the tablelands of central and northern NSW where the mean annual rainfall is 900 – 1700 mm. Forests NSW have evaluated *E. quadrangulata* for its suitability as a plantation species however it does not feature in any breeding programme.

4. *Eucalyptus argophloia* (western white gum)

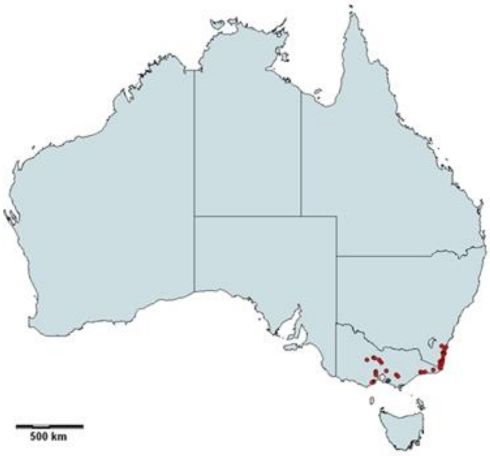
Natural Range: Southern Queensland



E. argophloia is only found naturally in southern Queensland, north east of Chinchilla. Despite its restricted distribution, the species can grow on a wide range of sites including those with heavy soils and frosts. The wood is a deep red and durable with high basic density. It is classed as a nationally threatened species and is protected under the Queensland Conservation Act.

5. ***Eucalyptus tricarpa* (red ironbark)**

Natural Range:



E. tricarpa occurs in southern New South Wales and is common in the Bendigo goldfields area, Gippsland and central Victoria, extending south to Anglesea.

E. argophloia and *E. tricarpa* have small breeding programmes in Australia and family seedlots were obtained from DPI Queensland and Forests NSW. Both species are closely related to *E. bosistoana* and offer the opportunity for hybridization to exploit wood properties such as colour and hardness.

Breeding objectives and selection criteria

In principle, one of the first steps when developing a breeding programme is to specify the breeding objective; that is, the list of tree traits that affect profit (often measured at rotation age) multiplied by their respective relative economic importance. In practice, that needs a good understanding of the value chain (including plantations and processing facilities) and markets intended for the species being improved, which in New Zealand has principally focused on commercial pulp wood production.

A distinctive feature of the NZDFI breeding programme is the clear definition of the end product wood qualities. Therefore, the trials are planned to be large enough to capture a broad range of genetic diversity from which to make very early and intensive selection for the key genetic traits for improvement. These are:

1. **Wood quantity:** stem diameter, tree height, stem form.
2. **Wood quality:** durability (extractive quantity and heartwood volume) and growth-strain (splitting-test). Data will also be available for stiffness (acoustic resonance), density and shrinkage (water displacement), but these traits will only be monitored to ensure that selections will not adversely effect the excellent wood properties of these species.
3. **Adaptation:** ability to coppice (propagation), survival to frost, drought, and pests.

Genetic evaluation

Each species will be initially evaluated in two types of trials:

1. A standard progeny test.
2. An early growth-strain screening trial.

Progeny Tests

All NZDFI breeding populations are tested as standard open-pollinated families at 3 contrasting sites across the Marlborough, north Canterbury, Wairarapa, and Hawkes Bay regions. Planting at three different sites will identify the relative contributions of genetic and environmental effects on the genotypes under test and reduces the consequences of damage or loss at a site. The sites are selected based on climatic factors, accessibility, and uniformity. Legal documentation with the landowner is completed to secure ongoing access for on-site operations and collection of germplasm.

Each site contains between 50 and 100 families with 30 or more trees per family (depending on seedling availability). All sites follow a single-tree-plot alpha-lattice incomplete block design. Spacing is 1.8 m within the row and 2.4 m between rows and equates to 2312 sph.
An establishment report is produced at the completion of the trial establishment. Trial records for each site have:

- Trial Description Form – Site information (location, landowner, experimental design)
- Location Map – including GPS coordinates
- Plot layout map

The standard open-pollinated trials are evaluated for height and survival a year after establishment. At age 3-5, trees are assessed for stem diameter and form. At ~7 years of age all families are assessed for heartwood quantity and quality (extractive content), growth and form. The timings of these assessment are indicative only, and are species and site dependent. The growth rate of these species at each site is not well defined. There may also be changes made in sampling techniques and other parameters added for measurement. In addition, the NZDFI monitors flowering at the trial level to support future deployment and breeding decisions. This involves recording when trees have one or more of the following; flower bud development, inflorescence present, seed capsule development and mature seed present. The NZDFI trial design allows for substantial within family selection and within-trial redundancy, with up to 80 trees for genetic entries, this permits the overlaying of other experiments. (e.g. wood properties, silviculture, forest health).

Early Growth-Strain Screening

It has been demonstrated in several studies that growth-strain in eucalypts is heritable. Therefore the opportunity exists to breed eucalypts with low growth-strain that are suitable for solid wood processing. A technique unique to the University of Canterbury allows the rapid screening of large sample numbers of 1-2 year old trees. (Chauhan and Entwistle, 2010). Whereby the measurement of growth-strain has previously been laborious and expensive this process can test 1000s of samples relatively efficiently. The technique is destructive and therefore the progeny trials cannot be used to screen for this trait. The breeding populations have been planted in a uniform tree nursery environment. The families are planted in blocks of 8 trees are replicated between 2 and 8 times, depending on the number of available seedlings per family, and the families are randomly allocated within the replication.

The outcomes of this trial are

1) A family ranking for growth-strain and (other wood properties i.e. stiffness, density and volumetric shrinkage). This information will be used to ensure that selections from the older progeny trials will priories those families with favourable properties.
2) A 2nd generation breeding population which is rouged from high growth-strain and slow early growth. Approximately a quarter of the planted trees will be propagated from cuttings taken from the coppice and new clonal trials will be established. The selection criteria are to maintain genetic diversity and the top individuals in the assessed traits. Therefore the top individuals of good form in each family for each trait (fast growth, low growth-strain, low density, high stiffness) will be selected with an additional global index selection for growth-strain and diameter.
3) A first generation deployment selection. Approximately the top 40 individuals (from families known for good heartwood) will be multiplied to create mother plants for a mass propagation in 2019. The 1st stage of improved material for the establishment of durable eucalypt plantations.

Woodville is the first trial where selections have been made, based on within-family selections and aiming to maintain as much genetic diversity as possible. An empirical estimate of genetic gain from this selection is obtained by taking the difference between the breeding values for the selected trees as a difference from the population mean. As a reference, the population means for traits of interest were: 11.19 GPa (stiffness), 2071 us (growth strain), 0.82 (dry density), 36.5 m (stem diameter).

When selecting trees in Woodville, emphasis was put on stem diameter and growth strain. The selected population shows a 0.5 GPa reduction of stiffness, while there was a 9 mm increase for stem diameter and 440 us reduction for growth strain. The average wood density remained unchanged. While there was a small trade-off between growth strain and wood stiffness, there was a substantial (21%) difference for growth strain. It is important to remember that at this stage we do not have a full set of genetic parameters for all traits nor do we have clear technical thresholds for growth-strain to aim for (we only know that lower is better).

Forest Health and Insect Screening

Eucalypts, like any plantation species, can suffer from insect and pathogen attack that can limit establishment, reduce productivity or cause tree death or deformity. As such the ability of breeds selected for elite growth and wood properties to thrive in the presence of established and future biological threats is fundamental to the success of this program. Furthermore a large part of our research programme is site-species and site-genotype matching and this will also reduce risk

The paropsine beetles are the main pests to affect both New Zealand and Australian eucalypt plantations. Of the 400+ species native to Australia several are already established and causing defoliation problems in New Zealand, while others are expected to arrive in the future.

However, many eucalypt pests show strong host preferences and there is a significant degree of variability in the susceptibility of eucalypt species, and even individual trees, to insect attack. Furthermore, some eucalypts can withstand substantial defoliation before growth impacts occur. To this end, the NZDFI trials provide a unique opportunity to assess and select those breeds that show the greatest and most consistent pest tolerance. Due to the geographic spread of trial sites, we will also have the ability to assess environmental impacts on any heritable measure of susceptibility to incorporate into evaluation of site-species matching.

As a number eucalypt pests are currently well established in New Zealand, occasional damaging outbreaks do occur on species such as *E. nitens*. If large enough, these outbreaks may still have the potential to affect NZDFI species selected for high pest tolerance. To ensure economically and ecologically sustainable management of these pests we will also assess the growth impacts of defoliation and the population biology of key pests to provide recommendations on when and how to manage outbreaks.

Our forest health program will have 3 main foci which are further outline in Appendix x:

1) Screening species and families for tolerance to established insect pests across sites using both natural defoliation in the field and manipulative experiments
2) Developing assessment tools for faster and more accurate insect pest monitoring for use land owners to use in their own Integrated Pest Management plans
3) To assess the impact insect defoliation on growth and recovery of young trees to assist in the development of damage thresholds for intervention (i.e. when is it necessary to manage pests).

Clonal Testing

Clonal testing provides more reliable breeding values and delivers higher genetic gain. The NZDFI eucalypt species all readily coppice and present the opportunity to produce cuttings derived from stump coppice growth. Selected ortets from progeny trials will be felled and resultant clones tested across sites. Generally 6-8 ramets per clones are required at each site. Clonal testing will be dependent on successful rooting and vegetative multiplication technology. The early growth-strain screening trial will managed after felling to produce coppice for clonal testing. These trials will test these low growth-strain selections for growth form and confirm wood property traits.

Scaling up assessments

The distinction between an objective trait and selection criteria in tree breeding can be subtle or extremely delineated.

1. An example of a subtle difference is when stem diameter and tree height is used to estimate tree volume at selection age and at rotation age. Breeders are measuring the same variables at different ages, and rely on age-age correlation to predict later performance.
2. An intermediate example is the forest industry using increment cores to estimate basic density for the whole tree. In this case, only a small portion of the tree is used at 1/4 of rotation to predict density for the whole tree at a later age. This section of the tree is extrapolated to the whole and age-age correlations applied.
3. Indirect assessments are used, like acoustics for predicting wood stiffness or Near Infrared Spectra for the prediction of chemical composition. This type of assessment relies on physical and/or chemical principles to predict the variable of interest and is often extrapolated from a small sample, with indirect measure and different ages.

Researchers and industry tend to feel more comfortable with subtle distinctions or even with intermediate examples; however, often it is practically impossible to use them at the scale required to evaluate thousands of individuals. In addition, for some traits it makes sense to redefine the aim of breeding. Both reasons relate to the general principles of genetic gain to capture variability and attain selection accuracy.

Early screening is targeted at assessing the worst part of a tree. Like MoE for radiata pine, wood durability (i.e. extractive content) increases from pith to bark. It is important to make sure that the worst wood is good enough for the intended use. Furthermore, early screening reduces trial costs, the environmental and financial risk associated with long-term trials and ensures timely deployment of improved material.

Data analysis

Each selection criteria in the standard open-pollinated progeny trials is analyzed using generalized linear mixed models with a multivariate approach, where performance in each site is considered as a different variable. If appropriate, the model will include spatial structure to account for microenvironment trend,

The early wood quality trial can be considered as one more site, with the additional potential complexity of left-censoring for growth-strain (Davies et al. 2016).

Following the analyses, we will have breeding values for growth, form and durability from the older open-pollinated trials, together with breeding values for wood properties and growth at age 2. An important point is that there is complete information at the *family level* (families screened for all the wood quantity, wood quality and adaptation variables), but all information at the *tree level* is partial (as no tree can be assessed for all variables). Breeding values at the tree level can be obtained using the relatedness between trees of the same family.

The assessment age for durability is currently being re-evaluated. Early data suggest that at age 7 years there is still a substantial proportion of trees that do not yet display heartwood. Assessment age is species dependent.

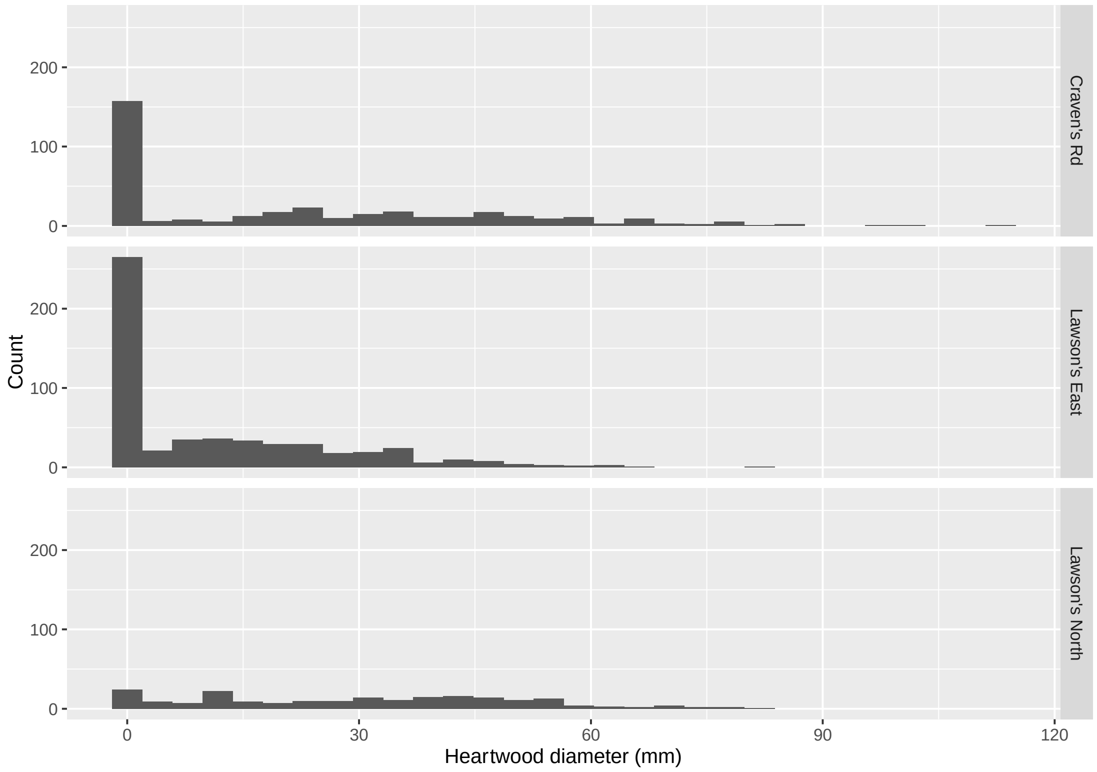


Figure 4: Heartwood diameter for *E. bosistoana* in the Lawson's and Craven's trials (2016 data collected by Yanjie Li).

Selection

The genetic analysis will produce a set of genetic values for all parents and individual trees for all traits. However, instead of selecting the best trees overall, restrictions will be set to account for two issues:

1. Seedlots selected from natural populations contain varying levels of inbreeding, depending on size and structure of the parent stand (e.g. Borralho and Potts 1996, Hardner et al. 1996). Differential family-level inbreeding can affect the rankings and poor initial performance can likely be more of a proxy for stand structure than for parental genetic worth.
2. Individual tree BLUP[1] tends to concentrate selection in too few, highly related individuals, drastically reducing population variability. There are a number of suggested alternatives to minimize this problem (e.g. Grundy et al 1994). A constraint on variability can be applied so the breeding programme sustains a larger number of unrelated and non-inbred trees – that is, status number. (Lindgren and Mullin 1998). This number is a measure of diversity, and it relates to the number of unrelated and non-inbred trees that would have an equivalent diversity to the population under study. From the long-term perspective, genetic diversity should not be constrained too early in the program.

A combination of animal-model BLUP and constrained selection would also account for the different sets of trees over which we are assessing the traits. During

---

[1] Best Linear Unbiased Prediction is the standard statistical genetic evaluation approach used in modern tree breeding programmes.

the first generation we will not completely rogue any family level, concentrating our selection efforts within families.

At this point the assessment of progeny trials and the Woodville growth strain trial will be combined in a selection index so as to identify the very poorest families to not include in the first generation.

Two traits that breeders aim to increase (basic density and wood stiffness) have already very high means in the NZDFI populations (at least when compared to *Pinus radiata*). Therefore, although these traits are being assessed, we may want to only constrain selection to avoid any significant decreases.

Data Management

NZDFI has a large network of experiments with an increasing database relating to each. This information is currently held by individuals contracted under the NZDFI project by the Marlborough Research Centre as well as various staff and students at the School of Forestry and not all being centrally accessible. In 2015 the decision was taken to import all NZDFI trial data into the database system Katmandoo developed by Department of Agriculture and Fisheries, Queensland.

Katmandoo is a data management system created for use in plant breeding to assist with the selection of superior genotypes with a range of traits (eg. yield, stem and wood quality characteristics and disease resistance). It aims to provide a single tool for managing phenotypic and genotypic data at both an individual and multiple experiment level.

The *Katmandoo* system consists of two (separate) fundamental components: a relational database to store the information and a software application to access it. The database structure can be designed to accommodate any information capture required by the user. The database includes:

- trial register information - trial type, location, design
- data management – individual tree assessment data
- pedigree management
- seed inventory subsystem

Deployment Strategies and Gain Assessment

Any effective tree improvement program incorporates both a breeding strategy for the ongoing genetic improvement of a species and a deployment strategy by which the genetic gains are captured rapidly and effectively for operational plantings via the use of seed or vegetative propagules. The following will be undertaken:

1. Conversion of unpedigreed planted stands to **seed stands** by selective thinning to produce a source of improved seed.
2. Conversion of progeny tests to **seedling seed orchards** by selected thinning and removal of poor performing families for the production of high quality seed. Other progeny trials will be retained with a full representation of the breeding population for the continued collection of data and as an archive of genotypes.

3. Establishment of **clonal seed orchards.** Selections will be made in breeding trials and grafted for producing seed with high genetic gain.
4. The potential advantages of a system based on **vegetative propagation** include: overcoming shortages of the best genetic material; rapid capture of increased genetic gains as the whole superior genotype is secured; enhanced uniformity of product; and matching of genotypes to specific sites.
   Juvenile coppice cuttings from elite selections can be used for clonal testing and future deployment.

Clonal seed orchards will be created from selections of the best individuals from the breeding trials; that is, they will consider individual tree growth and heartwood assessments at age 7 years, as well as family-level performance for the age 2 growth-strain test in Woodville. Selected trees will be grafted and an orchard established at Proseed NZ, Amberley or a site identified for exceptional flowering for the species. Orchard composition will be monitored, keeping track of any new information (assessments, traits) coming from breeding trials.

The strategies are designed to produce a range of genetically improved germplasm over time. The timetable for the delivery of improved seed will be dependent upon the degree of improvement required; existing genetic resources; species biological characteristics and the short- and long-term demand for improved seed.

Genetic gain trials are used to provide an accurate ranking and long-term evaluation of the performance of the different sources of genetic material. As new improved germplasm becomes available from the breeding programme genetic gain trials will be established with industry partners.

It is expected that genetic gains can be optimised in future deployment populations, moving to selections of best individuals across all families and removing complete families.

Hybrid development

Eucalyptus hybrids are of great importance in plantation forestry in some countries; for example, China, Congo, Brazil and South Africa (Eldridge *et al.*, 1993). The reason for the development of inter-specific hybrids in forestry is the capture of complementarity traits of certain pairs of species. However, only a few pairs of species—most notably *E. urophylla x E. grandis* in tropical environments—have been commercially successful, “when full account is made of losses through the life cycle, a picture of high incompatibility and inviability often emerges” (Potts and Dungey 2004). Therefore, the development of hybrids is high risk and high cost. Significant genetic gain and species adaptability can be achieved from improvement within the main species (*E. bosistoana, E. globoidea* and *E. quadrangulata*).

Nevertheless, a small number of selections of *E. argophloia* and *E. tricarpa* have been established in progeny tests for their potential to hybridize with *E. bosistoana* to introduce red timber colouring and pest tolerance. The best trees will have pollen collected and crossed in an *E. bosistoana* clonal orchard to test the viability of hybrid production. They are not being considered for development as pure species beyond one generation

Demonstration Species and Management Trials

The NZDFI has restricted the number of species for genetic improvement, concentrating efforts and resources on a few promising candidates. However, a broader list of species of interest is being tested in demonstration species and management trials. These were established in 2011, 2013 and 2014 and now provide data to evaluate species health, adaptability and performance across a matrix of sites with a variety of management. This series of trials ensure there are future candidates for the NZDFI tree breeding programme should that be necessary.

New Zealand has a long history of testing *Eucalyptus* species. Trials established between 1991 and 2004 by NZ Forest Research focused predominantly on naturally durable species and the stringybark group (McKenzie, 1993 and McConnochie *et al*, 2008). The results from these plantings and the experience of Farm Forestry growers was used to define the species for the demonstration species and management trials.

There are 11 species in the 2011 trials including NZDFI's five breeding species and radiata pine as a control. These are *E. argophloia, E. bosistoana, E. cladocalyx, E. camaldulensis, E. eugenioides, E. globoidea, E. longifolia, E. macrorhyncha* , *E. notabilis, E. quadrangulata*, and *E. tricarpa*. The number of species was reduced to 8 species in the 2013 and 2014 trials with *E. argophloia, E. eugenoides* and *E. notablis* not included.
Strict selection criteria were applied to choosing these species:

- Class 1 & 2/3 durable species (Australian Standard)
- Tree form and good growth
- Very high MOE (Stiffness) and MOR (Strength)
- Drought resistance (< 1000mm/year)
- Frost tolerance
- Proven timber use
- Potential for breeding hybrids
- Vigorous coppice

These trials are testing broad-based seedlots, with numbers of parents shown in Table 2. They are strategically located across regions in NZ and in different sites with varying environmental conditions. Figure A1.

Permanent Sample Plots (PSP) are established at age 3 and measured regularly prior to first thinning. Further measurement of these trials will yield useful productivity and crop management data. Knowledge gained will be transferred to landowners and other growers through our regional networks; and workshops/field days; videos etc.

References

Apiolaza LA. 2009. Very early selection for wood quality: screening for early winners. Annals of Forest Science 66(6): 601

Apiolaza LA, 2009. Exploiting different scales of variability in a generic *Eucalyptus* species. In: Apiolaza LA, Chauhan S and Walker JCF. Revisiting Eucalypts Workshop, Christchurch, February, 83-89.

Apiolaza LA, Raymond CA and Yeo B. 2005. Genetic variation of physical and chemical wood properties of *Eucalyptus globulus*. Silvae Genetica 54: 160-166

Apiolaza LA, McConnochie R, Millen P, Van Ballekom S and Walker JCF. 2011. Introducing durable species to New Zealand drylands: genetics of early adaptation of *Eucalyptus bosistoana*. In: New Zealand Dryland Forests Initiative Workshop. Blenheim. 2nd November, 137-145.

Borralho NMG and Potts BM. 1996. Accounting for native stand characteristics in genetic evaluations of open-pollinated progeny from a *Eucalyptus globulus* base population. New Forests 11(1): 53–64.

Chauhan S and Entwistle K. 2010. Measurement of surface growth stress in *Eucalyptus nitens* Maiden by splitting a log along its axis. Holzforschung 64: 267-272.

Davies N, Apiolaza LA and Sharma M. 2016. Heritability of growth strain in *Eucalyptus bosistoana*: a Bayesian approach with left-censored data. Submitted.

Dutkowski GW and Potts BM. 1999. Geographic patterns of genetic variation in *Eucalyptus globulus* ssp. *globulus* and a revised racial classification. *Australian Journal of Botany* 47: 237–263.

Dutkowski GW, Potts BM, Williams D, Kube PD and McArthur C. 2001. Geographic genetic variation in central Victorian *Eucalyptus nitens*. In IUFRO Conference *Developing the Eucalypt of the Future.* Valdivia, Chile, September 2001.

Ericsson T and Fries A. 1999. High heritability for heartwood in north Swedish Scots pine. Theoretical and Applied Genetics 98: 732-735.

Evison DC and Apiolaza LA 2015. Incorporating economic weights into Radiata pine breeding selection decisions. Canadian Journal of Forest Research 45: 135-140.

FAO. 1981 Eucalypts for planting. Rome.

Grundy B, Caballero A, Santiago E and Hill WG. 1994. A note on using biased parameter values and non-random mating to reduce rates of inbreeding in selection programmes. Animal Science 59(3): 465-468.

Hardner CM, Vaillancourt RE and Potts BM. 1996. Stand density influences outcrossing rate and growth of ppen-pollinated families of *Eucalyptus globulus*. Silvae Genetica 45(4): 226-228.

Jones RC, Vaillancourt RE, Gore PL and Potts BM. 2011. Genetic control of flowering time in *Eucalyptus globulus* ssp. *globulus*. Tree Genetics & Genomes 7: 1209-1218.

Miranda I, Gominho J, Aráujo C and Pereira H. 2014. Family effects in heartwood content of *Eucalyptus globulus* L. European Journal of Forest Research 133: 81-87.

Patterson BA, Vaillancourt RE , Pilbeam DJ and Potts BM. 2004. Factors affecting variation in outcrossing rate in *Eucalyptus globulus*. Australian Journal of Botany 52(6) 773-780.

Potts BM and Dungey HS. 2004. Interspecific hybridization of *Eucalyptus*: key issues for breeders and geneticists. New Forests 27: 115-138.

White T. 2001. Breeding strategies for forest trees: concepts and challenges. The Southern African Forestry Journal 190: 31-42.

Appendices

Appendix 1. List of eucalypt species tested in Marlborough from 2003 to 2006

Symphomyrtus
Eucalyptus bosistoana
Eucalyptus camaldulensis
Eucalyptus cladocalyx
Eucalyptus maidenii
Eucalyptus melliodora
Eucalyptus microcarpa
Eucalyptus moluccana
Eucalyptus quadrangulata
Eucalyptus saligna
Eucalyptus tereticornis
Eucalyptus wandoo

Monocalypts
Eucalyptus agglomerata
Eucalyptus blaxandii
Eucalyptus cameronii
Eucalyptus eugenoides
Eucalyptus fastigata
Eucalyptus globoidea
Eucalyptus laevopinea
Eucalyptus longifolia
Eucalyptus macrorhyncha
Eucalyptus microcorys
Eucalyptus muelleriana
Eucalyptus obliqua
Eucalyptus pilularis
Eucalyptus youmanii

Figure 5: Location of breeding and demonstration NZDFI Trials.

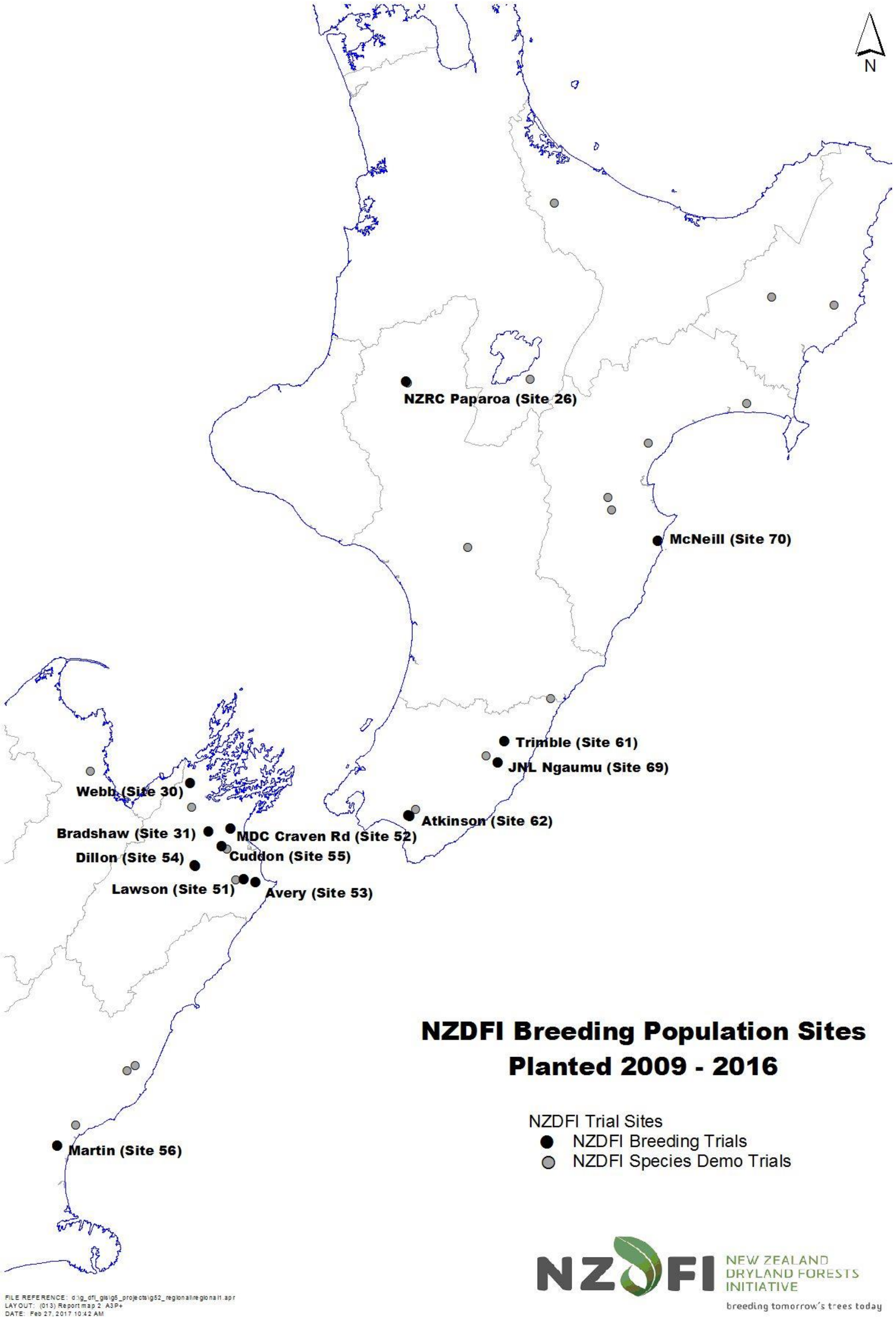


Figure 6: NZDFI tree improvement strategy

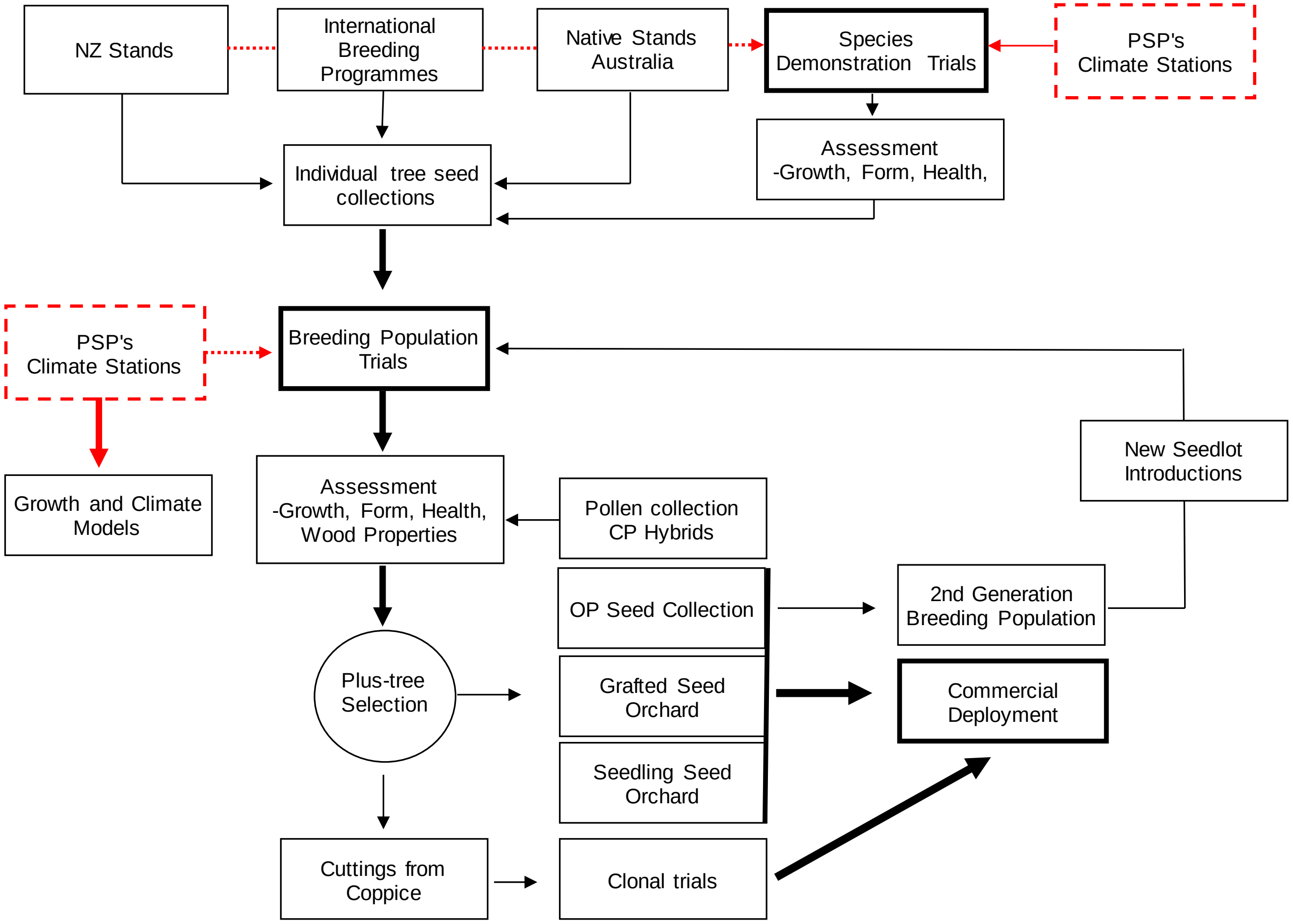


Table 3: Genetic material planted in NZDFI breeding trials

Est. Year	Site	E.bosistoana	E.globoidea	E.quadrangulata	E.argophloia	E.tricarpa
2009	Lawson	4500				
2009	Cravens	3750				
2009	Martin	3750				
2010	Avery	1750			270	
2010	Martin	2450				
2010	Cravens	1500				
2011	Atkinson		8640			
2011	Avery		10800			600
2011	Ngaumu		7200		1000	
2011	Martin			920		
2011	Cuddon			1680	1000	
2011	McNeil			1600		
2011	Trimble			1800		720
2011	Dillon				960	480
2012	Dillon	5040				
2012	McNeil	4320				
2012	Ngaumu	5040				
2016	Bradshaw			3600		
2016	Webb			3096		
2106	Paparoa			2880		
Total No. Seedlings Planted		32100	26640	15576	3230	1800
Total No. Families Tested		192	161	104	18	24

Table 4: Genetic material planted in NZDFI breeding trials (cont.)

Species	Trial Est. Year	No. Sites	No. Seedlings per Family	No. Families
E.bosistoana	2009	3	75	66
	2010	3	50-70	39
	2012	3	60-70	87
		Total No. families tested = 192		
E.globoidea	2011	3	80	161
		Total No. families tested = 161		
E.quadrangulata	2011	4	50-75	20
	2016	3	40-50	84
		Total No. families tested = 104		
E.tricarpa	2011	3	30	24
		Total No. families tested = 24		
E.argophloia	2010	1	15	15
	2011	3	50	18
		Total No. families tested = 33		

Table 5: List of species included in extension and management trials.

Species	Provenance	Parents
E. argophloia	*SSO Narromine*	42
E. bosistoana		50
E. camaldulensis	WAFPC SO	
E. cladocalyx	SSO Hamilton VIC	59
E. eugenioides	Sydney district	
E. globoidea	Cann River	10
2 seedlot mix	Yadboro SF	10
E. longifolia	Kerikeri	
E. macrorhyncha	Gunning NSW	4
3 seedlot mix	Stromlo Forest	5
	Uriarra Road	10
E. notabilis	Lake Burragorang	
E. quadrangulata	Mt Skanzi	
E. tricarpa	Tucker Box	11
2 seedlot mix	Martins Creek	11