**Environment and Agriculture**

# An Analytical Framework for the Domestication of Mallee: the Agronomy, Genetics, Productivity and Economics of Biomass Production in a Dryland Agricultural System

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To the best of my knowledge and belief this thesis contains no material previously published by any other person except where due acknowledgement has been made. This thesis contains no material which has been accepted for any other degree or diploma in any university.

Beren Spencer-Grayling

10th of February 2021

We acknowledge that Curtin University works across hundreds of traditional lands and custodial groups in Australia, and with First Nations people around the globe. We wish to pay our deepest respects to their ancestors and members of their communities, past, present, and to their emerging leaders. Our passion and commitment to work with all Australians and peoples from across the world, including our First Nations peoples are at the core of the work we do, reflective of our institutions' values and commitment to our role as leaders in the Reconciliation space in Australia.

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# List of publications

While a mallee-based biomass processing industry remains prospective, no large-scale commercial operations have yet emerged. The objective of the General Introduction, and the series of papers to follow, is to provide access to the knowledge now available on operational practice, design of integrated mallee systems, productivity, harvest and supply chain development, and economics of mallee crops in wheat/sheep agriculture in Western Australia, with a view to facilitating future industry development.

This thesis comprises of five published peer-reviewed journal articles (1 to 5 below). Additional to these are three peer-reviewed articles that are collaborations directly related to the thesis (6 to 8 below). The primary author of the three additional articles is Richard Mazanec.

1) Spencer B, Abadi A, Bartle J, Sudmeyer R, Van Gent S, Gibberd M, Zerihun A (2021). Determinants of the economic viability of mallee eucalypts as a short rotation coppice crop integrated into farming systems of Western Australia. GCB Bioenergy 13:242-256 https://doi.org/10.1111/gcbb.12775

2) Spencer B, Mazanec R, Gibberd M, Zerihun A (2021). Sapling and coppice biomass heritabilities and potential gains from *Eucalyptus polybractea* progeny trials. Tree Genetics & Genomes 17:15 https://doi.org/10.1007/s11295-021-01499-7

3) Spencer B, Mazanec R, Abadi A, Gibberd M, Zerihun A (2020). Flowering phenology in a *Eucalyptus loxophleba* seed orchard, heritability and genetic correlation with biomass production and cineole: breeding strategy implications. Scientific Reports 10:15303 https://doi:10.1038/s41598-020-72346-3

4) Spencer B, Bartle J, Abadi A, Gibberd M, Zerihun A (2021). Planting configuration affects productivity, tree form and survival of mallee eucalypts in agroforestry systems. Agroforestry Systems 95(1):71–84 https://doi:10.1007/s10457-020-00543-0

5) Spencer B, Bartle J, Huxtable D, Mazanec R, Abadi A, Gibberd M, Zerihun A (2019). A decadal multi-site study of the effects of frequency and season of harvest on biomass production from mallee eucalypts. Forest Ecology and Management 453:117576. https://doi.org/10.1016/j.foreco.2019.117576

6) Mazanec R, Grayling P, Doran J, Spencer B, Turnbull, P (2021) Genetic parameters and potential gains from breeding for biomass and cineole production in three-year-old Eucalyptuspolybractea progeny trials, Australian Forestry, 84:1, 13-24, https://doi.org/10.1080/00049158.2021.1892999

7) Mazanec R, Grayling P, Doran J, Spencer B, Neumann C (2020). Provenance variation, genetic parameters and potential gains from selection for biomass and cineole production in three-year-old *Eucalyptus loxophleba* subsp. *gratiae* progeny trials. Australian Forestry 83:75-90. https://doi.org/10.1080/00049158.2020.1753294

8) Mazanec R, Grayling P, Spencer B, Doran J, Neumann C (2017). Provenance variation, genetic parameters and potential gains from breeding for biomass and cineole production in three-year-old *Eucalyptus loxophleba* subsp. *lissophloia* progeny trials. Australian Forestry 80:34-42. https://doi.org/10.1080/00049158.2016.1275100

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# Abstract

The conversion of 16 million ha of native perennial vegetation to agriculture in the 300 to 600 mm winter rainfall region in the south west of Western Australia (WA) during the period 1850 - 1975 has caused extensive environmental change. One of the more serious changes has been hydrological, where groundwater has been able to accumulate and mobilise stored salt, generating land and stream salinity on a regional scale. Salinity has degraded some 10% of the region and has been projected to damage up to 30% over several decades as a new hydrologic balance is generated. To help control this threat, research to develop deep rooted perennial crops commenced. Mallee eucalypts emerged as a strong prospect due to an existing global market for Eucalyptus oil (in particular 1,8-cineole) and more recently, for their bioenergy, biochar and carbon sequestration potential.

Mallee species selection in the 1990s and 2000s, based initially on oil production and later on biomass production, narrowed the number of species to *Eucalyptus polybractea*, *E. loxophleba* (subsp. *gratea* and *lissophloia*) and *E. kochii* (subsp. *plenissima*, *kochii* and *borealis*). A breeding program was established by the WA State Government for seed production. Progeny trials were thinned to seed orchards with retained trees selected on the basis of foliar 1,8-cineole concentration, and later, on a combination of foliar 1,8-cineole and biomass productivity. These species were adopted as a tool to address the hydrological imbalances with the potential of regular harvest to diversify incomes for landholders. However, little was known about the yield potential, genetic parameters of traits of interest, frequency of harvest to maximise yield or the consequence of mallee integration on farm economics.

To assess the yield potential of mallee, 19 experimental sites were established on mature belts of mallee (5 to 12 years old) across the WA wheatbelt of the three targeted species. These were subjected to two harvest frequency treatments (3 to 8 year cycles based on growth rates) and two seasons of harvest (autumn and spring) treatments over a decade. Mortality was low at 16 sites ranging from 1% to 12% while the other three sites failed due to shallow soil profiles. Dry biomass yield on successful sites ranged between 2.2 to 32.8 dry Mg ha$^{-1}$ year$^{-1}$. There was a species response to season of harvest with *E. loxophleba* more productive following spring harvests while *E. kochii* yielded more when subjected to autumn harvests. Sites with higher productivity generally had acidic soil, low EC$_e$ and comparatively high fertility. At nine of the ten sites subjected to three 3-year harvests, there was no indication of declining yields demonstrating that at productive sites, 3-year rotations are appropriate for mallee biomass production.

To determine the optimal planting configurations for mallee biomass production, one *E. polybractea* and one *E. loxophleba* trial were established around Narrogin (WA). Each trial consisted of 20 spacing treatments: four row treatments (1, 2, 4 and 6 row belts); and five within-row spacing treatments (1, 1.5, 2, 3 and 4 m). Biomass yields ranged from 2.7 to 21.2 dry Mg $ha^{-1}$ $year^{-1}$ with significant rows and within-row spacing effects. Across both trials, productivity was highest in narrower belts and shorter within-row spacing. The large variation in productivity will determine the planting distribution and future profitability from mallee plantings.

In the *E. loxophleba* seed orchards, individuals were recorded flowering at all times of the year. The timing of key flowering traits determines the potential of gene exchange and self-fertilisation. To investigate this, monthly flowering observations were made at an *E. loxophleba* seed orchard comprising of 1142 trees, from 60 families and nine provenances. Foliar 1,8-cineole concentrations and biomass of each tree were also assessed. Heritability for start and end of flowering and 1,8-cineole were moderate to high ($\hat{h}^2$ = 0.75 to 0.45) while the propensity to flower, flowering duration, and biomass estimates were moderate to low ($\hat{h}^2$ = 0.31 to 0.10). Strong genetic correlations were found between most flowering traits; correlation between flowering traits and biomass or 1,8-cineole were weak. To hasten genetic improvement, selection for in-phase flowering, in addition to biomass productivity and cineole content, should be adopted.

The mallee breeding program initially applied selection for biomass on sapling biomass. For mallee, which are coppice crops, gain from selection on sapling vs coppice is unknown. In this work, we assessed if selection based on coppice biomass provides better gain than on sapling biomass. At three *E. polybractea* progeny trails, biomass assessments were made on saplings at the age of three and six years. The trials were then harvested and coppice biomass was assessed 3.5 years later. We found very low mortality (1-2%) at each trial. Sapling biomass heritability ranged from 0.06 to 0.36 and 0.07 to 0.17 for coppice. Within-site genetic correlations were strong between each assessment and cross-site genetic correlations were not different from unity. Simulation revealed that if the selection imposed on 3-year old saplings was applied to coppice, negative biomass gains would be achieved (-7.1% to 3.4%); whereas gains of 8.4% to 14.8% could be achieved if coppice selection was applied to the coppice assessment. Hence selection for coppice biomass productivity should be made on data generated from coppice growth data.

Experimental mallee biomass and crop yield data were used to assess the economic viability at 11 sites in the WA wheatbelt over the period 2006 to 2012. Levelised cost (LC) of biomass production ranged across sites from $40 to $257 fresh $Mg^{-1}$ and was most strongly correlated with mallee productivity. The loss of crop and pasture yield through mallee competition account for 38% of mallee costs, mallee harvesting was 32% of costs, opportunity cost of land was 16% and mallee planting establishment was 14% of costs. If income generated from carbon credits of below-ground biomass is included, LC dropped by 11%. LC from unharvested mallee ranged from \$33 to \$237 fresh $Mg^{-1}$. If above- and below-ground biomass are converted to carbon credits, three of the eleven sites were profitable.

In summary, the results from this thesis show that mallee can be harvested at productive sites on a 3-year rotation. Mallee should not be planted at sub-optimal sites (alkaline soil, high soil $EC_e$ with poor nutrients) due to high risk of mortality and high costs incurred to the landholder. Future mallee planting should be in narrow belts (1 or 2 rows) with short (1 to 2 m) within-row spacing. A well-managed breeding program can achieve gains in both 1,8-foliar cineole and biomass concurrently. Reproductive phenological observations are important to determine the outcrossing potential of individual trees and families in a breeding population. To achieve gains in coppice biomass, breeding programs should select on coppice biomass. Harvested mallee biomass needs to be sold at under \$60 fresh $Mg^{-1}$ at farm gate to foster profitable processing. Mallee planted in unfavourable conditions are unlikely to be economically viable except when the landholder attaches value to the secondary benefits.

# Chapter 1

## General Introduction

Mallee species are multi-stemmed lignotuberious eucalypts suitable for harvest and have been planted in the Western Australian (WA) wheatbelt for decades as a land management tool with additional economic benefits from the production of cineole and biomass, and more recently, for carbon sequestration. This thesis aims to fill key knowledge gaps remaining after two decades of research into mallee domestication and commercialisation carried out by the government and university sectors. This introduction provides the historical and land-use context of mallee development as a perennial crop. It also reviews the progress that has been made in both biomass production capability and mallee economics.

### *Early agricultural development in WA*

The region lying between latitudes 29° and 34° south and the annual rainfall isohyets 300 and 600 mm in the south west corner of WA was extensively developed for dryland agriculture in the late 1800s and 1900s. The area has a warm temperate Mediterranean type climate with an agriculture based on introduced annual, winter-growing crop and pasture species, and is locally known as the wheatbelt region (Turner & Asseng, 2005). Geologically the region consists of an ancient stable plate of granitic rocks called the Yilgarn Plateau. It has been eroded to a low elevation (300 m) and low relief landscape, with a mantle of highly weathered materials retained in deep (20-30 m), infertile soil profiles (Mulcahy, 1967). Conversion of the native woodlands and shrub-lands to agriculture did not commence on any scale until phosphate fertiliser became available in the early 1900s. Development accelerated in the 1950s following the introduction of trace element fertilisers, mechanisation, and the adoption of legume based annual pastures used in rotation with cereal crops (Connor, 2004; Nichols *et al.*, 2012; Kirkegaard *et al.*, 2014). There is now 16 million ha of land in agricultural production supporting competitive, export-oriented industries in grains, wool and sheep meat with export revenues of over $8 billion in 2016 (Carberry *et al.*, 2011; ABARE, 2016). New land development slowed to a halt in the 1980s with the introduction of land clearing controls due to the recognition of sustainability issues, including dryland salinity and soil loss, as threats to both conservation and agricultural production (Australian Greenhouse Office, 2000).

### *Salinity*

Secondary salinisation is a consequence of hydrological change following conversion of native perennial woody vegetation to introduced annual crop and pasture species (Clarke *et al.*, 2002; Hatton *et al.*, 2003; Peck & Hatton, 2003; George *et al.*, 2008). The shallow-rooted annuals have lower plant water use and this leads to extensive accumulation of the surplus water in the deeply weathered (20-30m) regolith formed over the basement igneous rocks. In the generally low relief landscape this creates permanently moist sub-soils, slowly expanding groundwater systems, mobilisation of previously stable sub-soil salt storage, and extensive slow discharge of saline groundwater in low landscape positions and drainage lines.

The subsoil salt is mostly of marine origin and arrives in rainfall and as dry fallout at 20-50 kg ha$^{-1}$ year$^{-1}$ (Hingston & Gailitis, 1976). Storage of total soluble salts over the full profile depth (average depth of regolith was 33.6 m) can be up to 20 Gg ha$^{-1}$ (McFarlane & George, 1992). Peck and Hurle (1973) showed that under native vegetation salt input and output in stream flow are approximately in balance, but the reduced water use under agriculture enhances the leaching of stored salt such that salt output far exceeds input.

Shallow saline water tables and saline seepage has degraded some 1.1 million ha of previously productive farmland or about 7% of the total 16 million ha of farmland in the wheat/sheep region. Estimates show that the potential total area under threat of salinity may be up to 4.5 million ha or 28% of the region (George *et al.*, 2005; Simons *et al.*, 2013). In addition to damage to productive land, the extensive shallow water tables on valley floors degrade water resources (Schofield & Ruprecht, 1989); compromise regional community and transport infrastructure (Sparks *et al.*, 2006); and exacerbate the fragmentation of remnant native habitat in a region recognised as one of the 25 global biodiversity hot-spots (Myers *et al.*, 2000; Hopper & Gioia, 2004). Natural resource management (NRM) is a generic term covering a range of activities, including those aiming to overcome these forms of degradation.

### *Salinity impact on water supply catchments*

During the 1970s increasing stream salinity in five catchments with water supply potential that had been subject to recent agricultural development, was recognised as a problem that required active intervention. These catchments are confined to higher rainfall areas west of the wheat/sheep region but have comparable hydrologic processes and salinity risk. The right to clear land in these catchments was withdrawn and land acquisition for revegetation was imposed (Sadler & Williams, 1981). This was supported by a period of new investment in salinity related research (Anon., 1981; Sharma, 1984; Peck & Williamson, 1987) which added momentum to other long-standing salinity research in the wheat/sheep region (Peck, 1978; Nulsen & Henschke, 1981).

Revegetation commenced on acquired farmland on the Collie River catchment in 1977. Some 10,000 ha of mostly lower slopes and valley floor land was planted in the period up to 1996 with some success (Mauger *et al.*, 2001). Revegetation was also undertaken in other water supply catchments (Bari *et al.*, 2004; Smith *et al.*, 2007). This was the first example of large-scale revegetation for salinity control in WA. It provided an opportunity to design, apply and assess the performance of a large range of revegetation species and methods. The results are reviewed by Schofield *et al.* (1989) and Mauger *et al.* (2001).

*Revegetation for profit*

The forced acquisition of land by the State government was viewed as an unwelcome imposition by some farmers. A major conclusion from this early experience was that while many tree species might be well adapted for revegetation of salt affected catchments, those with potential commercial value, and able to be integrated into local farming systems, should be preferred (Schofield & Ruprecht, 1989). This preference acknowledged that the up-front cost and long delayed benefits of salinity control make extensive non-commercial revegetation difficult to finance (State Salinity Council, 2000; Sparks *et al.*, 2006). Furthermore, the efficient revegetation and management of the land would be best left to farmers, and if it was profitable and compatible with their conventional farm business, tree crops might be readily adopted (Lefroy *et al.*, 1993; Bartle, 2009). It was recognised that a conflict was emerging; landholders were willing to revegetate, but not to the magnitude that was going to risk their income by conversion of agricultural land to trees (O'Sullivan, 2004). This restricted revegetation projects to certain soil types and landscape positions that were suboptimal for highly productive plantings.

The momentum behind the confidence in commercially-driven tree crops was partly due to the development of the forestry share-farming concept. Share-farming was first applied to *Pinus radiata* plantation forestry in WA by the State Department of Conservation and Land Management (CALM) in the mid-1980s. This stimulated development of a legal instrument called *profit á prendre* to enable the ownership of a forestry operation to be separated from the ownership of the land, including transferability to a new owner upon sale of the land (Heenan, 1987). In this way a processing industry with a payback period of a decade or more could sponsor development of a secure long-term resource without having to own the land. This form of legal agreement also proved amenable to a wide range of options in planting configuration (including agroforestry where tree crop and agriculture are integrated for mutual benefit), and for cost and revenue sharing between the owners of the land and the timber crop.

This in turn facilitated the emergence of bluegum (*Eucalyptus globulus*) forestry in the Albany region in the late 1980s. Bluegum had potential for large scale export of wood chip for paper manufacture (Shea & Bartle, 1988; Bartle & Shea, 1989). To enhance the attractiveness of bluegum to farmers, widespread demonstration of agroforestry options were undertaken (Bartle, 1989; Bartle, 1991). However, the wider economic and policy settings proved even more attractive to agribusiness ‘managed investment schemes’ (MIS). During the early 1990s these schemes proliferated rapidly, undertaking large scale land purchase and whole-farm plantation establishment that increased the price of suitable farmland and overwhelmed the interest in agroforestry. While MIS activity was effective in delivering some public benefits, such as improved stream water quality in the Denmark River catchment (Bari *et al.*, 2004), the community was divided into those who liked trees or profited from selling for forestry, and those that resented trees displacing conventional agriculture (Schirmer & Tonts, 2003; Schirmer, 2007; Williams, 2014). Furthermore, the MIS business model proved to be flawed and by 2009 two major companies (Great Southern and Timbercorp) had become insolvent (Brown *et al.*, 2010). In spite of problems with MIS development, a viable bluegum industry was established utilising other business models which were less tax driven. All of the commercial development outlined in this section occurred outside the wheatbelt. Given the sustainability issues in the wheatbelt, and that perennial revegetation could contribute to their resolution, the question arose as to whether the experience in the higher-rainfall agricultural zone could be extended to the wheatbelt.

### *Woody crops for the wheat/sheep region*

In 1992 CALM sought to build on the positive aspects of the bluegum experience and develop some form of agroforestry or tree crop industry that would be attractive to farmers in the wheat/sheep region (Bartle & Shea, 2002). An assessment of the options showed that mallee eucalypts with leaf oil and bioenergy production potential held promise for large scale development (Bartle, 1999; Barton, 2000; Bartle, 2001; Bartle & Shea, 2002). Building on earlier trials by Murdoch University, the participation of several farmer groups across the wheat/sheep region was enlisted by CALM. Two years of species and silvicultural trials, mainly in the form of demonstration plots, were followed by the commencement of extensive planting and research in 1994 (Department of Agriculture, 1989). Much of this work was never published, but (Eastham *et al.*, 1993) provides an example of a multi-species experiment used to demonstrated biomass production, survival and cineole yield of 12 species following multiple harvests. This experiment was later used to report on water use for salinity mitigation (Eastham *et al.*, 1994).

There was a further period of intensification of salinity research during the 1990s, coincident with the Decade of Landcare and supported by Commonwealth Government sponsored programs, such as the National Landcare Program, the Natural Heritage Trust and the National Dryland Salinity Program (Alexander *et al.*, 2000; Cary & Webb, 2000; Pannell, 2001a). This was a period of intense activity instigated by questions concerning the sustainability of agriculture in Australia and its environmental impacts in general, including loss of biodiversity. This period saw the development of national and state policy, which included some planning concerning revegetation, including tree planting. In WA, a salinity action plan and strategy were developed (Anon., 1996b, 1996a; State Salinity Council, 2000; Wallace, 2001). The period generated debate and controversy, including comments by some who criticised a ‘spirit of forced optimism’, lack of rigorous analysis, and weak development of the necessary technologies (Pannell, 2001b). For example, salinity planning in WA generated considerable argument as to the role of revegetation. Hydrologists analysed new data and argued that the proportion of revegetation required to control recharge in some catchments may be as much as 80%, in contrast to the then common assumption that 10-20% might be adequate (George *et al.*, 1999). This challenged the recommendations for agroforestry with belt plantings of potentially commercial species like mallee. Bartle (1999) responded that, with careful design to intercept surface run-off and shallow sub-surface interflow, the effectiveness of a small area of narrow belt plantings could be amplified; but perennials on recharge areas should only ever be seen as a complement to other measures such as discharge zone treatments (including perennial pastures and drainage) to achieve a satisfactory level of salinity control. The concept of designing agriculture to be a structural and functional mimic of natural ecosystems was the subject of an international meeting that provided useful insights (Lefroy & Stirzaker, 1999). This ecological perspective of how to better manage the recharge proportion of the landscape was complemented by a later international meeting that focused on managing discharge areas using engineering methods such as drainage and groundwater pumping (Dogramaci & Degens, 2003; Dogramaci, 2004).

No consensus was achieved among researchers on the role of revegetation with perennial species in salinity management. However, there was a general view that such plantings could offer advantage in specific situations, especially if a broader set of outcomes – such as water production, protection of rural towns, and conservation of biodiversity – were taken into consideration. Nevertheless, these debates and related developments provided the impetus for the foundation of two Co-operative Research Centres – the CRC for Plant-based Management of Dryland Salinity (Salinity CRC), and subsequently the Future Farm Industries CRC (FFI CRC). Here, sustained and coordinated research and development of herbaceous and woody perennials technology was undertaken over the period 2000-2014 and in WA, some of this centred on oil mallee.

### *Climate change*

During the 1990s, climate change emerged as a global and national issue. The United Nations took the lead in trying to achieve coherent global action to better understand the science of climate; devise remedies and rules for management of anthropogenic greenhouse gas emissions; and enlist commitment from all nations. An international treaty called the United Nations Framework Convention on Climate Change (UNFCCC) was signed by 154 states and the European Community in 1992, which, however, was criticised due to a lack of financial or market mechanisms to reduce emissions of greenhouse gases (Bodansky, 1993). The Kyoto Protocol, which extended the UNFCCC, was signed by Australia in 1997 and established legally binding obligations for most industrialised nations to limit their net emissions of greenhouse gases to an extent designed to keep atmospheric heating within an acceptable range (Kyoto Protocol, 1998). Kyoto was not ratified by Australia until 2007. However, this stimulated a period of discussion and innovation in Australia on all forms of mitigation of climate change, the most relevant in the agricultural context being: restricting new land clearing; sequestering carbon dioxide by revegetation of farmland; and the use of crop residues and new biomass crops for renewable energy (Kyoto Protocol, 1998; Harper *et al.*, 2007).

In his national review, Garnaut (2008) described climate change as a 'diabolical policy problem'. This difficulty is reflected in the national and international weakening of the resolve to mitigate climate change and slippage to support the UN coordinated action. However, more recently there has been a strengthened resolve from a number of cities, regions, countries and companies including the declaration of zero-net-emissions by 2050 targets (NewClimate Institute, 2020; van Soest *et al.*, 2021). Agriculture has the opportunity to add climate change to the forces driving profitable new regional biomass industries (Stucley *et al.*, 2012; Climate Change Authority, 2019).

In 2015, the Paris Climate Agreement, further extended the UNFCCC, was ratified by Australia and mandates emission reductions targets (Paris Agreement, 2015). To achieve these, Australia legislated the Australian Carbon Credits (Carbon Farming Initiative) Act 2011 (Department of the Environment, 2015b), as a mechanism for generating carbon credits from the reforestation of land.  This legislation implements the creation of Australian Carbon Credit Units (ACCU), a unit tonne of carbon dioxide equivalent, or $CO_2e$, sequestered by specific projects under approved methodology. The number of ACCUs generated from a revegetation project can be assessed either using the FullCAM carbon accounting model (Paul *et al.*, 2013) or direct biomass measurement methods (Department of the Environment, 2014).

There are four methods under which ACCUs can be generated from new revegetation projects: Reforestation and Afforestation (2015); Reforestation by Environmental or Mallee Plantings – FullCAM (2014); Measurement based methods for new farm forestry plantations (2014); and Plantation forestry (2017). These methods differ with limits on the spatial extent where planting are allowed and restrict certain management options. For instance, the Reforestation by Environmental or Mallee Plantings – FullCAM, and the Reforestation and Afforestation methods do not allow for plantings to be harvested, making these methods unsuitable for mallee biomass supply; the Plantation Forestry method is spatially restricted to current National Plantation Inventory regions which excludes much or the northern and eastern wheatbelt of WA. Thus, for the purposes of this thesis, the Measurement based methods for new farm forestry plantations methodology is used hereafter. This methodology is suitable for farm forestry plantings, including mallee, over a minimum timeframe of 25 years, although there is a reduction of 20% of the $CO_2e$ net abatement if harvested before 100 years (Department of the Environment, 2015a). Currently, ninety five percent of ACCUs are purchased by the government through the Emissions

Reductions Fund (ERF) but these can be on-sold to commercial ventures (Climate Change Authority, 2020). The four methodologies as outlined above only produced 1.2% of the total ACCUs generated under the ERF due to the large up-front costs of revegetation (Roxburgh *et al.*, 2020).

## Screening process for commercialisation of perennial species

Many perennial species adapted to the wheat/sheep region environment have the potential to contribute to salinity control by increasing the annual duration of active growth and the depth of root penetration and thus access to soil water stores. Overall, this contributes to increasing plant water use, including from soil water that is not accessible to annual plants. While mallee was an early standout group for development, there are many other species and many product prospects. Bartle and Reeves (1992) proposed a strategy for systematic assessment of potentially suitable species and products. They suggested parallel assessment pathways for species and products with progressively more intensive assessment of those showing the most promise. This strategy was applied by Olsen *et al.* (2004) to the woody native flora of south west of WA, and subsequently by Hobbs *et al.* (2009) to the flora of south-eastern Australia. There are several generic conditions that these studies adopted to sharpen the focus on the most prospective species and products. Importantly, this work was underpinned by perennial revegetation used as a focus on commercial production. Other drivers included revegetation to better conserve the native biota, and to protect water and rural infrastructure assets. Many criteria were used to assess woody species for commercial production; below are some of the main factors that highlight mallee as a strong candidate for widespread adoption. These pertain to ecological, physiological and industrial applications.

### *Preference for native woody species*

The work by Olsen *et al.* (2004) and Hobbs *et al.* (2009) took the position that new woody crop development should focus on domestication of natives rather than introduction of alien species. The native flora is adapted to local conditions, presents a low risk of generating weed problems and provides great diversity from which to choose. Biosecurity became a global issue during the 1990s and a National Weed Strategy was developed (Anon., 1999). The focus on regional native woody species avoided the identified weed risks (Pheloung *et al.*, 1999; Virtue *et al.*, 2004; Wallace, 2006; Stone *et al.*, 2008).

### *The imperative of scale*

To achieve the sustainable land use goals and the economies of scale necessary for efficient farm and processor operations, it was essential to obtain broad-scale plantings of woody species. In this context, it was predicted that commercially viable woody crops used in the most effective designs could occupy more than a million ha and produce an annual production tonnage comparable to the wheat crop (Bartle, 1999, 2001). However, to achieve the necessary scales of plantings requires products with large volume markets in order to avoid oversupply constraining product options (see Table 1).

### *Harvest frequency*

An important issue in replacing annual crops and pastures with woody perennials is that while annual plants provide annual cash flows, harvested woody plants provide much more episodic cash flows. Woody crops amenable to frequent harvest will provide early and more regular cash flow, and moderate any competition imposed by belt planting on the adjacent annual crops. The early revenue return would also help the farmer finance the cost of establishment. But frequent harvest pushes selection towards coppicing species (i.e. able to re-sprout after harvest and avoid re-establishment costs). This discounts the potential of conventional forestry and sawn timber product options without annuity payments.

### *Whole plant harvest and whole biomass utilisation*

Forestry and grain cropping operations evolved with a focus on their major product. Hence harvest equipment and whole supply chains were developed to capture and deliver their single product as efficiently as possible. Residues had little or no value and were left in the forest or field. Globally improved residue collection capability is now being built into supply chains to capture a proportion of these residues for their emerging product value (Richard, 2010; Stucley *et al.*, 2012; Wolfsmayr & Rauch, 2014) and a new tool has been developed which maps biomass residues across Australia (ARENA, 2021a). New industries using coppicing species on a short harvest cycle will be best suited to whole biomass (a mixture of wood, leaf, twig and bark) harvest and supply chain operations. Likewise processing operations and products able to utilise whole biomass would avoid the cost of segregation of biomass components. Such total harvest operations must be careful to compensate or account for the removal of nutrients and energy from the system (Grove *et al.*, 2007; Yu *et al.*, 2015).

### *Product transport horizon*

The competitiveness of biomass production systems is dependent on transport supply chains. Annual crops have product values in the range \$200 to \$400 $Mg^{-1}$ (ABARE, 2020). Transportation of grain to a coastal port accounts for less than 15% of the value of the crop. In contrast, a bulk biomass product will be limited to a short transport horizon that will generally exclude export of raw product (Yu *et al.*, 2009). This constrains product options to those for which at least a preliminary value-adding step can be undertaken locally, and this was a selection criterion for the products in Table 1.

Table 1 Potential large-scale product types that might be derived from woody biomass. This table is a modified version from Section 3 in Olsen *et al.* (2004)

| Product category | Product group | Examples of particular products |
|---|---|---|
| Reconstituted wood products | Panel Board | Particleboard<br>Medium density fibreboard<br>Oriented strand board<br>Wood/cement/plastic composites |
| | Processed wood | Pulp and paper<br>Charcoal<br>Activated carbon |
| Bioenergy | Solid fuel | Electricity<br>Industrial steam and heat<br>Wood pellets<br>Desalinated water |
| | Biofuel | Cellulosic ethanol<br>Bio-oil by pyrolysis<br>Fischer-Tropsch liquids |
| Industrial products | Chemicals by transformation | Chemicals from bio-oil refinery<br>Bio-plastics, biomaterials |
| | Extracted chemicals | Oils, solvents, tannins, gums and resins |
| Fodder | Direct gazing | Drought fodder reserve |
| | Processed animal feed | Feed pellets |

*Other prospective species for domestication*

Table 2 shows the species selection steps used in the Search Project sponsored by the Salinity Co-operative Research Centre to screen for potentially economic species in the south west WA native flora (Olsen *et al.*, 2004). Expert knowledge, existing herbarium collections and botanical databases were used to narrow the selection of species by applying the simple screening steps outlined in Table 2.

Table 2: Progressive selection of species for domestication potential in the wheat/sheep region. This table is a modified version from Section 5 in Olsen *et al.* (2004) and Bartle (2009)

| | | |
|---|---|---|
| Total number of native species in Western Australia (WA). | | 9977 |
| Eliminate species listed as rare (likely to lack adaptability for use as crops) | | 7965 |
| Eliminate monocotyledons (to select for woody species) | | 6339 |
| Species that occur in at least one of the 4 botanical regions of the wheat/sheep zone | | 3664 |
| Species taller than 4m (available height data used as a surrogate for growth potential) | | 484 |
| Species taller than 4m distributed across the four wheat/sheep botanical zones | Avon wheatbelt | 309 |
| | Esperance sandplain | 266 |
| | Geraldton sandplain | 219 |
| | Mallee | 293 |
| Species >4 m tall occurring in all 4 botanical regions (adapted to a wide range of sites) | | 68 |

This process arrived at a manageable number of species which then made it feasible to progressively undertake more expensive product testing to arrive at a short-list of best prospects. The short-list included a selection of mallee eucalypts, some *Acacia* species (Maslin & McDonald, 2004; Bartle, 2006) and the salt tolerant fodder shrub, *Atriplex nummularia* (Barrett-Lennard *et al.*, 2005).

While this dual species/product strategy was necessary for new woody crop development, herbaceous perennials can also contribute to salinity control but have the substantial advantage of supplying feed to existing livestock industries and acceptable to farmers resistant to tree crops. Furthermore, there is a track record of herbaceous perennial use and potential to develop new species to expand the geographic range over which they might be used (Dear & Ewing, 2008; Dear *et al.*, 2008). Categories of herbaceous perennials subject to further development include legumes (Cocks, 2001), grasses (Sanford *et al.*, 2005; Moore *et al.*, 2014; Reed, 2014) and forage species (Barrett-Lennard *et al.*, 2005; Masters *et al.*, 2007; Hughes *et al.*, 2008). Perennial forms of the conventional cereal crops have also been proposed (Bell *et al.*, 2010).

There is a wide range of halophytic perennials that have good salinity and waterlogging tolerance and potential for productive use on areas with shallow saline water tables or active saline discharge (Barrett-Lennard *et al.*, 2005; Rogers *et al.*, 2005; Masters *et al.*, 2007). These include the saltbush group of species where new selection and breeding programs have generated improved varieties of *Atriplex nummularia* and extensive adoption of saltland pastures (Hobbs *et al.*, 2009). Although productivity and water use on salt-affected sites may be relatively low compared to upslope land, Bennett *et al.* (2014) show useful saline groundwater level recession and reduced salt and nutrient discharge.

### *Two decades of mallee R&D*

This brief historical introduction provides a context for the following review of the two decades of research and development of a mallee-based industry in WA over the period 1992 to 2015. The main objective of mallee development is to make it a competitive farm crop that is able to justify its integration into crop or pasture land on the basis of its economic performance alone. Success in this regard would open the way to efficient integration of tree crops with annual plant agriculture, and to achieve whatever salinity control and the other improvements in farm performance they might deliver. Furthermore, with a profitable woody crop option, farmers may find it more attractive to implement additional treatments for improving salinity management and farm sustainability.

This concept and its development over two decades attracted considerable support from research sponsoring bodies including Commonwealth and WA government departments, universities (UWA, Murdoch and Curtin), two Co-operative Research Centres (Salinity CRC and Future Farm Industries CRC) and many hundreds of wheatbelt farmers. The mallee development lost momentum after the close of the Future Farm Industries CRC in 2015, and the emergence of a period of uncertain national leadership on climate change policy, regulation of carbon emissions, and renewable energy that still prevails.

While a mallee-based biomass processing industry remains prospective, only a few small commercial operations have emerged. The objective of this overview is to provide a background to the knowledge now available on operational practice, design of integrated mallee systems, productivity, harvest and supply chain development, and economics of mallee crops in wheat/sheep agriculture in WA, with a view to facilitating future industry development.

## Why mallee?

### *Mallee growth habit and taxonomy*

Mallee are a group of species within the genus *Eucalyptus* characterised by multiple stems of low height (3 to 18 m) that arise from a prominent lignotuber sitting just below the ground surface that also forms the apex of the root system. The lignotuber is well provided with meristematic tissue and readily re-sprouts or coppices after damage or removal of the canopy by fire, drought or harvest (Noble, 1982; Noble, 2001; Wildy & Pate, 2002; Burrows, 2013)

The *Eucalyptus* genus has some 900 species classified by Brooker (2000) into several major and several minor sub-genera. Brooker included *Angophora* and *Corymbia* within the genus but a now substantial body of work using molecular genetics supports their exclusion (Ladiges & Udovicic, 2000; Steane *et al.*, 2002), thus reducing the eucalypts to about 800 species. About one-third of these can be described as mallee, but a sharp definition of mallee numbers is problematic given that expression of the mallee habit can be facultative and transitory (Nicolle, 2006). Of the more than 300 eucalypts species that occur in WA, about two-thirds are mallee (Hopper, 1990). Mallee species occur in all the polytypic sub-genera of *Eucalyptus* suggesting that the mallee habit is not a primary evolutionary branch within the genus and that mallee species therefore embrace considerable genetic diversity (Hill, 1989; Parsons, 1994).

### *Water use*

Mallee are a dominant component of the flora across southern Australia (latitude 25-36 degrees south and longitude 117-147 degrees east) within climates characterised by cool winters with 250-600 mm annual rainfall (winter dominant in the west to uniform annual distribution in the east) and hot summers with annual potential evaporation of 1500-2800mm (Parsons, 1994). The wetter part of this climate range includes the areas now developed for dryland wheat/sheep agriculture. Mallee also occur in a wide range of landforms ranging from late Quaternary sandy dune systems in the east to ancient, highly-weathered, deep profiles formed over granitic rocks on the Yilgarn Plateau in the west (Wasson, 1989).

The 3 to 10 fold excess of potential evaporation over rainfall indicates that water is a major constraint in biological productivity across this region. Mallee species are well adapted to this constraint. In a classification of hydraulic functional types, Mitchell *et al.* (2008) put mallee into the ‘year-round active water use’ category. Some adaptations to year-round water use were investigated by Nulsen *et al.* (1986). In a native stand of *E. pileata* and *E. eremophila* near

Newdegate in WA, they showed mallee to be deep rooted (roots were observed at 28 m); to actively channel stem flow along deeply penetrating roots; and to occupy deep more permeable soil types sited downslope of less permeable soils carrying low shrub cover that generated local run-off during major rainfall events. Verboom and Pate present the concept of bio-engineering, where plant and soil interact to modify the profile and enhance survival in semi-arid environments (Verboom & Pate, 2006; Verboom & Pate, 2013).

These contemporary observations help explain the ancient practice of the aboriginal people of excavating the long lateral roots of mallee (that also have occasional vertical 'sinker' roots (Wildy & Pate, 2002) to extract drinking water. Noble and Kimber (1997) list widespread historical accounts of this practice and also document that the word 'mallee' was derived from the name of the trees tapped for water by aboriginal people in the Lower Murray River region who were prominent in this practice.

### *The case for mallee domestication*

Based on the previous comments, the primary reason for selection of mallee as a candidate for domestication is its impressive pedigree. There are numerous mallee species native to the wheat/sheep and adjacent pastoral regions. These provide a large pool of genetic diversity from which can be drawn species with attributes that make them attractive prospects for 'short-rotation coppice' crops. Mallee provide the potential for development of large scale woody crops to be a semi-arid, warm-temperate climate analogue for the cool temperate woody biomass crops, willow and poplar, extensively used in the northern hemisphere (Dickmann, 2006; El Kasmioui & Ceulemans, 2012).

The second reason for selection of mallee is the long history and accumulation of knowledge in the use of native stands for the production of eucalyptus oil in Australia (Shiel, 1985; Boland *et al.*, 1991; Coppen, 2002a). In particular, *E. polybractea,* which occurs in two disjunct populations in West Wyalong in NSW and Bendigo in Victoria, has been harvested for eucalyptus oil production for about 100 years (Davis, 2002). Over the past couple of decades, the two major producers of eucalyptus oil have also established plantations on farmland to increase production, further building confidence that mallee can make the transition to being a domesticated crop plant.

The final reason for the selection of mallee was the work done on eucalyptus oil in WA during the 1980s. Associate Professor Allan Barton, who resided at Murdoch University, identified cineole, a major component of eucalyptus oil, as an attractive feedstock to produce large volumes of useful industrial chemicals (Barton, 2000). Of particular focus were to use cineole as a degreasing agent (Barton & Knight, 1997) and as a co-solvent in water/ethanol-fuel mixtures (Barton & Tjandra, 1988; Barton & Tjandra, 1989). Barton also assisted in the development of a rapid and accurate methodology to quantify the cineole concentration in eucalypts leaves (Ammon *et al.*, 1985a, 1985b). This facilitated the screening of many mallee to identify species and individuals with high foliar cineole concentration. Barton's work inspired others to explore the variation in foliar cineole from native populations which led to the exploration of the genetics and taxonomy from some high performing candidate species (Brooker *et al.*, 1988; Barton *et al.*, 1991; Grayling, 1996). Barton also established species comparison trials at several wheat/sheep region locations during the period 1985-88 and participated in the first experiments on planted mallee establishment and productivity (Eastham *et al.*, 1993). The WA Department of Agriculture became interested in using mallee eucalypts to assist to mitigating land degradation issues and to produce commercial eucalypt oil (Department of Agriculture, 1989).

Global Eucalyptus oil production has been volatile over the last few decades. During the 1990s the global Eucalyptus oil market volume increased from approximately 3,000 to 7,000 tonnes and much of this increase came from China, where Eucalyptus oil was a by-product of *E. globulus* timber production (Coppen, 2002b; Rajkumar, 2004; Pain, 2007). This oil has lower 1,8-cineole concentration (65%) and attracts a lower price than the high cineole content (70 to 90%) of the mallee species selected for cultivation in WA. There was a four-fold increase in Eucalyptus oil prices during the 2010-2020 period (Butlin pers comm). Given the motivation to utilise all potentially useful components of mallee biomass to stimulate industry development, it was deemed as important to maximise both mallee cineole content and biomass yield.

## Planning and progress in industry development

Creating new crops with associated regional industry is inevitably a long-term undertaking. An overview of the major streams of development that have been undertaken for mallee is presented in Fig 1.

Developing any new industry is inherently risky, especially in the early stages, and this discourages interest from commercial investors. The State Government Department Conservation and Land Management (CALM) provided capital and entrepreneurial input to initiate the bluegum industry in the higher rainfall regions in the late 1980s and early 1990s (Shea & Bartle, 1988; Bartle & Shea, 1989). CALM was keen to further apply the knowledge gained from this experience, especially given the greater potential for NRM benefits in the wheat/sheep region. They undertook long-term investment in mallee industry development starting in 1993 (Bartle & Shea, 2002). As the most prominent investor, CALM took a leading role in coordinating inputs from other parties including those associated with NRM activities, researchers, R&D funding agencies, regional economic development interests and potential farmer-growers. In this role CALM (and its successor departments) provided resources and coordination for more than two decades supported by annual budgets of about $0.75 million.

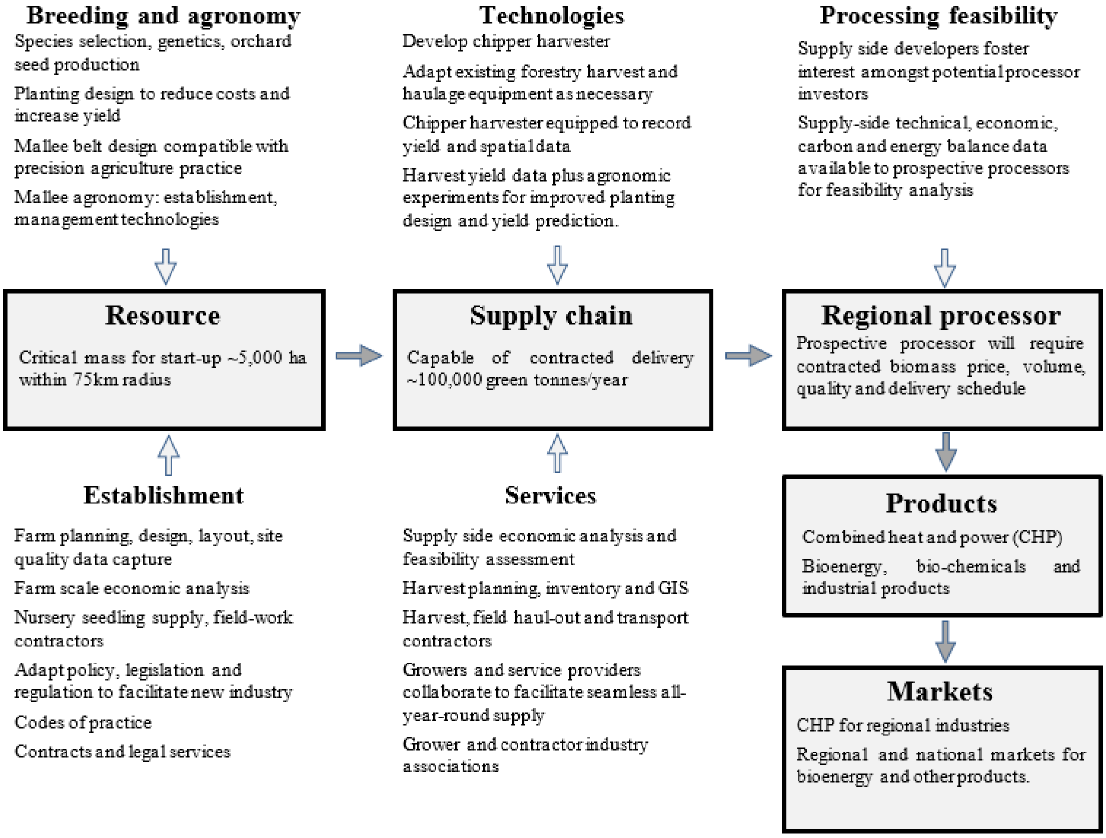

Figure 1: Conceptual overview of the major streams of development required to create a large-scale mallee biomass based industry (John Bartle *pers comm*)

An initial objective was to develop a sufficient mallee resource to demonstrate the potential to supply a large-scale biomass processing industry. This required new technology and service infrastructure to plant and manage an initial resource (Fig. 1). Six locations representative of the full range of site conditions that a large mallee industry would encounter were selected from across the wheat/sheep region. At each location a local farmers' land-care group was approached to support participation and provide local leadership. The aim was to confine planting within a 50-75km radius at each location with a view to developing critical mass for harvest efficiency and efficient future utilisation of the resource. Government funds were readily available to help revegetate to improve NRM. However, some farmers who were less engaged in the land care benefit of mallee were aware that revegetation for tree cover alone, even with a subsidy, would still cost more than the on-farm benefit they could expect. This was reflected in strong support for the mallee concept, the ready availability of farmers wanting to make exploratory plantings of mallee, and the emergence of the slogan 'profitable landcare' (OMA, 1999).

The local mallee groups were active, innovative and collegial. They combined to form the Oil Mallee Association (OMA) in 1996 and became active participants in industry development.

They were prominent in developing and pooling practical knowledge on mallee establishment and management. Growers were impressed with the vigour of mallee and its tolerance of sheep grazing which meant expensive fencing was not required. By 2008 some 12,500 ha of mallee had been planted with about 20% of all wheat/sheep region farmers undertaking exploratory planting, most commonly in belt form (URS, 2008). Grower experience was consolidated into an establishment manual (O'Sullivan, 1998), a code of practice (OMA, 2003, 2012) and an industry development plan (URS, 2008).

Grower interest extended to developing supply chain technologies. Growers became aware that an Australian Austoft 7700 cane harvester had been adapted and tested in Sweden for willow coppice harvest (van der Meijden & Gigler, 1995). They sponsored the purchase of a second-hand machine from Queensland in 1997. It was found that this machine could not be effectively adapted to cut the dense mallee stems but this exercise initiated more substantial investment in supply chain development (Giles, 1999; Bartle, 2001; Giles & Harris, 2003). Eucalyptus oil was always seen as a possible early source of revenue from mallee (Bartle & Reeves, 1992). Growers from the group located at Kalannie (30.367$^{o}$S 117.121$^{o}$E) attempted to develop small scale oil extraction. They tested a variety of harvest machines, but even on short harvest cycles with smaller stem diameters, they had problems with poor machine durability. After several years of perseverance, a successful small viable operation emerged and continues to operate successfully (Ian Stanley *pers comm*).

In 2000 OMA sponsored the establishment of a company (Oil Mallee Company, OMC) to provide a vehicle for commercial development. The OMC attempted to raise capital to support research and development but this was not successful. OMC was later sold to a carbon sequestration company.

In the late 1990s the Kyoto Protocol attracted strong commercial interest in carbon sequestration and renewable energy (Kyoto Protocol, 1998). In Australia, under the National Greenhouse Strategy a national Mandated Renewable Energy Target (MRET) was legislated in 2001 where generators were obliged to deliver 2% of their output from renewable sources, or purchase the equivalent on an open market. This was later increased to 20% (Crowley, 2013). OMA engaged with several major corporations to explore the potential of mallee both as a form of carbon sequestration and renewable energy. This delivered several years of valuable collaborative work on mallee research as well as promise of processor investment. The work with the WA State electricity generator, Verve, proceeded to the stage of construction of a demonstration processing facility. This was established at Narrogin and designed to test conversion of mallee biomass to activated carbon, electricity and eucalyptus oil (Enecon, 2001). Operational testing was not completed until 2006, but three years passed before it became clear that the process would not be commercially viable (Bartle & Abadi, 2010). This was a dismal outcome for growers who had been hopeful that their substantial commitment to biomass production would attract an early bioenergy start-up processor. This adversity was compounded by the growers’ dependence on regular mallee harvest to moderate the effect of competition from mallee belts on adjacent crop or pasture (Sudmeyer *et al.*, 2012).

By this time it was clear that wind and solar would capture most of the demand for renewable electricity generation (Lu *et al.*, 2017) and it is anticipated that batteries will be used to stabilise the grid (Datta *et al.*, 2018; Faunce *et al.*, 2018) However, there remains a potential biomass market amongst existing small regional enterprises such as abattoirs and stock feed manufactures who had experienced recent steep price increases for electricity and who were looking to reduce their costs for both heat and power (Colley, 2012; Colley *et al.*, 2020). With a view to support these developments, small scale supply chain options using available equipment were undertaken (Huxtable & Giles, 2014; Spinelli *et al.*, 2014).

The major product outlook for mallee has now turned to biofuels and biochar. The technologies for these are rapidly developing but there are still impediments to large-scale investments (Englund *et al.*, 2020). Recently however, a number of small- to medium-sized enterprises have moved into production (GHD, 2019). A small-scale plant converting municipal solid and forestry waste has recently been announced in the Shire of Collie, Western Australia, which aims to produce bioenergy, biofuels and biochar (ARENA, 2021b).

## Growing the resource

### *Mallee genetics and agronomy*

A breeding program was established by CALM in 1993 with the initial aims of producing out-crossed seed orchards with improved foliar cineole concentrations and to provide information on genetic parameters for key traits (Bartle *et al.*, 1998). It became apparent, that the progeny trials were not able to generate accurate estimation of genetic correlation due to the insufficient number of families (R. Mazanec *pers. comm.*). For each species, wild trees with available seed were screened for foliar cineole, using gas chromatography, and by 2002, some 15,000 wild individuals had been sampled (Bartle & Brennan, 2002). Initially, a threshold lower limit of foliar cineole of 2.7% was applied to the selection process (Bartle *et al.*, 1998), however, that was later reduced to 2.5% (Mazanec *et al.*, 2020; Mazanec *et al.*, 2021). For the candidate trees, the seed was harvested and used to establish progeny trials for the breeding program (Table 3) predicated on the basis that foliar cineole concentration was a heritable trait (Barton *et al.*, 1991). Progeny trials were established, saplings screened for foliar cineole concentration and a crude measure of biomass was undertaken (dead, runt, healthy, sub-dominant, dominant). Based on a combination of these two factors, but heavily weighted toward cineole concentration, the progeny trials were thinned to produce a seed orchard. However, it was clear that current cineole markets alone were not large enough to support a large scale industry and new markets were required for mallee biomass (Bartle *et al.*, 1996; Enecon, 2001; Wu *et al.*, 2008; Bartle & Abadi, 2010).

Table 3 The species included in the mallee breeding program, the wild parent trees that comprised the breeding population and the number of trials established in a range of years.

| Species | Number of parent trees | Number of established trials | Planting years |
|---|---|---|---|
| *E. loxophleba* subsp. *lissophloia* | 100 | 11 | 1995 - 1999 |
| *E. kochii* subsp. *borealis* | 82 | 8 | 1993 - 1997 |
| *E. kochii* subsp. *kochii* | 29 | 4 | 1996 - 1997 |
| *E. kochii* subsp. *plenissima* | 72 | 8 | 1994 - 1997 |
| *E. kochii* subsp. *borealis* & *kochii* & *plenissima* | mix from above | 3 | 1998 - 1999 |
| *E. angustissima* | 117 | 3 | 1998 - 2001 |
| *E. polybractea* | 100 | 12 | 1993 - 2000 |

Concurrently, it had become clear that as a result of the strong selection for cineole, the breeding program may have inadvertently selected against biomass productivity. This partly arose from the findings of Milthorpe *et al.* (1998) who suggested that selections based on concentration of leaf oil may result in reductions of total cineole yield. To test this, a series of yield trials were established to compare the biomass performance of the progeny from the seed orchards to bulk collections of wild seed from the same species. Seven *E. loxophleba*, four *E. polybractea* and three *E. kochii* yield trials were planted between 2006 and 2011 which were assessed for biomass at ages three and six, and screened for foliar cineole. The results were clear, the breeding program had indeed increased cineole concentrations but there was a corresponding loss of biomass (unpublished data). This demonstrated the requirement for a new breeding direction with a stronger focus on biomass.

Another potential issue became apparent in the mallee breeding program. The selections for biomass improvement were done on sapling biomass assessments, not coppice. The heritability of coppice biomass and the genetic correlation between sapling and coppice had never been assessed. Wildy *et al.* (2000) found that when nine mallee taxa were subjected to a harvest at 2.5 years of age and 12 months later, coppice biomass production was strongly correlated with sapling biomass. Goodger *et al.* (2007) found from an *E. polybractea* trial comprising of 20 individuals subjected to harvest at 3.5 years of age, and two coppice harvests 12 and 24 months later, that there was a strong correlation between biomass and oil yield after each harvest. However, phenotypic correlation should not be the basis of a breeding program and the genetic parameters of coppice biomass production were identified as an important area for future research. This gap in breeding knowledge relating to saplings and coppice biomass heritabilities and the potential gains that could be achieved in progeny trials for *E. polybractea* has been assessed in Chapter 5 of this thesis.

Speculation arose as to the level of outcrossing from some of the mallee seed orchards. While undertaking cineole and biomass assessments from these trials, it became clear that while the flowering in the *E. kochii* and *E. polybractea* orchards was quite consistent between years, in the E. *loxophleba* orchards, trees were flowering throughout the year. Other eucalypt species have weak reproductively barriers and can self-pollinate which reduces fitness of the next generation (Eldridge & Griffin, 1983; Griffin & Cotterill, 1988; Hardner & Potts, 1995). Assessments of the flowering phenology became necessary to determine the synchronicity of flowering within seed orchards, especially for the two *E. loxophleba* subspecies. The implications for breeding strategy of the flowering phenology in a *E. loxophleba* seed orchard were assessed in Chapter 4 of this thesis. The work also included assessment of the genetic correlation between flowering traits with tree biomass and leaf cineole.

### *Second-round breeding population establishment*

In 2008 the Future Farm Industries Cooperative Research Centre (FFI CRC) was established with a major stream of research on new short-cycle woody crops for use in farming systems. It provided funds to establish new breeding populations of *E. polybractea* and two *E. loxophleba* subspecies, *lissophloia* and *gratiae* with a focus of genetic improvements in biomass production. These trials were established in 2010 in Western Australia (Brookton, Lake Bryde and Newdegate), South Australia (Monarto), New South Wales (Condobolin) and Victoria (Drummartin). Wild seed was again collected from native papulations but this time without initial screening for cineole. By the end of the FFI CRC, each individual mallee at each trial had been assessed for biomass (3-year old) and the trials in Western Australia had been screened for cineole (Mazanec *et al.*, 2017; Mazanec *et al.*, 2020; Mazanec *et al.*, 2021). Initial results from the two *E. loxophleba* subsp. *lissophloia* and *gratiae* trials indeed revealed a weak negative genetic correlation between biomass and foliar cineole concentrations, but positive genetic correlations between biomass to total cineole yield. These results suggest that gains in cineole yield could be realised while also selecting for biomass, and that strong selection for cineole was not required. Indeed, selection simulation incorporating biomass and cineole yield would not substantially reduce foliar cineole concentrations, but would increase total cineole yield, whereas selecting trees on foliar cineole concentrations would favour smaller trees (Mazanec *et al.*, 2017; Mazanec *et al.*, 2020).

### *Mallee response to harvest regime*

Early work on assessment of potential mallee yield in WA (Eastham *et al.*, 1993; Wildy *et al.*, 2000) preceded the emergence of a common understanding of best practice on species selection, planting site quality, design of planting layout and management. A major benefit of the early mallee planting undertaken by farmers was that it provided virtually every combination of species, site and design to help form a qualitative view of what works best (URS, 2008; Bartle, 2009). In addition, it provided a large choice of sites for experimental work on response to harvest treatments.

In spite of being well adapted for coppice regeneration, full canopy loss imposes a major stress on mallee. This burden can be expressed as sensitivity to the season and frequency of removal of the canopy by fire or harvest. This was demonstrated in a series of experiments conducted near Pooncarrie (mean rainfall 250 mm/year, evaporation 2400 mm/year) in the far west of NSW in the early 1980s (Noble, 1982; Noble, 1989; Noble & Diggle, 2014). This work examined the population dynamics of extensive mixed-species native mallee woodland in a region subject to pastoral use where fire risk, grazing productivity and conservation had become issues of concern. Survival following multiple decapitations, using both fire and manual harvests, were followed over several years. The findings show a progressive decrease in the rate of mortality for stands subject to less frequent autumn harvest, and a greater mortality rate in successive annual autumn compared to spring harvests. Furthermore, severe mortality occurred in autumn from two successive harvests one year apart. These results indicate the need for caution if shorter harvest intervals are applied, especially if this is done in autumn.

The long history of harvest of native mallee stands of *E. polybractea* (and more recently of planted stands) for eucalyptus oil production in NSW (West Wyalong) and Victoria (west of Bendigo), shows that a whole canopy, ground-level harvest is sustainable, as long as the harvest cycle is longer than 18 months (Davis, 2002). This is much more frequent than can be sustained at Pooncarrie. The more favourable climate in these locations (rainfall 550 mm and evaporation 1500 mm/year) is probably the major factor accounting for this difference. This is supported by recognition that harvest can be a little more frequent at West Wyalong during periods of above average rainfall (Davis, 2002). At nearby Condobolin, with a little less favourable climate, Milthorpe *et al.* (1998) observed that planted stands of *E. polybractea* and *E. kochii* could not sustain yield under annual spring harvests.

The physiological processes mediating response to harvest have been examined for *E. kochii* subsp. *plenissima* at Kalannie in WA where mean annual rainfall is 320 mm and potential evaporation 2400 mm year$^{-1}$ (Wildy & Pate, 2002; Wildy, 2003; Wildy *et al.*, 2004b). They observed that harvest caused loss of fine roots, reduced production of new fine roots, and retention of structural roots but with delayed secondary thickening. Root system recovery delayed recovery of coppice shoots to favour restoring a functional root to shoot ratio. The root to shoot ratio was more slowly restored for autumn harvests (2.5 years) than for spring (1.7 years). By contrasting young paddock-planted *E. kochii* in belts with the same species in nearby mature native vegetation they observed higher water use efficiency in coppice as gauged by mallee dry matter production. Several factors may explain this, e.g. the extant root system; preferential deployment of assimilates to coppice growth; the small ratio of stem to leaf biomass in coppice shoots; and the low ratio of leaf area to root system water catchment. This indicates that optimum biomass production from the available water supply will be achieved by mallee belts subject to a harvest frequency limited to the period needed for full root system recovery, including some active secondary root growth. These authors suggests a 2-3 year cycle depending on season of harvest. However, factors such as the biomass yield required for economic harvest, and impact of harvest frequency on the lateral root zone competition by belts on adjacent crop or pasture must also be considered in deciding the harvest frequency. Hence any assessment of yield must be done within the context of likely harvest regimes extended over multiple coppice harvests with the expectation of lengthened harvest frequencies with decreasing rainfall. The impact of harvest frequency and yield to these factors remained unknown and Chapter 2 of this thesis aims to address some of the knowledge gaps in yield prediction by assessing the impact of harvest regimes on biomass production from 19 mallee sites over 10-years.

### *Mallee biomass yield*

With the interest from industry, government and universities, many studies directly assessed the yield potential of mallee. These field plantings display large variation in yield as might be expected from long narrow belts across a typical WA wheat/sheep farm paddock that may include several different soil types and a range of site quality (Moore, 2001). Indeed, within belts, localised access to fresh ground-water can increase growth rates ten-fold (Brooksbank *et al.*, 2011). Huxtable *et al.* (2012) reviewed the yield from nine mallee trials and found a large range across all species and sites from 1 to 23 fresh Mg ha$^{-1}$ year$^{-1}$ (Wildy *et al.*, 2003; Wildy *et al.*, 2004a; Wildy *et al.*, 2004b; Pracilio *et al.*, 2006; Grove *et al.*, 2007; Carter & White, 2009; Liew, 2009; Sudmeyer & Daniels, 2010; Brooksbank *et al*., 2011). More recent work indicates similar productivity (Bennett *et al.*, 2015; Sudmeyer & Hall, 2015).

These studies, although useful, were not established to estimate long-term timeframes required to assess the stability of coppice production over several rotations which was viewed essential for mallee commercialisation. The previous studies included a range of methodologies including: a range of landscape positions, different ways of calculating the paddock space the mallee utilised, different species, and a wide range of mallee ages and harvest frequencies. In order to establish accurate estimates of mallee yield potential, in 2006, 19 long-term mallee harvest sites were established to capture the range of mallee yields using large plot size with appropriate replication. Due to the temporal and seasonal yield differences observed in mallee harvest, different harvest treatments were imposed. These 19 sites contained the largest long-term assessment of the range of biomass yield potential of mallee and results are contained in Chapter 2 of this thesis.

### *Mallee belt systems design*

It is well understood that for the widespread adoption needed for a mallee industry, mallee needs to be economically viable (Cooper *et al.*, 2005; Bartle & Abadi, 2010; Abadi *et al.*, 2012). The prospect of commercial viability will be improved if the additional landscape benefits including increased water use, erosion control and stock shelter are considered. So, the question emerges, what is the best planting configuration to balance these multiple objectives? There are a number of options to be considered including: whether to segregate or integrate trees; what is an ideal range for within and between row spacing; and if integrating, what is the optimal row number per belt, and distance between belts. These matters have been addressed to varying degrees, however, the most fundamental debate centres on the question of whether to integrate or separate.

Lefroy and Stirzaker (1999) argue that integration will generally be the preferred method with narrow belts of tree crops dispersed to manage salinity by exploiting excess water. However, there are two caveats; 1) that mallee would have to be commercially competitive with traditional agriculture; and 2) that for complete hydrological control, mallee would need to be planted on a significant proportion of the landscape, a point endorsed by George *et al.* (1999) and Hatton and Nulsen (1999). However, extensive integration of tree belts will impose extensive competition with adjacent annual crops and pasture. Lefroy and Stirzaker (1999) found that the integration of tagasaste did indeed reduce crop yield.

Sudmeyer *et al.* (2012) measured crop yield adjacent to mallee belts at 15 of the 19 sites from the long-term mallee project established in 2006. This project found that mallee did reduce agricultural yields adjacent to the belt; and that there were negative correlations between the magnitude of the crop loss and rainfall and a positive correlations with mallee age and mallee size with crops being more sensitive to competition than pasture. Across all sites, mallee competition reached an average of 11.3 m into the alley and yield was reduced by 36% (2 – 20 m from belt). The crop yield losses were reduced by 22 or 27% compared to open paddock yield when mallee was subjected to 3 or 6 year harvests respectively. This led Sudmeyer and Hall (2015) to argue for segregation of mallee from agriculture, but conceding the choice will ultimately be determined by what a biomass market will pay for mallee biomass.

In the meanwhile, mallee integration was still being asserted on the basis that while mallee might not solve salinity, it could mitigate the extent of the problem (Bartle, 2009), especially given the potential scale of mallee biomass markets emerging in the bioenergy and biofuels sector (Abdullah & Wu, 2009; Bartle & Abadi, 2010; McGrath *et al.*, 2016). The extensive reach of mallee belts would also achieve greater water use and biomass production per unit area, provide erosion control, shade and shelter for livestock protection and local and regional biodiversity benefits (Abadi *et al.*, 2012). It was also recognised, from the work by Ritson (2006) on *Pinus* *pinaster,* that much faster growth rate could be achieved on edge rows from both plantations and belts, compared to the internal rows. This was because of increased competition for resources. Applying these principles to mallee, the faster growth rates of belts would result in more frequent harvests which would in turn reduce competition to agriculture. The number of rows in belts, the within-row tree spacing, and the distance between belts were widely varied in planting undertaken in the 1990s and 2000s (Huxtable *et al.*, 2012).

The most common planting configuration of mallee in belt planting has historically been 4-row belts, with 2 m between rows and within-row trees spacing of 1.5 m (URS, 2008; Bartle, 2009). Four-row belts were historically preferred because this was the minimum width able to be monitored by the remote sensing technology available at the time by the Australian Greenhouse Office (URS, 2008). However, this restriction has since been retracted and for carbon sequestration (below- and above-ground) the new methodologies allowed greater flexibility of planting configurations including 2-row belts (Paul *et al.*, 2013; Department of the Environment, 2015a). It was also recognised that suppression of internal rows within a mallee belt could be severe reducing yield by up to 50% (Bartle *et al.*, 2012c; Huxtable *et al.*, 2012; Peck *et al.*, 2012). This is analogous to the edge effect observed in *P. pinaster* plantations (Ritson, 2006). Early trial plantings had highly variable within-row spacing. The comparative assessment of yield responses to number of rows and within-row spacing remained unknown and Chapter 3 of this thesis aims to assess how planting configuration affects mallee growth rates and survival of mallee in agroforestry plantings.

### *Mallee economics*

A common theme of this introduction has been that large scale adoption of mallee will only occur if they are demonstrably commercially viable. To determine aspects of the economics of mallee, various reports have been completed. The first was a simple economic analysis presented by Herbert (2000) who calculated cash-flow and break-even periods. He considered 2-row belts encompassing 15% of a paddock with three different harvest regimes. At productive sites initial harvest was at 4 years with subsequent harvests every two years. The other extreme was at saline sites, with first harvest at 6 years followed by harvest every three years. He estimated that at harvest age, the average tree would be 15 kg and estimated a price of \$30 Mg$^{-1}$ for mallee biomass. Transport costs within 50 km were \$15 thus the on-farm price was \$15 Mg$^{-1}$. Herbert (2000) found that mallee biomass would need to attract a price of between \$13 and \$24 Mg$^{-1}$ to be more profitable than existing enterprises. Mallee was found to be more profitable with more frequent harvests, with the lowest frequent harvest regime not profitable. When secondary landscape benefits were included, the system became more economically viable with a lower biomass price required for profitability. This model did not take into account the competition of mallee on agriculture, or of opportunity cost of land.

A more comprehensive mallee economics was completed by Cooper *et al.* (2005) who presented a conceptual model matching biophysical and economic conditions. At this time, experimental long-term biomass estimates were scarce and thus, mallee production was estimated as a function of capture and conversion of water to biomass. The work estimated the amount of biomass that could be produced within transport distance from an industrial processing facility. This model incorporated agricultural crop yields and crop foregone due to displacement by mallee plantings and by competition with mallee. The estimated yields to break-even with annual crops and pasture were between 16 to 41 dry Mg ha$^{-1}$ year$^{-1}$ depending on rainfall zone and transport horizon. To test this model, Narrogin and Merredin were selected and using the base-case scenario; higher rainfall Narrogin could generate 266,000 Mg dry biomass with 7% of the land planted and supplying enough biomass for a large-scale biomass industry, whereas biomass production at low rainfall Merredin was not economic. However, the model was sensitive to biomass price and very sensitive to water use efficiency. Changes to biomass price from \$30 to \$35 Mg$^{-1}$ and water use efficiency from 1.5 to 1.8 g/L revealed that Narrogin could produce up to 1.6 million Mg dry matter year$^{-1}$ and Merredin 239,000 Mg dry matter year$^{-1}$ with 16% of the land planted. This could support large-scale biomass industry based on biomass processing.

Abadi *et al.* (2006) also used biophysical and economic data to model the economics of several agroforestry crops, including mallee, in WA and NSW. Paddocks with and without agroforestry crops were compared using; net present value, annual equivalent returns, break-even periods and pay-back periods. Mallee biomass price was estimated to be \$30 per fresh Mg and at this price, mallee did not improve the economics of a paddock and would only be implemented for secondary benefits (e.g. salinity). However, like Cooper *et al.* (2005), this report found the model was sensitive to biomass price and mallee could be economic if a greater price could be obtained for biomass.

The Oil Mallee Association contracted URS to complete an Industry Development Plan which included an economic assessment of mallee over 30-years (URS, 2008). They used four regions based on rainfall, reduced crop revenues (competition and opportunity) and included income based on revenue from the Renewable Energy Target scheme of between \$0 and \$80 Mg$^{-1}$ of $CO_2$e. Biomass growth was estimated to be between 9 and 16 fresh Mg ha$^{-1}$ year$^{-1}$. They found that at \$0 $CO_2$e, mallee biomass would be profitable at \$40 per fresh Mg delivered to processing facility in the higher rainfall regions but \$45 to \$50 was required in the lower rainfall areas. However, with price of \$40 Mg$^{-1}$ of $CO_2$e, all regions were profitable at \$35 per fresh Mg delivered to processing facility.

Bartle and Abadi (2010) contemplated the economics of mallee biomass over a 50-year period to be used as a bioenergy feedstock. They subjected paddocks to 8% mallee configured in two-row belts to an initial harvest cycle at 5 years, and regular on-going harvests every 3-years with 50 Mg $ha^{-1}$ (fresh biomass) for each harvest. They also incorporated a price on below-ground carbon. Their delivery price of fresh biomass was \$45 $Mg^{-1}$. They found that cash flow from mallee agroforestry would exceed agricultural after 12 years with profits increasing over time.

By 2012, data were starting to emerge from the mallee harvest project comprised of 19 sites (Mendham *et al.*, 2012; Peck *et al.*, 2012). This increased the quality of the estimates used in the models twofold: firstly, it provided standardised experimental mallee biomass data; and secondly, it provided a function of competition of mallee on crops and pasture, later published as Sudmeyer *et al.* (2012). This allowed for greater accuracy of the economic models detailed below.

Abadi *et al.* (2012) included all aspects of mallee and crop costs and estimated mallee yield of 13 to 15 fresh Mg $ha^{-1}$ $year^{-1}$. They also estimated the secondary landscape benefits of mallee from \$2 to \$15 per fresh Mg of mallee produced and found that mallee cost the landholder (subtracting the secondary benefits) between \$37 and \$68 per fresh Mg of mallee biomass delivered to processing facility. They found that 35% of costs could be attributed to both opportunity and crop competition costs and also, harvest and haulage.

Bartle *et al.* (2012b) used mallee biomass data from the mallee harvest project and separated mallee yield into four rainfall zones which ranged from 4 to 20 fresh Mg $ha^{-1}$ $year^{-1}$. They used the new experimental mallee biomass data to estimate mallee yields within each rainfall zone. Harvest regimes of 3, 4, 5 and 6 years were applied. They also included a below-ground component generating carbon credits at \$25 $Mg^{-1}$ of $CO_2e$. To calculate the value of biomass, they considered the emissions of electricity generation from coal. Biomass supply chain (harvest and haulage) was modelled at \$44 fresh $Mg^{-1}$ and the model was run over a 50-year timeframe. The base case results show that mallee biomass systems were economically viable in the two higher rainfall zones and marginal at the 400-500 mm of annual rainfall and unviable at 300 – 400 mm. They found harvest and delivery costs account for 53% of the total costs of mallee and 28% due to crop competition and opportunity cost of land.

An Excel based paddock simulation model called IMAGINE was developed by Abadi and Cooper (2004) to estimate mallee biomass production. This was updated in the FFI CRC and is capable of bio-economic simulation for a range of land use systems (Bartle *et al.*, 2012a). It is flexible and can model agricultural production for any climatic or edaphic zone over one or more paddocks up to 50 years. It simulates the financial outcomes from land-use sequence of annual agriculture with perennial pastures or agroforestry, including mallee belt plantings. For agroforestry, IMAGINE can estimate the amount of above- and below-ground carbon sequestration. It can also estimate the competition imposed on the agricultural crop by mallee. Costs of production and commodity prices can either be generated from historical data or estimated using probability distributions. Results of IMAGINE are detailed in (Bartle *et al.*, 2012a) and later applied to various sites in WA and NSW (Farquharson *et al.*, 2013). Although a very useful tool, this has yet to be updated with more recent biomass and crop competition data.

The economic assessments of mallee, as summarised above, have been useful in assessing the economic viability and thus the scope for commercialisation potential of mallee. This has been constrained by the very limited market for mallee biomass, and consequently, the price per Mg of mallee biomass is unknown. Secondly, the input values for each model have been limited by the lack of long-term data sets. With the new mallee biomass and crop competition data generated from the FFI CRC, a new economic approach is possible. There are now 10-years of mallee biomass data with different harvest regimes from 19 trial sites. From the same sites, crop yield data was collected for 6-years including detailed assessments of crop competition imposed by mallee. These data are suited to Levelised Cost analysis, an economic tool which calculates the break-even cost of mallee biomass production, to inform industry of the real price of production. This economic assessment using actual agricultural inputs and production modelling had not been performed historically making economic decisions difficult for landholders. Chapter 5 of this thesis uses production and market data to assess the determinants of the economic viability of mallee eucalypts when integrated into annual agricultural systems.

## Thesis objectives and knowledge gaps

This thesis aims to advance the knowledge base by analysing historical and new mallee datasets. With more information available on mallee agronomy, breeding, yield potential, harvest management and economics, this thesis aims to fill gaps that may help to advance mallee commercialisation. An increase in deep rooted woody perennial cover could sequester $CO_2$ and mitigate hydrological imbalances (and other landscape sustainability management issues) arising from the widespread clearing of native vegetation across southern Australia.

This thesis draws on the nearly 30 years of research into the commercialisation of mallee agroforestry in WA by university, government and farmer groups. With the current Australian Government reluctance to adopt comprehensive carbon abatement, and carbon revenues being essential for a viable mallee industry, mallee industry research and development has declined over the past several years.  This thesis has allowed datasets from some of the long-term research work to be collected, interpreted and published in the public domain.

I aim to address some targeted questions, which may generate further interest in the potential of a mallee agroforestry industry. There is still some support in the current environment with a legislated price and mechanism to generate carbon credits from mallee planting in Australia (Paul *et al.*, 2013; Department of the Environment, 2015b). The current Australian price is about half the price of $CO_2e$ compared to other industrialised countries (Ramstein *et al.*, 2019) but is expected to rise. Further, as a result of the increased awareness of climate change in Australia, there has been a concerted effort made by industry to appear to be tackling climate change, for example, Australian financial institutions reluctance to finance a proposed coal mine in the Galilee Basin (Jolley & Rickards, 2020) and other fossil fuel projects. There is now an expectation that greenhouse gas emitting industries will take action on climate change to appease their shareholders. Mallee is one of the many tools available to mitigate climate change.

The mallee agroforestry system is well advanced compared to other woody biofuel perennial crops in Australia. There are technologies to convert mallee biomass to biofuels and bioenergy and methodology to generate carbon credits from mallee planting. However, there have been significant knowledge gaps to be addressed to assist an industry development based on integrated mallee agroforestry. These can be separated into five broad categories: yields, management, planting configuration, breeding and economics and are visually represented in Fig 2. This thesis addresses these by:

- Assessing mallee biomass production. Many studies have assessed mallee biomass yield using various methodologies which makes the yield potential for mallee difficult to estimate and this may be a barrier for industry. We address this by using site-specific allometric equations to estimates mallee biomass yield from 19 sites over a 10-year period. At each site, both harvested and unharvested mallee treatments are included. At more productive sites, this includes three 3-year harvest cycles. This study also correlates mallee yield response to site characteristics (edaphic and climatic) to assist in defining optimal sites (chapter 2; Forest Ecology and Management 2019).

- Examining management of mallee harvesting. The effect of frequency and season of harvest on biomass production. Decadal datasets are used with four different harvest regimes (spring and autumn harvests subjected to both short and long harvest cycles). How the three mallee species respond to the different harvest regimes is examined (Chapter 2; Forest Ecology and Management 2019).

- Assessing the effect of mallee planting configurations on productivity. Across two sites, this study estimates the biomass potential of 20 planting configuration treatments. There were four belt treatments with 1, 2, 4 and 6 row belts divided into five within row spacing treatments with 1, 1.5, 2, 3 and 4 m. Results are expressed on a plot level to compare planting treatments, including the alley to determine total biomass production at paddock level (Chapter 3, Agroforestry Systems 2020).

- Investigating the synchronicity of flowering in a mallee seed orchard and genetic parameters of biomass and cineole. This study determines the reproductive phenology from an *E. loxophleba* seed orchard. Data is presented from two annual assessments from a seed orchard comprised of two subspecies and 60 families. The assessment determines the variation in flowering between and within families, and the heritability of flowering traits and traits of interest: biomass and foliar cineole. This will allow for breeding decisions to remove out-of-phase families from the breeding populations without reducing gains in biomass or cineole (Chapter 4, Scientific Reports 2020).

- Assessing the heritability and genetic parameters of sapling and coppice production. This provides the first assessment of the heritability of mallee coppice success and biomass production. The genetic correlation between two sapling, and one coppice biomass assessments from three *E. polybractea* progeny trials in southern Australia is estimated. This determines optimal timing of selection to maximise mallee biomass production. (Chapter 5, Tree Genetics and Genomes 2021).

- Examine the economic viability of mallee and determines the levelised cost using experimental data. Six-year mallee biomass and crop yield datasets are used to determine the break-even price per Mg of fresh mallee biomass. Total costs of mallee production are calculated and separated into establishment and maintenance costs, harvesting costs, opportunity cost (of the land the mallee occupies) and competition cost imposed from mallee on agriculture (Chapter 6, GCB Bioenergy 2021).

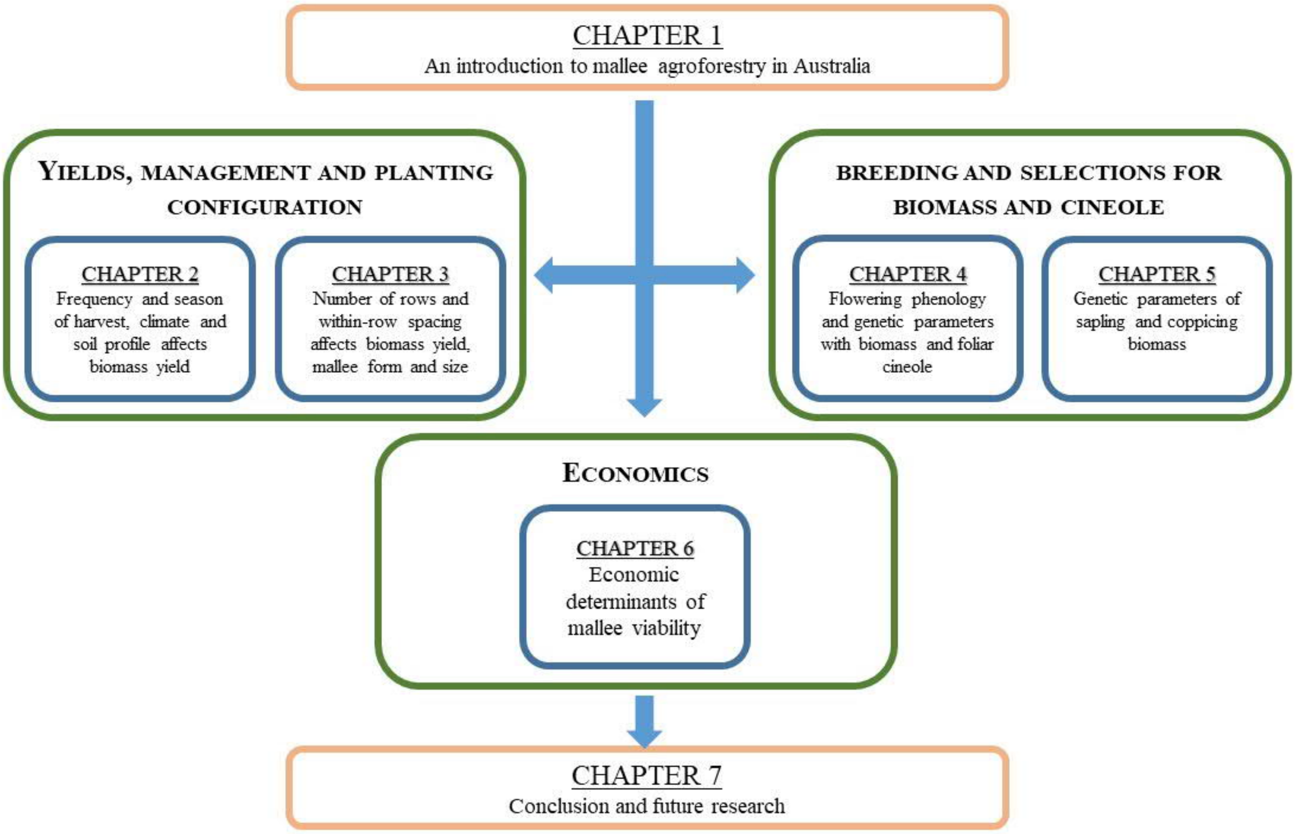

Figure 2 Schematic of thesis structure with arrows linking the broad research areas into chapters

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# Chapter 2

## Frequency and season of harvest, climate and soil profile affects biomass yield

Spencer B, Bartle J, Huxtable D, Mazanec R, Abadi A, Gibberd M, Zerihun A (2019). A decadal multi-site study of the effects of frequency and season of harvest on biomass production from mallee eucalypts. Forest Ecology and Management 453:117576. https://doi.org/10.1016/j.foreco.2019.117576

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# A decadal multi-site study of the effects of frequency and season of harvest on biomass production from mallee eucalypts

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## ABSTRACT

Mallee eucalypts are hardy, woody perennials that are being developed as a short-rotation coppice crop in Australia for the production of eucalyptus oil, biofuels and other biomass products. The economic viability of this prospective crop is dependent on its ability to survive and regenerate following repeated harvesting of the above ground component. Here we report on survival and biomass yield of mallee belt plantings of *Eucalyptus polybractea*, *E. loxophleba* ssp *lissophloia* and *E. kochii* ssp *plenissima,* at 19 sites, under two harvest-frequencies (3–8 year cycles) and harvest seasons (autumn or spring) over a decade from 2006 to 2015. 16 sites had post-harvest mortality ranging from 1.0% to 12.2% while the remaining three sites with either shallow saline water tables or a silcrete hardpan failed. Average site dry biomass yield across treatments ranged from 2.2 to 32.8 Mg $ha^{-1}$ $yr^{-1}$. Higher yielding sites were generally characterised by pH between 3.8 and 8, $EC_e$ below 15.0 dS $m^{-1}$ and high soil fertility. Lower yielding sites were generally near saline valley floors. After 7-years, biomass yield from unharvested treatments exceeded the average cumulative yield of harvest treatments at eight of the 16 sites, including all three *E. kochii* sites. For *E. loxophleba*, significant interactions were found between season and frequency of harvest with highest yields in long rotation spring treatments. There were also interactions between site and frequency of harvest, which were mainly driven by the variable performance of the uncut treatment. On average *E. loxophleba* yielded more biomass following spring harvests whereas *E. kochii* yielded more following autumn harvests. *E. polybractea* yield was unaffected by season or frequency of harvest; however, harvest treatments yielded more biomass than uncut treatments. After 10 years, at eight of the nine sites subjected to three 3-year cycles, no decline in biomass yield was observed. The site that declined in production was attributed to depletion of a sandplain aquifer by extensive mallee plantings. Overall, the results from this decadal study indicate that in warm-temperate semi-arid areas, such as the south-west of WA, mallees biomass can be harvested sustainably at most sites even in short (3-year) rotation cycles.

## 1. Introduction

The issues of sustainability in agriculture and climate change are driving development of options to improve the performance of agricultural systems (Foley et al., 2011). Perennial biomass crops have potential to be profitable and may also improve the sustainability of intensive annual crop/pasture agriculture (Brandes et al., 2016; Brandes et al., 2018; Dale et al., 2016; VanLoocke et al., 2016). Here we present biomass yield data from three decades of investment in the domestication of native mallee eucalypts for use as short-rotation coppice crops in the wheatbelt agricultural region in the south west of Western Australia. This development was motivated by the prospect of being able to better manage sustainability problems in traditional farming systems with the targeted integration of new perennial crops, but without any reduction in the economic viability of the farm (Bartle, 2009; Bartle and Abadi, 2009; Barton, 2000; URS, 2008).

The major sustainability problem in the Western Australian (WA) wheatbelt is dryland salinity (GHD, 2019), arising from the small change in the water balance that followed 20th century conversion of some 15 million hectares of native vegetation to agriculture based on annual crops and pastures (George et al., 1997; George, 1992; Hatton et al., 2003; Peck and Hurle, 1973). The native woody perennial

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vegetation is adapted to exploit any subsoil water infiltration (Robinson et al., 2006; Verboom and Pate, 2006) and this allowed a large storage of cyclic salt to accumulate (Hingston and Gailitis, 1976). The conversion to annual plant agriculture permits some deep soil water penetration, slow accumulation of groundwater and mobilisation of stored salt (George, 1992). Large areas of Western Australian farmland have already been degraded by this process, and it is projected that over several decades some 30% of the landscape could be affected (Simons et al., 2013) with adverse regional consequences for infrastructure, water resources and biodiversity protection (George et al., 2008).

The process of water infiltration into WA wheatbelt soil profiles and the movement of groundwater is dispersed and locally variable (George, 1992). Lefroy and Stirzaker (1999) considered the options to achieve complementarity from tree crops in the farm setting, i.e. to segregate, integrate or rotate. For salinity mitigation, integration in the form of widely separated, permanent, mallee belts on the recharging proportion of the farm was adopted as likely best practice for prime cropland. However, this could be complemented by segregated stands of mallee forests on poorer quality land. Design of mallee belt layout needs to achieve economically competitive biomass yields and contribute to salinity control, and as far as possible deliver other on-farm benefits including: reduction in wind erosion and provision of stock shelter (Baker et al., 2018; Bird et al., 1992; Sudmeyer et al., 2002; Sudmeyer and Scott, 2002); reduction of waterlogging (Ellis et al., 2006; Rundle and Rundle, 2002; Silberstein et al., 2002); protection and complementation of remnant native vegetation (Smith, 2009); and managing greenhouse gas emissions (McGrath et al., 2016; Wu et al., 2007).

Mallee eucalypts are a genetically diverse group within the genus *Eucalyptus*, with about 300 species widely distributed across the southern semi-arid region of Australia (Nicolle, 2006). They are tall shrubs or low trees that characteristically have multiple stems arising from a large below-ground lignotuber (Eastham et al., 1993; James, 1984; Wildy et al., 2000b). The lignotuber of mallee eucalypts is well endowed with meristematic tissue, an adaptation that enables prolific coppicing after decapitation by fire or drought, and enables regeneration after harvest (Noble, 1982; Noble, 2001; Noble and Diggle, 2014; Wildy and Pate, 2002).

Confidence in the potential of including mallee as a farm crop developed initially from native woodland stands of *E. polybractea* harvested on short rotations in both New South Wales (NSW) and Victoria. This commenced about a century ago to provide the international market for high cineole containing eucalyptus oil (Coppen and Hone, 1992; Davis, 2002). In recent decades this industry expanded into production from plantations established on former farmland (Davis, 2002).

In addition to cineole production, mallee eucalypts have been the focus of research as a bioenergy feedstock crop to mitigate climate change (Bartle and Abadi, 2009; McGrath et al., 2016; O'Connell et al., 2007; Shepherd et al., 2011). Many WA mallee species were screened for high biomass yield potential and leaf cineole content. Subspecies of both *E. loxophleba* L.A.S. Johnson and K.D. Hill and *E. kochii* C.A Gardner, were identified as prospective candidates (Wildy et al., 2000a; Wildy et al., 2000b). Together with *E. polybractea*, these three species of mallee eucalypts, hereafter referred to as ‘mallee’, provide a semi-arid climate analogue for the northern hemisphere cool temperate short rotation coppice species willow and poplar (Dickmann, 2006; Volk et al., 2016) and perennial grasses in the Mississippi Basin (Brandes et al., 2018; VanLoocke et al., 2016).

Early studies investigating fire and harvest of natural stands of mallee provide an indication of survival and biomass yield responses to various harvest regimes. In the pastoral region of NSW (mean rainfall 250 mm/year, evaporation 2400 mm/year) mallee mortality rates were sensitive to the frequency of ‘harvest’ (Noble, 1982; Noble, 1989a,b; Noble and Diggle, 2014); the effect was more pronounced when harvests were undertaken in autumn compared to spring. In a later study, Milthorpe et al. (1998) subjected 1-year old plantations of *E. polybractea* and *E. kochii* in Condobolin NSW (mean rainfall 460 mm/year, evaporation 1800 mm) to annual harvest for 5-years. Yield improved on the second harvest, but thereafter declined from 6 to 2 Mg ha$^{-1}$. Similarly, Davis (2002) observed failing vigour after 9-years of annual harvests for *E. polybractea* at West Wyalong NSW (mean rainfall 480 mm/year and evaporation 1600 mm). These observations suggest implications for mallee biomass production arising from the choice of season and frequency of harvest, especially in more arid regions.

Wildy and Pate (2002) and Wildy et al. (2004b) examined the mallee root system responses to harvest. They demonstrated that newly harvested trees (*E. kochii* ssp *plenissima*) showed loss of fine root ($<$ 2 mm) biomass, arrested production of roots, secondary thickening of structural roots, and an increasing shoot to root ratio to restore a functional balance. The recovery of root biomass to pre-harvest levels took 1.7- to 2.5-years. Spring harvests showed earlier and stronger root and shoot growth than late summer harvest.

The growth of individual trees and tree stands through time have been well described e.g. Richards (1959). In the general growth function, a tree grows slowly at an early age, reaches exponential growth at middle age and then declines with old age (e.g. Johnson and Abrams (2009), Zhao-gang and Feng-ri (2003)). In a coppice system, the growth function model is reset after harvesting. Thus, a key driver of biomass production is likely to be the frequency of harvest and, to maximise production, management decisions can be imposed to align timing of harvest with growth rate. Estimations have been made concerning when to harvest mallee under short-rotation. For instance, Bartle and Abadi (2009) proposed a conceptual model where mallee are first harvested at 5-years of age, then every subsequent 3-years. Yu et al. (2015) conducted a life cycle analysis based on harvest at 7-years with subsequent harvests every 4- to 5-years. Both of these cases contain models where biomass estimations were based on best available data and stability of productivity over time was inferred.

This paper presents results of different harvest regimes at 19 sites, covering a range of landforms and soil types across the WA wheatbelt, where mallee belts were monitored and harvested over a 10-year period. A subset of these results have been previously presented in research reports (Mendham et al., 2012; Peck et al., 2012). This paper reanalyses the original data, with the inclusion of three additional years of biomass data, testing the following hypotheses:

1. That frequency and season of harvest have no effect on biomass production or mortality;
2. There is no decline in the biomass yield caused by the cumulative effect of regular harvest.

This paper also presents the allometric relationships used to calculate total dry biomass and dry components (wood, bark, twig and leaf) of the three mallee species: *E. loxophleba* subsp. *lissophloia*, *E. polybractea* and *E. kochii* subsp. *plenissima*.

## 2. Materials and methods

This study concludes a long term research project that was set up across 19 sites with the dual purpose of quantifying biomass production of mallee belts subjected to contrasting harvest regimes, and to determine the competition imposed on adjacent annual crop and pasture over the harvest cycle (Mendham et al., 2012; Peck et al., 2012; Sudmeyer et al., 2012a). The work reported here is principally concerned with mallee biomass production. Site 5 from the previous reports was excluded from the statistical analysis, as this site traversed three distinct site types rendering the 3 replications inadequate. Site names were left consistent with previous reports to allow for further examination of data.

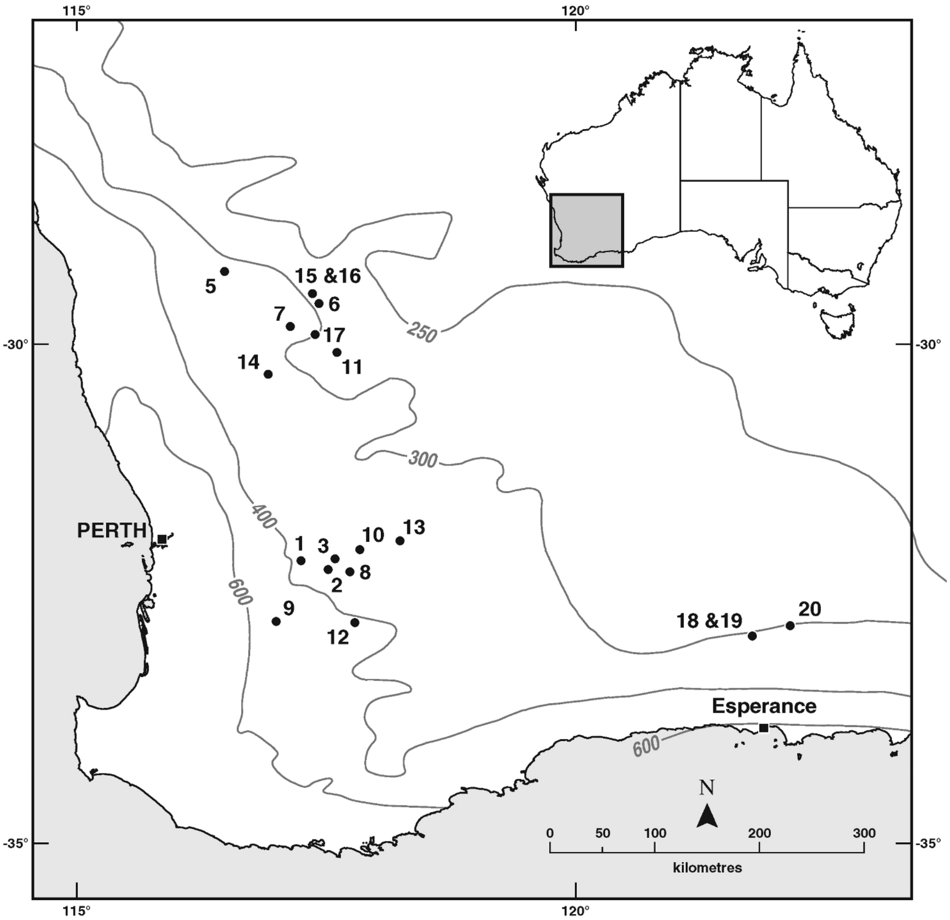


**Fig. 1.** Location of mallee trial sites within the Western Australia wheatbelt; also shown are selected rainfall isohyets (grey line). The site numbers shown on the map correspond to those in Table 1.

### 2.1. Species and study site

This study investigated the three major mallee species widely adopted for planting by farmers in WA: *Eucalyptus loxophleba* subsp. *lissophloia*, *E. polybractea* and *E. kochii* subsp. *plenissima*. These species will be hereafter referred to as $E_{lox}$, $E_{pol}$ and $E_{koc}$ respectively. Typically $E_{pol}$ has been planted in higher rainfall zones of the southern wheatbelt, $E_{koc}$ has been planted in the northern wheatbelt where it naturally occurs and $E_{lox}$ has been planted throughout the wheatbelt (*c.f.* Fig. 1 and Table 1).

This study ran for 10-years (2006–2015) using 19 mallee belt plantings that had been established several years earlier by farmers in the WA wheatbelt (Fig. 1). For site details see Table 1. Sites were chosen to be representative of the range of species and environmental conditions where belt plantings of mallee had been undertaken (for detailed landscape and soil information see Table S1).

The wheatbelt has a Mediterranean climate with hot dry summers and mild wet winters. Over the duration of this experiment, mean annual rainfall ranged from 628 mm on the south-coast to 304 mm in the north-east, and evaporation ranged from 1646 mm to 2575 mm (Table 1). The crops and pastures of this region are predominantly non-irrigated, winter-growing annuals.

### 2.2. Experimental design

All sites were planted in belt configurations of two, three, four or six rows within paddocks subject to rotations of annual crops and pasture. However, each plot within this study consisted of only two rows, an edge row and the adjacent row (Fig. 2). The plots ran along the belt and were either 20 m long with a 10 m buffer between plots or 25 m long with a 12.5 m buffer.

Each site consisted of 15 plots, three replicates of five treatments in a randomised block design. The four harvest treatments were arranged in a 2x2 factorial design with the addition of an uncut control treatment. There were two season of harvest treatments, spring and autumn; and two frequency of harvest treatments, 3- and 4-year. All mallee in each plot were measured annually and harvested in the allocated seasons. However, it became apparent prior to the second harvest that the allocated 3- and 4-year harvest frequencies were not viable on poorer sites due to concerns of mortality and increased unit costs of biomass harvesting. Harvest frequency was reduced until coppice biomass exceeded approximately 10–15 dry kg/tree to keep harvest cost of below AU\$30 $Mg^{-1}$ (Spinelli et al., 2014). With these considerations, the final frequency of harvest treatments imposed were: short with a minimum of 3-years between harvests and long which was the short harvest cycle duration plus one year. The more productive sites (9 sites in total) completed three short and two long coppice cycles while the least productive sites only underwent one full cycle.

### 2.3. Site soils physico-chemical characteristics

Between 2008 and 2010, soil cores were drilled at each site using an EVH Rhino 2100 drill rig 20 m from the centre of each uncut treatment replicate. The total core depth was limited to 10 m or by the presence of a hardpan or water table. Soil cores were tested for $EC_e$ (dS $m^{-1}$), pH ($CaCl_2$) and nutrients ($NH_3$, $NO_3^-$, Cowell P, Cowell K, S and organic C) approximately every 50–60 cm. The soil nutrient content from each core was estimated in the top 50 cm of the soil profiles and corrected for

**Table 1**
Site characteristics and planting configuration for all field sites. Meteorological data obtained through SILO (Jeffrey et al., 2001) datasets from 2005 to 2014. Site 4 was never established. Sites names have been kept consistent with previous publications for traceability.

| Site | Lat (°S) | Long (°E) | Species | Year planted | Mean annual rainfall (mm) | Mean annual pan evaporative (mm) | Mean maximum temperature (°C) | Mean minimum temperature (°C) | Number of rows | Alley width (m) | Plot length (m) |
|---|---|---|---|---|---|---|---|---|---|---|---|
| 1 | −32.87 | 117.25 | $E_{pol}$ | 1996 | 443 | 1781 | 23.4 | 10.0 | 2 | 70 | 20 |
| 2 | −32.96 | 117.52 | $E_{lox}$ | 1997 | 406 | 1775 | 23.3 | 9.9 | 2 | 20 | 20 |
| 3 | −32.85 | 117.59 | $E_{lox}$ | 2000 | 396 | 1811 | 23.4 | 9.9 | 3 | 50 | 20 |
| 5 | −29.98 | 116.48 | $E_{lox}$ | 1998 | 308 | 2577 | 27.0 | 12.4 | 4 | > 250 | 20 |
| 6 | −30.30 | 117.43 | $E_{lox}$ | 1999 | 304 | 2556 | 26.8 | 12.5 | 3 | 95 | 20 |
| 7 | −30.52 | 117.14 | $E_{lox}$ | 2000 | 305 | 2493 | 26.5 | 12.2 | 2 | 40 | 20 |
| 8 | −32.99 | 117.73 | $E_{pol}$ | 1998 | 396 | 1785 | 23.2 | 9.8 | 4 | 125–250 | 20 |
| 9 | −33.48 | 117.00 | $E_{lox}$ | 1999 | 486 | 1646 | 23.2 | 9.4 | 2 | 35 | 20 |
| 10 | −32.76 | 117.83 | $E_{lox}$ | 1998 | 364 | 1889 | 23.9 | 10.2 | 2 | 36 | 20 |
| 11 | −30.78 | 117.61 | $E_{koc}$ | 1999 | 322 | 2401 | 26.1 | 11.9 | 2 | 30–250 | 20 |
| 12 | −33.49 | 117.79 | $E_{lox}$ | 2000 | 417 | 1722 | 22.9 | 9.8 | 6 | 55 | 20 |
| 13 | −32.67 | 118.24 | $E_{lox}$ | 1997 | 362 | 1972 | 24.6 | 10.4 | 2 | 48 | 20 |
| 14 | −30.01 | 116.91 | $E_{lox}$ | 2000 | 340 | 2357 | 26.1 | 11.7 | 4 | 40–120 | 20 |
| 15 | −30.21 | 117.36 | $E_{koc}$ | 1998 | 319 | 2567 | 26.8 | 12.4 | 2 | 95 | 25 |
| 16 | −30.18 | 117.37 | $E_{koc}$ | 1994 | 324 | 2575 | 26.8 | 12.4 | 2 | 95 | 25 |
| 17 | −30.60 | 117.39 | $E_{lox}$ | 1999* | 306 | 2466 | 26.4 | 12.2 | 2 | 50 | 25 |
| 18 | −33.63 | 121.77 | $E_{pol}$ | 2001 | 610 | 1929 | 22.8 | 11.2 | 6 | 90–100 | 25 |
| 19 | −33.63 | 121.76 | $E_{pol}$ | 2001 | 628 | 1935 | 22.8 | 11.4 | 6 | 120–140 | 25 |
| 20 | −33.52 | 122.16 | $E_{lox}$ | 2001 | 513 | 1951 | 22.6 | 10.8 | 6 | 150–250 | 25 |

* Initially harvested in Feb 2002 prior to trial.

the bulk density of the soil horizon. pH and $EC_e$ were expressed as the maximum and minimum recorded from each core. Further methodology for soil data collection are detailed in Sudmeyer et al. (2012b) whereas soil properties and classification for each site are described in Appendix B from Peck et al. (2012). The soil data that were used for analyses described in this study are presented in Table S2.

### *2.4. Assessment of above ground fresh biomass (AGFB)*

Assessments of AGFB were made bi-annually across the 10-years of the trial. The autumn harvests and uncut treatments were measured in March and April while the spring treatments were measured in September and October. To estimate AGFB for uncut mallee, stem diameters in mm were measured at 10 cm above ground level with a diameter tape, or callipers, in which case two perpendicular measurements were taken. Where buttressing or lignotubers affected the measurement, the stems were measured at the point closest to 10 cm above the ground where it most accurately reflected the true stem diameter.

To provide a single diameter estimate for multiple stemmed uncut mallee the Equivalent Diameter near Root Collar (EDRC) in cm was calculated as in Chojnacky and Milton (2008):

$$EDRC = \sqrt{\sum_{i=1}^{n} \mathrm{drc}_i^2} \qquad (1)$$

where *drc* equals the diameter of each stem near the root collar and n equals the number of stems from an individual plant. Diameter was used for AGFB estimation except at Site 11 that was dominated by small mallee with bushy forms and multiple stems. For such uncut and coppiced mallee, Crown Volume Index (CVI) was deemed a more accurate biomass assessment method. CVI estimates the volume of the coppice in $m^3$ as:

$$CVI = H \times W_i \times W_j \qquad (2)$$

where $H$ is the height and $W_i$ and $W_j$ are perpendicular, horizontal crown widths in cm. Site 17 had been harvested in February 2002, 4-years before the commencement of this project and the EDRC method

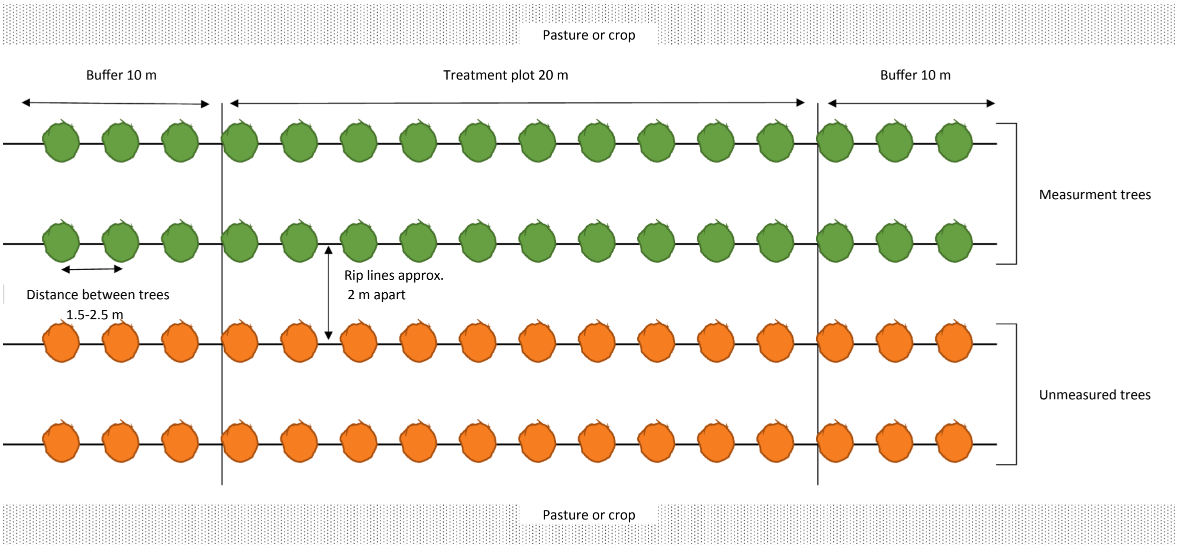


**Fig. 2.** Schematic showing layout of a four-row mallee belt, and location of measurement plot and buffers. Mallee rows are planted on 0.5 m deep rip lines typically 2 m apart and a within-row spacing of between 1.5 and 2.5 m. Sites 15–20 had 25 m plots with 12.5 m buffers.

was used for the initial measurement.

Measurements at three of the nineteen sites were terminated due to high mortality and poor performance. At Sites 6, 7 and 14 coppice measurement ceased in 2012, 2008 and 2009 respectively. Measurements continued for the uncut treatment until the conclusion of the project at Site 6 whilst at Sites 7 and 14 uncut treatments were measured until 2011 (see Table 7).

### 2.5. Destructive measurements

Biomass was harvested with a chainsaw close to ground level and weighed in a trailer fitted with Ruddweigh 600 mm load bar (precision ± 0.1 kg). Uncut and coppiced mallee were selected using a stratified random selection method as detailed in (Snowdon et al., 2002). The uncut and coppiced mallee were stratified into 12 diameter or CVI size classes and samples from each group were randomly selected. A range of 20–122 uncut and 45–142 coppice were destructively sampled from each site (see Tables S4 and S5 for more details). Trees for uncut treatments were destructively sampled in 2006 with the initial spring and autumn harvests. Most of the coppice were destructively sampled at first coppice harvest in 2009–2011. Additional coppice and uncut mallee were destructively sampled during latter years when the size of the mallee out-grew the range of the initial size classes, increasing the size range of the allometric model. The additional uncut samples were harvested from areas of the planting not part of the experiment.

### 2.6. Partitioning and moisture content

For all but site 14, uncut mallee were stratified into three size classes and two to four mallee per size class were selected per site, as described by Snowdon et al. (2002), except at Site 13, where only four trees were sampled. Total weight of wood with bark (> 20 mm diameter over bark), and twig with leaf (< 20 mm diameter over bark) were recorded. These were further partitioned into four biomass components (wood, bark, leaf and twig). Sub-samples of each of the four components were weighed in the field. The subsamples were transported to a laboratory and oven dried at 70 °C. When the weights of the samples had stabilised, the final weight was recorded. A similar method was applied to coppice with data collected from 11 of the 19 sites.

### 2.7. Mallee survival

Gaps in belts were recorded at commencement in 2006 and later deaths were recorded annually during biomass assessments. Percent survival was the difference between the mallee counts at the initial and the subsequent years of measurement. Mallee deaths were verified the following year and thus no new deaths were recorded in 2015, the last measurement year. This method accommodated the occasional observation of delayed coppice regeneration.

### 2.8. Allometry

Mallee allometry has been published in Paul et al. (2016) and Paul et al. (2013b), however new equations were developed for this study because of site and species bias inherent in using generalised equations. Dry biomass components of uncut or coppiced mallee were estimated by two-step allometric modelling. In the first step, generic and species-specific allometric models were developed to estimate AGFB from EDRC/CVI. In the second step, a further set of allometric models were developed to estimate dry components from AGFB. The two models are described below:

$$ln(AGFB_{Indiv}) = ln(a) + b * ln(x) + \varepsilon \tag{3}$$

where $\mathrm{AGFB_{Indiv}}$ is above ground fresh biomass of the individual mallee, $a$ and $b$ are parameter estimates, $x$ is $EDRC$ in cm or $CVI$ in $m^3$ from Eqs. (1) and (2), respectively and ε is the error term. To estimate dry components further allometric equations were developed using:

$$ln(AGDBC_i) = ln(c_i) + d_i * ln(q) + e_i \tag{4}$$

where $AGDBC_\mathrm{i}$ is above ground dry biomass for $component_i$ (i.e., leaf, twig, bark or wood) of the individual mallee, $c_i$ and $d_i$ are parameter estimates for $\mathrm{component_i}$, $q$ is $AGFB$ and $\mathrm{e_i}$ is the error term for $component_i$ biomass model. In all cases, estimates $a/c_i$ and $b/d_i$ were calculated using linear least-squares procedures. Due to the time-demand of data collection for component partitioning, species-specific models were tested and site-level analysis was omitted. General linear modelling (GLM) was used to test whether species improved the performance of the models.

For all allometric equations, the residuals were checked for heteroscedasticity. Data from 10 uncut and 24 coppiced mallee were removed from analysis as those were deemed data entry errors or were small individuals (EDRC or CVI < 26 mm or < 0.6 $m^3$) which may not conform to scaling laws (Enquist et al., 2007). Two correction factors were assessed to remove bias from back-transformation (Baskerville, 1972; Beauchamp and Olson, 1973) but in this study the two methods had a maximum difference in biomass estimation of 0.67%, so the more common methodology of Baskerville (1972) was used.

### 2.9. Plot biomass estimation

The individual biomass of uncut and coppiced mallee was calculated by converting EDRC or CVI to fresh biomass (Eq. (3)) then applying the component allometrics (Eq. (4)) and summing the partitioned dry components:

$$Total\ \ AGDB = \sum\nolimits_{i=1}^{n} C_i \tag{5}$$

where AGDB is above ground dry biomass, $C_i$ is the dry weight of tree/coppice biomass component, $i$: wood (> 20 mm diameter over bark), bark, twig (< 20 mm diameter over bark) and leaf.

To standardise land area under different planting configuration and mallee size, the edge of the plot adjacent to the crop/pasture was calculated as half the internal distance between rows within a belt extended into the adjacent crop, as explored in Appendix 9 by Paul et al. (2013a). Plot-level biomass estimates were divided by the total number of years of growth and expressed in dry biomass per hectare per year (Mg $ha^{-1}$ $yr^{-1}$).

A preliminary analysis showed biomass yield symmetry between rows within a belt Figs. S1–S20. For example, in a four row belt, row one biomass is equivalent to biomass from row four; row two is equivalent to row three. This was tested from replicated datasets from 20 sites and at 18 sites there was no difference between the stem diameters between the outer two rows each side of a mallee belt in paired comparison $t$-tests ($P > 0.05$) (Figs. S1–S20). Therefore in this study, to standardise biomass estimates between sites with a different number of rows, sites with more than two-rows have been compensated with the additional biomass from the unmeasured rows using belt symmetry principles (e.g. doubling the measured two-row biomass from a four-row belt to estimate total biomass).

### 2.10. Statistical analysis

#### 2.10.1. Treatment effect evaluation

Treatment effects were evaluated using general linear models:

$$\mathrm{y}_{ijklm} = \mathrm{s}_i + \mathrm{r}_k(\mathrm{s}_i) + \mathrm{u}_j + \mathrm{a}_l(\mathrm{u}_j) + \mathrm{f}_m(\mathrm{u}_j) + \mathrm{s}_i.\ \mathrm{a}_l(\mathrm{u}_j) + \mathrm{a}_l.\ \mathrm{f}_m(\mathrm{u}_j) + \mathrm{s}_i\ .\ \mathrm{f}_m(\mathrm{u}_j) + \mathrm{s}_i.\ \mathrm{a}_l.\ \mathrm{f}_m(\mathrm{u}_j) + \mathrm{e}_{ijklm} \tag{6}$$

$$\mathrm{y}_{ijkm} = \mathrm{s}_i + \mathrm{r}_k(\mathrm{s}_i) + \mathrm{u}_j + \mathrm{f}_m(\mathrm{u}_j) + \mathrm{s}_i.\ \mathrm{f}_m(\mathrm{u}_j) + \mathrm{e}_{ijkm} \tag{6a}$$

where y is AGDB, $\mathrm{s}_i$ is the $\mathrm{site}_i$, $\mathrm{r}_k(\mathrm{s}_i)$ is the $\mathrm{replicate}_k$ nested within $\mathrm{site}_i$, $\mathrm{u}_j$ is the $\mathrm{cut/uncut}_j$ harvest treatment, $\mathrm{a}_l$ is the $\mathrm{season}_l$ of harvest, $\mathrm{f}_m$ is the $\mathrm{frequency}_m$ of harvest and $\mathrm{e}_{ijklm}$ is the residual error. All main effects and interactions were nested within $u_j$. The site effect was tested using rep (site) as an error term. Because the spring treatments were modified at most sites after 7-years, two sets of analyses were performed to test: (i) the effect of all treatments (season of harvest and frequency of harvest) on the 7-year dataset (Eq. (6)), and (ii) the effect of frequency of harvest on the 10-year dataset with the season factor $a_l$ excluded (Eq. (6a)). Further, because each species occurred at one site only, separate analyses were done for each species. Where necessary data were transformed using Box-Cox transformation to meet assumptions of normality and variance homogeneity. Tukey-Cramer tests were used to compare least square means (LSM) at different sites, treatments and site by treatment interactions.

Eq. (6) was also used to test the factors responsible for mortality. The arcsine transformation was used to approximate the model. Other models were tested, including binomial distribution with logit link function but these specifications did not converge. Since the failed sites were only measured for part of the experiment, two analyses were performed: (i) a three-year analysis that including the three failed sites excluding the effect of frequency of harvest because none of the sites were harvested and (ii) a 10-year analysis using Eq. (6a). Site 6 was excluded from the analysis because neither long autumn nor long spring treatments were established.

### *2.10.2. Relationships of mallee growth to environmental variables*

Stepwise multiple regression of annual growth on soil, climatic and landscape variables (listed in Table S3), were carried out to identify environmental variables that explain observed growth responses. Variables were included in the regression if they improved model fit ($P < 0.05$). Collinearity was tested by checking the variance inflation factor with a cut off of 10 (Chatterjee and Ali, 2015). Every site, including the failed sites, was included in this analysis to identify site variable/s predicting poor performance.

All analyses reported here were performed using SAS v9.4 (SAS, 2017).

## 3. Results

### *3.1. Estimation of mallee biomass*

Allometric models were used to estimate dry biomass in a two-step process. For the first step of the estimation process (Eq. (3)), species- and site-specific allometric models based on EDRC in cm or CVI in $m^3$ explained between 79% and 98% of the variation in AGFB (Tables 2, S4 and S5). For all species-specific and most of the site-specific models, the uncut mallee models had a better fit than the coppice models.

The second stage of the allometric modelling (Eq. (4)), estimated dry component biomass (wood, bark, twig and leaf) from AGFB models and accounted for 87–98% of variances, depending on component biomass (Tables 3 and 4). GLMs revealed species differences for both uncut and coppiced mallee. For uncut mallee equations, there were significant differences in both slope and intercept between species, primarily due to $E_{lox}$ allometry being different from allometries of the other two species (Table 3). Inclusion of species in coppice allometries also improved fit of the component biomass models. The species effects were realised in intercepts (Table 4). The biomass components changed with tree size for all treatments and species. For the uncut treatment dry components across the species ranged from 26.5 to 30.3% for wood, 3.9–7.5% for bark, 14.5–16.3% for twigs, and 8.9–10.6% for leaves. For the coppice treatments, the respective ranges were 10.7–18.1% for wood, 2.9–3.4% for bark, 21.7–24.4% for twigs and 15.4–17.9% for leaves. Thus, in the harvested treatments, foliage accounted for a larger percentage of total biomass than in the uncut treatments, which conversely had a larger percentage of wood and bark.

**Table 3**
Allometric models for estimating above ground dry components (wood, bark, twig and leaf) of uncut mallee from AGFB (Eq. (4)). Numbers in parentheses are standard errors. Sample sizes and the largest biomass per uncut mallee for each species model were: $E_{koc}$ n = 25, upper domain = 327.7 kg, $E_{lox}$ n = 89, upper domain = 702 kg, and $E_{pol}$, n = 34 and upper domain = 683.8.

| Component | Species | Intercept ($d_i$) | Slope ($c_i$) | MSE | $R^2$ |
|---|---|---|---|---|---|
| Wood | $E_{pol}$&$E_{koc}$ | −2.640 (0.092) | 1.271 (0.016) | 0.039 | 0.978 |
| | $E_{lox}$ | −2.295 (0.140) | 1.224 (0.034) | 0.036 | 0.98 |
| Bark | $E_{pol}$ | −4.180 (0.126) | 1.173 (0.030) | 0.042 | 0.973 |
| | $E_{koc}$ | −2.556 (0.284) | 1.006 (0.064) | | |
| | $E_{lox}$ | −3.197 (0.152) | 0.990 (0.038) | | |
| Twig | $E_{pol}$&$E_{koc}$ | −1.190 (0.098) | 0.858 (0.018) | 0.047 | 0.947 |
| | $E_{lox}$ | −1.296 (0.045) | | | |
| Leaf | $E_{pol}$&$E_{koc}$ | −0.909 (0.118) | 0.695 (0.021) | 0.064 | 0.897 |
| | $E_{lox}$ | −0.991 (0.052) | | | |

**Table 4**
Allometric models for estimating above ground dry components (wood, bark, twig and leaf) of coppiced mallee from AGFB (Eq. (4)). Numbers in parentheses are standard errors. Sample sizes and the largest biomass per coppice for each species model were: $E_{koc}$ n = 17, upper domain = 77.8 kg; $E_{lox}$ n = 75, upper domain = 49.8 kg; and $E_{pol}$, n = 59 and upper domain = 194 kg.

| Component | Species | Intercept ($d_i$) | Slope ($c_i$) | MSE | $R^2$ |
|---|---|---|---|---|---|
| Wood | $E_{pol}$&$E_{lox}$ | −3.424 (0.213) | 1.408 (0.052) | 0.119 | 0.878 |
| | $E_{koc}$ | −3.753 (0.097) | | | |
| Bark | $E_{koc}$&$E_{lox}$ | −4.520 (0.109) | 1.303 (0.046) | 0.094 | 0.877 |
| | $E_{pol}$ | −4.798 (0.177) | | | |
| Twig | $E_{koc}$&$E_{pol}$ | −0.801 (0.091) | 0.822 (0.021) | 0.023 | 0.939 |
| | $E_{lox}$ | −0.955 (0.030) | | | |
| Leaf | $E_{koc}$ & $E_{lox}$ | −1.298 (0.136) | 0.896 (0.032) | 0.051 | 0.87 |
| | $E_{pol}$ | −1.422 (0.121) | | | |

### *3.2. Mortality*

Mortality varied from about 1% to 45% across sites; the highest mortalities (> 30%) occurred at valley floor Sites 6 and 7 which had saline ground water close to the surface or Site 14 with a silcrete hardpan at a depth 2–4 m, while the lowest mortality rates were observed at sites with relatively low soil constraints (Fig. 3(a and b)).

**Table 2**
Parameterisation of allometric model for species-specific prediction of above ground fresh biomass (AGFB) of uncut tree from 'equivalent diameter near root collar' (EDRC) in cm and coppice from Crown Volume Index (CVI) in $m^3$ (Eq. (3)). Sample number (n), coefficient of determination ($R^2$), Mean Squared Error (MSE), and values in parentheses are standard errors of parameter estimates.

| Species | Treatment | n | AGFB range (kg) | Intercept ($b$) | Slope ($a$) | $R^2$ | MSE |
|---|---|---|---|---|---|---|---|
| $E_{koc}$ | Uncut tree | 178 | 0.8–331.5 | −2.315 (0.138) | 2.471 (0.053) | 0.926 | 0.058 |
| $E_{lox}$ | Uncut tree | 724 | 0.5–702.0 | −2.120 (0.054) | 2.535 (0.025) | 0.936 | 0.068 |
| $E_{pol}$ | Uncut tree | 112 | 4.5–771.5 | −2.008 (0.154) | 2.463 (0.058) | 0.943 | 0.052 |
| $E_{koc}$ | Coppice | 156 | 1.0–107.3 | 0.786 (0.084) | 0.956 (0.027) | 0.892 | 0.033 |
| $E_{lox}$ | Coppice | 775 | 0.1–152.5 | −0.094 (0.043) | 1.074 (0.014) | 0.877 | 0.067 |
| $E_{pol}$ | Coppice | 508 | 0.1–316.0 | 0.221 (0.050) | 1.055 (0.015) | 0.902 | 0.066 |

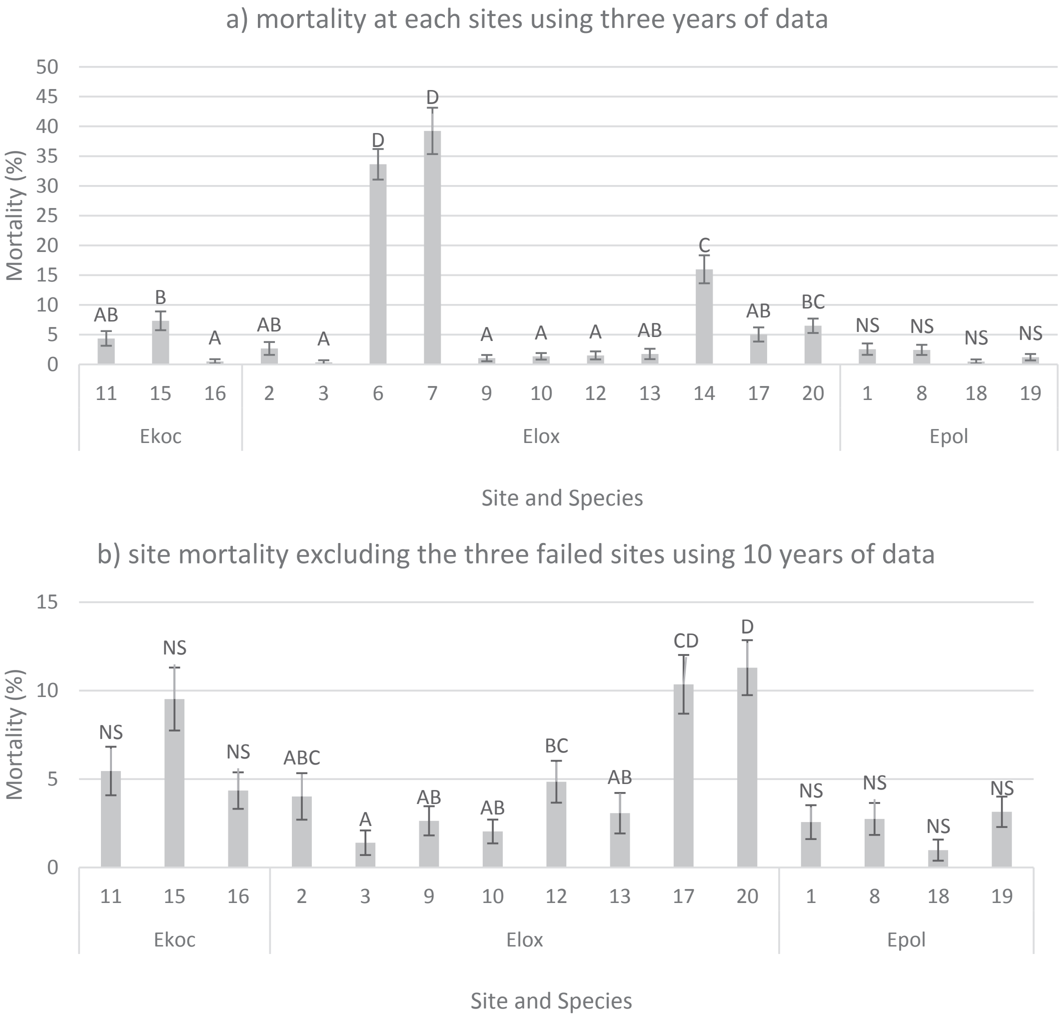


**Fig. 3.** Mortality (%) by site and species averaged across all harvest treatments ( ± s.e.). (a) Analysis included all trial sites using 3-years of data and (b) analysis excluded three failed sites and used 10-year data sets. Within a species, site means that are followed by different letters signify a difference in mortality at $P < 0.05$. Site was not significant (NS) for $E_{koc}$ or $E_{pol}$ sites.

By the end of the study, $E_{koc}$ had overall mortality rate of 6.2%. The season of harvest had more effect on survival than frequency of harvest, with spring treatment mortality of 9.1% compared to autumn harvest treatment of 3.4% or uncut treatment of 6.0% ($P < 0.05$) (Table 5). There was a site by season interaction ($P < 0.05$) due to elevated spring mortality at Site 15 (23.1%) and the uncut treatment at Site 16 (10.0%). Most of the spring mortality was from the initial harvest. Using the 3-year dataset, there was a significant site by season effect ($P > 0.05$) due to high spring mortality at Sites 15 and 11 with no spring deaths at Site 16. However, season as a main effect proved more significant ($P < 0.0005$) primarily due to the high mortality of the spring treatments compared to the consistently low mortality of the autumn harvest (Table 5). Site was also significant with mortality of 7.3% at Site 15 when compared to 0.5% at Site 16 ($P < 0.05$) (Fig. 3a).

**Table 5**
Effects of harvest regimes (season and frequency) on mallee mortality using 3-years of data for the failed sites, 3-years of data for the successful sites (excluding the three failed $E_{lox}$ sites) and 10-year data. Mortality means of treatments are followed by different letters are significantly different at $P < 0.05$ based on Tukey's HSD test.

| | | 3 Year data failed sites | | | 3 Year data successful sites | | | 10 Year data | | |
|---|---|---|---|---|---|---|---|---|---|---|
| Species | FreOfHarv | Autumn | Spring | Control | Autumn | Spring | Control | Autumn | Spring | Control |
| $E_{koc}$ | Control | | | | | | $1.1^{AB}$ | | | $6.0^{AB}$ |
| | Long | | | | $0.5^{A}$ | $7.1^{BC}$ | | $4.0^{AB}$ | $7.1^{AB}$ | |
| | Short | | | | $0.0^{A}$ | $9.3^{C}$ | | $2.8^{A}$ | $10.9^{B}$ | |
| $E_{lox}$ | Control | | | $5.4^{A}$ | | | $1.8^{A}$ | | | $4.9^{A}$ |
| | Long | $12.7^{AB}$ | $34.6^{BC}$ | | $3.4^{AB}$ | $1.5^{A}$ | | $6.9^{AB}$ | $2.6^{A}$ | |
| | Short | $30.8^{B}$ | $62.3^{C}$ | | $4.7^{B}$ | $1.7^{A}$ | | $8.2^{B}$ | $3.2^{A}$ | |
| $E_{pol}$ | Control | | | | | | $0.7^{NS}$ | | | $2.5^{ns}$ |
| | Long | | | | $1.4^{NS}$ | $2.1^{NS}$ | | $2.7^{ns}$ | $3.1^{ns}$ | |
| | Short | | | | $0.7^{NS}$ | $2.8^{NS}$ | | $1.1^{ns}$ | $2.8^{ns}$ | |

In contrast to the $E_{koc}$ sites, by the end of the study, $E_{lox}$ had very low spring mortality (2.9%), less than half the mortality of the autumn harvest treatments (7.9%) with season being highly significant ($P < 0.001$) (Table 5). The site effect was also significant ($P < 0.05$) ranging from 1.3% at Site 3 to 11.3% at Site 20 (Fig. 3b). Two separate analyses of the mortality data were carried out after 3-year; the three failed sites were analysed separately to reduce the impact of mortality on the successful sites (Fig. 3a). In the first analysis using the failed site data only, season of harvest had a significant effect on mortality in the spring treatments (51.3%) compared to the autumn treatments of (23.3%) ($P < 0.01$). In the second analysis that excluded the failed sites, there was a significant site by season interaction ($P < 0.05$) with mortality at Sites 17 and 20 autumn treatments of 11.2% and 7.5% respectively, compared to Sites 3 and 10 which had mortality of under 1.0%. Season as a main effect was highly significant ($P < 0.01$) with mortality of 4.1% across autumn treatments compared to 1.6% for the spring treatments. The site effect was also highly significant ($P < 0.0001$) mainly due to mortality of under 2% at Sites 3, 9, 10, 11 and 13 compared to 6.5% and 5% at sites 17 and 20 respectively.

$E_{pol}$ had low mortality rate throughout the trial period; neither site nor harvest treatments impacted on its survival.

The annual mortality of coppice and uncut treatments was not uniform. For coppiced mallee treatments, mortality rates peaked the first year after the initial harvest for $E_{lox}$ and $E_{pol}$ at 1.9% and 1.6% respectively whereas $E_{koc}$ mortality rate peaked 2-years after the initial harvest at 2.5% (Fig. 4). There was a weaker trend observed in the uncut treatments with lower mortality rates occurring for each species throughout the trial period.

### 3.2.1. *Annual biomass production*

Annual biomass data was used to analyse all the spring and autumn treatments after 7-years because at many sites the spring treatments were modified. The frequency of harvest of all autumn and uncut treatments over 10 annual measurements were analysed separately.

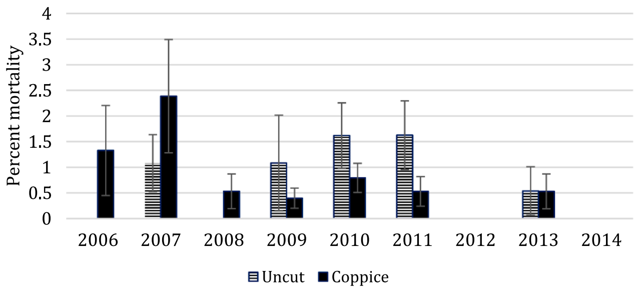
### 3.2.2. *$E_{koc}$ sites*

For the 7-year spring/autumn analysis, there was greater than a three-fold yield difference between the highest and lowest yielding sites ($P < 0.0001$) that remained for the duration of the experiment (Table 6). For 7-year analysis with all treatments, the uncut and autumn treatments significantly outperformed the spring treatments ($P < 0.001$) as seen at Sites 11 and 15. Additionally, the uncut treatment produced more biomass than the coppice treatments after 7-years ($P < 0.005$); however, these differences were not significant after 10-years. There were no differences in annual growth rate between the frequency of harvest treatments at 7- and 10-years, nor were any interactions present.

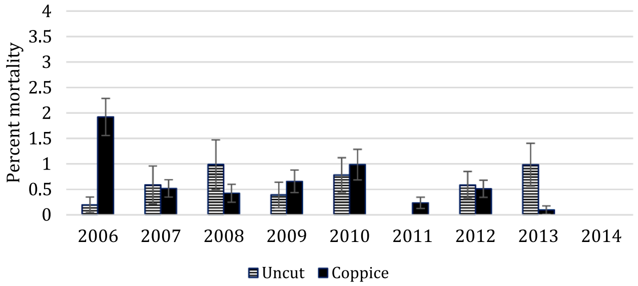
### 3.2.3. *$E_{lox}$ sites*

Similar to the $E_{koc}$ result, site was the most important factor influencing yield ($P < 0.0001$) at the 7- and 10-yr results with a three-fold difference between the highest and lowest yielding sites (Table 7). There was also a significant difference between harvest seasons; however, there was a reversal of the performance of the treatments compared to the $E_{koc}$ sites in that the spring coppice yielded more biomass than the uncut treatments or autumn treatments ($P = 0.005$). This difference was not realised across all sites, with Site 13 showing the opposite trend. There was no significant interaction between site and season. There was no difference between frequency of harvest treatments but there were significant interactions between frequency of harvest and season ($P < 0.005$), in which the autumn treatment yield was higher under the short rotation, whereas the spring treatments benefited from longer harvest cycles. Excluding the failed sites, there was a significant interaction between performance of the harvested and uncut treatments and site ($P < 0.0001$) for both the 7- and 10-year analyses. At five of the eight sites (Sites 2, 3, 9 12 and 20) all coppice treatments outperformed the uncut treatments whereas the reverse was found at Sites 10, 13 and 17. However, the main effect of harvested or uncut treatment was not significant in the 7-year analysis but the coppice treatments yielded more biomass in the 10-year analysis ($P = 0.01$).

(a) E. kochii



(b) E. loxophleba



(c) E. polybractea

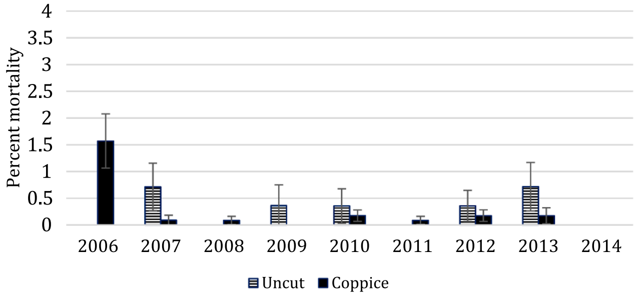


**Fig. 4.** Mortality of all coppice and uncut treatments ( ± s.e.) of three mallee species (a) *E. kochii*, (b) *E. loxophleba* and (c) *E. polybractea* for each year. Only autumn coppice treatments were used after 2012. The three failed $E_{lox}$ sites were excluded from analysis.

### 3.2.4. *$E_{pol}$ sites*

The $E_{pol}$ sites included the highest yielding sites across this study and there was a two- to three-fold difference between site yields ($P < 0.0001$) (Table 8). There was no effect of season and frequency of harvest, but there were interactions between site and the harvested or uncut treatments ($P < 0.001$). Higher yields were observed for all coppice treatments at Sites 8 and 18 and three of the four coppice treatments at Site 1. In contrast, at Site 19 the uncut treatment outperformed the coppice treatments ($P < 0.05$). Across all sites, the main effect of harvesting compared to the uncut treatment was significant ($P = 0.0001$) with more biomass produced following harvesting. For the 10-year analysis, there was no difference between the yield of frequency of harvest treatments but the interaction and main effect that were observed at 7-yr persisted to 10-yr $P < 0.001$ and $P = 0.005$ respectively.

**Table 6**

Annualised increments of total dry biomass yield (Mg ha$^{-1}$ yr$^{-1}$) of $E_{koc}$ for each treatment and harvest cycle. The bracketed numbers are the duration in years of the harvest cycle. Tukey's tests were performed to test difference between site, treatment and site by treatment interaction – means with the same letter do not differ at the 0.05 significance level. Tests for all treatments are given in the 7-year column, and tests for autumn and uncut treatments are given in 10-year column.

| Site | Season | Frequency | 7-year | 10-year | Cycle 1 | Cycle 2 | Cycle 3 |
|---|---|---|---|---|---|---|---|
| 11$^{C}$ | Autumn | Short | 4.8 (6)$^{FG}$ | 4.8 (9)$^{E}$ | 4.5 (7) | 5.6 (2) | |
| | Spring | Short | 3.0 (6)$^{G}$ | | 3.0 (6) | | |
| | Autumn | Long | 4.2 (6)$^{G}$ | 3.7 (9)$^{E}$ | 3.4 (8) | 6.0 (1) | |
| | Spring | Long | 3.5 (6)$^{G}$ | | 3.5 (6) | | |
| | Control | Control | 5.3 (6)$^{EFG}$ | 4.9 (9)$^{DE}$ | | | |
| 15$^{B}$ | Autumn | Short | 9.4 (6)$^{DEF}$ | 10.2 (9)$^{BC}$ | 10.3 (5) | 10.0 (4) | |
| | Spring | Short | 4.6 (6)$^{G}$ | | 5.2 (5) | 1.9 (1) | |
| | Autumn | Long | 9.4 (6)$^{DEF}$ | 8.9 (9)$^{CD}$ | 9.4 (6) | 8.1 (3) | |
| | Spring | Long | 4.8 (6)$^{G}$ | | 4.8 (6) | | |
| | Control | Control | 10.1 (6)$^{CDE}$ | 9.1 (9)$^{C}$ | | | |
| 16$^{A}$ | Autumn | Short | 17.0 (6)$^{AB}$ | 14.9 (9)$^{A}$ | 18.6 (3) | 15.4 (3) | 10.6 (3) |
| | Spring | Short | 13.1 (6)$^{BCD}$ | | 12.3 (3) | 13.8 (3) | |
| | Autumn | Long | 15.7 (6)$^{AB}$ | 14.1 (9)$^{AB}$ | 16.8 (4) | 12.0 (5) | |
| | Spring | Long | 14.3 (6)$^{BC}$ | | 14.4 (4) | 14.1 (2) | |
| | Control | Control | 19.5 (6)$^{A}$ | 15.9 (9)$^{A}$ | | | |

### 3.3. *Yield comparison across multiple cycles*

By the conclusion of this experiment, nine sites had undergone three complete cycles of 3-year autumn harvests and biomass yield was stable except for Site 1, 16 and 18 (Fig. 5). Sites 1 and 18 produced less biomass in the first harvest cycle. Site 16 was the only site that showed progressive decline over the three harvest cycles.

### 3.4. *Relationships of mallee growth to environmental variables*

Stepwise multiple linear regression of biomass on environmental variables identified soil and/or climate factors that accounted for some of the variation in biomass across sites and species. While the growth of uncut controls and coppice treatments were affected by some common factors, coppice growth variation was also influenced by variation in soil nutrients status and climatic factors. For uncut mallee biomass, soil pH, salinity constraints and soil carbon status accounted for over half the biomass variance across sites and species (Eq. (7)):

$$T = 28.16 - 3.37MpH - 0.227MEc + 0.007OC, \quad R^2_{adj} = 0.56 \tag{7}$$

where T is biomass of the uncut control treatment (Mg ha$^{-1}$ yr$^{-1}$), MpH is maximum pH ($CaCl_2$), MEc is maximum soil electrical conductivity ($EC_e$ dS m$^{-1}$) and OC is organic carbon (g/m$^3$) of soil.

For coppice, the variability in biomass across sites and species was explained by a combination of soil and climate constraints (Eq. (8)):

$$C = 162.6 - 4.66MpH - 2.77T_{max} - 0.269MEc + 0.102\ N + 0.007OC \quad R^2_{adj} = 0.72 \tag{8}$$

where C is coppice biomass (Mg ha$^{-1}$ yr$^{-1}$), MpH is maximum site pH ($CaCl_2$), $T_{max}$ is Maximum Temperature (°C), MEc is maximum soil electrical conductivity (dS m$^{-1}$), N is $NO_3^-$ plus $NH_4^+$ (g/m$^3$), and OC is organic carbon (g/m$^3$) of soil.

## 4. Discussion

The data presented here show substantial variation in both biomass yield and mortality across sites, associated with species, climatic and edaphic variables. There is no consistent effect on biomass or mortality from frequency of harvest. However, there was a species response to season of harvest. $E_{koc}$ responded better to autumn harvest, $E_{lox}$ performed better with spring harvest and $E_{pol}$ performed well in each season and displayed low mortality.

### 4.1. *Site effect*

The variation in climatic and edaphic factors across the 19 sites more strongly influenced biomass production than the treatments imposed. The combination of rainfall, evaporation and soil depth has been found to accurately predict biomass production of juvenile mallee plantings (Wildy et al., 2000a). In this study, we found that most biomass yield variance (56–72%) was accounted for by edaphic (pH, $EC_e$, N and organic C) and climate ($T_{max}$) variables. The impacts of pH, $EC_e$ and $T_{max}$ on yield were negative, while soil nutrients (N and OC) had positive effects. Although climate is a key determinant of plantation productivity in the WA wheatbelt by virtue of its impact on landscape water balance (Brooksbank et al., 2011; Robinson et al., 2006; Smettem and Harper, 2009; Sudmeyer et al., 2004; Wildy et al., 2004a), our findings emphasise the importance of localised, edaphic factors controlling water and nutrient availability and uptake by mallee at any given site.

Soil pH was a good predictor of growth for both uncut and coppiced treatments indicating that the studied mallee species prefer acidic soils. pH varied across the sites from 3.6 to 9.1. Symonds et al. (2001) found in a nursery trial that many eucalypt species prefer acidic conditions, including $E_{pol}$ which produced significantly less biomass above a pH of 7.6. The distribution of some endemic eucalypt species or provenances are restricted by soil pH, and perform poorly when grown out of their natural range (Anderson and Ladiges, 1978; Ladiges and Ashton, 1977; Parsons and Specht, 1967). This suggests that pH should be considered in species and site selection, and that agricultural soils too acidic and expensive to ameliorate for other crops may be suitable for mallee.

Another factor that reduced yield across the sites was soil salinity which has been shown, especially in combination with waterlogging, to reduce growth and survival of plants (Barrett-Lennard, 2003). Site 11 exhibited the highest soil salinity of 36.6 dS m$^{-1}$ but had a dry profile, whereas Sites 6, 7, 10, 12, 15, and 17 all had high salinity with shallow saline groundwater (Peck et al., 2012). These root zone soil constraints may have contributed to the high mortality at two of the failed sites (Sites 6 and 7) and to low biomass yield at the other sites. In contrast, Site 1, which was located mid-slope and considered to have access to fresh groundwater at 6–10 m (Peck et al., 2012), had the highest yield. Of the three mallee species, $E_{lox}$ is considered to be moderately salt-tolerant and can survive at $EC_e$ 30.0 dS m$^{-1}$ (Pepper and Craig, 1986). Brooksbank (2011) found that $E_{pol}$ and $E_{koc}$ will not use saline groundwater exceeding 16.1 dS m$^{-1}$ whereas $E_{lox}$ actively sourced groundwater at 30.1 dS m$^{-1}$.

$E_{lox}$ was most commonly planted on lower slopes and valley floors, reflecting its natural occurrence on such sites. However, under agricultural use such sites have been compromised by the accumulation of saline groundwater and as for conventional annual crops, have reduced yield. At Sites 6 and 7 (< 5 m elevation above adjacent saline discharge area) establishment was achieved, but high mortality occurred after first harvest. In contrast, the uncut treatment at Site 7 achieved yields of 17.6 Mg ha$^{-1}$ yr$^{-1}$. These sites had soil salinities of 15.2 and 20.0 dS m$^{-1}$ respectively, suggesting that $E_{lox}$ salt tolerance is reduced as a result of harvesting. There are perennial grazing plant options for salt affected valley floor sites (Barrett-Lennard et al., 2006; Bennett et al., 2009), but these sites may be usefully bounded on lower slopes by plantations of $E_{lox}$ to be harvested on longer cycles. The four $E_{pol}$ sites had relatively deep soil profiles and were only saline at depth (> 6 m) and could be considered relatively unconstrained by soil salinity. Mallee plantings will span multiple soil types and it is inevitable that yield variability will occur.

Nitrogen ($NO_3^-$ and $NH_4^+$) and organic carbon had a positive effect on growth. It was thought that nutrient supply might only slowly become limiting because all sites were within fertilised annual cropping

**Table 7**
Annualised increments of total dry biomass yield (Mg $ha^{-1}$ $yr^{-1}$) of $E_{lox}$ for each treatment and harvest cycle. The bracketed numbers are the duration in years of the harvest cycle. Tukey's tests were performed to test difference between site, treatment and site by treatment interaction – means with the same letter do not differ at the 0.05 significance level. Tests for all treatments are given in the 7-year column, and tests for autumn and uncut treatments are given in 10-year column.

| Site | Season | Frequency | 7-year | 10-year | Cycle 1 | Cycle 2 | Cycle 3 |
|---|---|---|---|---|---|---|---|
| 2$^{C}$ | Autumn | Short | 9.7 (6)$^{BCDEF}$ | 8.6 (9)$^{BC}$ | 9.7 (6) | 6.2 (3) | |
| | Spring | Short | 8.1 (6)$^{CDEFG}$ | | 8.1 (6) | 6.7 (2) | |
| | Autumn | Long | 7.7 (6)$^{FG}$ | 7.5 (9)$^{CD}$ | 7.1 (7) | 8.6 (2) | |
| | Spring | Long | 7.1 (6)$^{FGH}$ | | 6.2 (7) | 4.7 (1) | |
| | Control | Control | 5.6 (6)$^{HIJKL}$ | 5.3 (9)$^{F}$ | | | |
| 3$^{C}$ | Autumn | Short | 7.6 (6)$^{FG}$ | 7.7 (8)$^{CD}$ | 7.7 (3) | 7.5 (3) | 8.0 (2) |
| | Spring | Short | 7.5 (6)$^{FG}$ | | 6.5 (3) | 8.5 (3) | |
| | Autumn | Long | 6.4 (6)$^{GHIJ}$ | 7.1 (8)$^{CDE}$ | 6.7 (4) | 7.4 (4) | |
| | Spring | Long | 8.1 (6)$^{DEFG}$ | | 8.6 (4) | 7.1 (2) | |
| | Control | Control | 4.9 (6)$^{JKML}$ | 5.5 (8)$^{F}$ | | | |
| 9$^{A}$ | Autumn | Short | 10.3 (6)$^{BCD}$ | 10.0 (9)$^{AB}$ | 8.9 (3) | 11.6 (3) | 9.4 (3) |
| | Spring | Short | 12.3 (6)$^{AB}$ | | 8.5 (3) | 16.1 (3) | |
| | Autumn | Long | 10.1 (6)$^{BCDE}$ | 10.3 (9)$^{AB}$ | 9.1 (4) | 11.2 (5) | |
| | Spring | Long | 14.6 (6)$^{A}$ | | 11.2 (4) | 21.4 (2) | |
| | Control | Control | 8.8 (6)$^{CDEF}$ | 7.6 (9)$^{CD}$ | | | |
| 10$^{A}$ | Autumn | Short | 6.4 (6)$^{GHIJ}$ | 6.5 (9)$^{DEF}$ | 5.7 (5) | 7.4 (4) | |
| | Spring | Short | 7.8 (6)$^{EFG}$ | | 6.2 (5) | 15.9 (1) | |
| | Autumn | Long | 8 (6)$^{DEFG}$ | 7.4 (9)$^{CD}$ | 8.0 (6) | 6.1 (3) | |
| | Spring | Long | 10.7 (6)$^{BC}$ | | 10.7 (6) | | |
| | Control | Control | 12.1 (6)$^{AB}$ | 11.0 (9)$^{A}$ | | | |
| 12$^{E}$ | Autumn | Short | 4.2 (6)$^{KLM}$ | 3.9 (9)$^{G}$ | 4.2 (6) | 3.2 (3) | |
| | Spring | Short | 5.1 (6)$^{IJKL}$ | | 5.1 (6) | 3.0 (2) | |
| | Autumn | Long | 4.2 (6)$^{KLM}$ | 3.7 (9)$^{G}$ | 3.7 (7) | 3.7 (2) | |
| | Spring | Long | 4.3 (6)$^{KLM}$ | | 3.1 (7) | 1.7 (1) | |
| | Control | Control | 2.4 (6)$^{N}$ | 2.2 (9)$^{H}$ | | | |
| 13$^{B}$ | Autumn | Short | 7.7 (6)$^{FG}$ | 7.8 (9)$^{CD}$ | 8.4 (3) | 7.0 (3) | 8.0 (3) |
| | Spring | Short | 6.7 (6)$^{GHI}$ | | 6.5 (3) | 6.9 (3) | |
| | Autumn | Long | 6.2 (6)$^{GHIJ}$ | 7.2 (9)$^{CDE}$ | 6.9 (4) | 7.4 (5) | |
| | Spring | Long | 6.6 (6)$^{GHI}$ | | 7.1 (5) | 4.3 (1) | |
| | Control | Control | 10.2 (6)$^{BCDE}$ | 10.7 (9)$^{AB}$ | | | |
| 17$^{B}$ | Autumn | Short | 7.1 (6)$^{JIHK}$ | 7.1 (9)$^{EF}$ | 7.6 (3) | 6.7 (3) | 7.2 (3) |
| | Autumn | Long | 6.8 (6)$^{JKLM}$ | 7.1 (9)$^{F}$ | 7.0 (4) | 7.1 (5) | |
| | Spring | Long | 8.5 (6)$^{GHI}$ | | 8.9 (4) | 7.7 (2) | |
| | Control | Control | 10.1 (6)$^{EFG}$ | 9.1 (9)$^{CDE}$ | | | |
| 20$^{D}$ | Autumn | Short | 4.6 (5)$^{KLM}$ | 6.7 (8)$^{F}$ | 6.7 (6) | 6.9 (2) | |
| | Spring | Short | 4.2 (5)$^{LM}$ | | 5.3 (5) | | |
| | Autumn | Long | 3.9 (5)$^{M}$ | 4.7 (8)$^{G}$ | 4.7 (8) | | |
| | Spring | Long | 5.2 (5)$^{IJKL}$ | | 6.5 (5) | | |
| | Control | Control | 3.7 (5)$^{M}$ | 3.9 (8)$^{G}$ | | | |
| 5 | Autumn | Short | | | 12.6 (4) | 12.4 (4) | |
| | Spring | Short | | | 5.3 (4) | 10.1 (2) | |
| | Autumn | Long | | | 9.8 (5) | 12.6 (4) | |
| | Spring | Long | | | 12.1 (5) | 4.8 (1) | |
| | Control | Control | | | 12.5 (9) | | |
| 6 | Autumn | Short | | | 4.9 (3) | 3.7 (3) | |
| | Spring | Short | | | 0.3 (3) | 0.8 (2) | |
| | Autumn | Long | | | 6.1 (4) | 3.0 (2) | |
| | Spring | Long | | | 4.7 (5) | | |
| | Control | Control | | | 17.6 (9) | | |
| 7 | Autumn | Short | | | 0.3 (2) | | |
| | Spring | Short | | | 0.1 (2) | | |
| | Control | Control | | | 0.7 (5) | | |
| 14 | Autumn | Short | | | 1.1 (3) | | |
| | Spring | Short | | | 0.5 (2) | | |
| | Autumn | Long | | | 0.8 (3) | | |
| | Spring | Long | | | 0.3 (2) | | |
| | Control | Control | | | 3.9 (5) | | |

and sheep-grazed legume-based pastures. However, the level of nutrient removal appears too great especially for N and P, and fertiliser application will be required following harvesting (Grove et al., 2007; Yu et al., 2015). Soil sampling occurred between the first and the second harvests at most sites. Soil organic carbon was positively correlated with biomass production for both the coppice and unharvested treatments. This suggests that concentrations of organic carbon were unaffected by biomass removal. However, the presence of nitrogen in the coppice model suggests removal of biomass may have reduced available soil nitrogen pools. Also, poor inherent site soil fertility may have limited growth from the time of establishment. In 2012 the spring treatments for 10 sites were harvested and then converted to nutrient trials. The addition of 320 kg/ha of ammonium sulphate, 230 kg/ha of double phosphate and 85 kg/ha of nitrate of potash between the rows in

**Table 8**

Annualised increments of total dry biomass yield (Mg ha$^{-1}$ yr$^{-1}$) of E$_{pol}$ for each treatment and harvest cycle. The bracketed numbers are the duration in years of the harvest cycle. Tukey's tests were performed to test difference between site, treatment and site by treatment interaction – means with the same letter do not differ at the 0.05 significance level. Tests for all treatments are given in the 7-year column, and tests for autumn and uncut treatments are given in 10-year column.

| Site | Season | Frequency | 7-year | 10-year | Cycle 1 | Cycle 2 | Cycle 3 |
|---|---|---|---|---|---|---|---|
| 1$^{A}$ | Autumn | Short | 30.4 (6)$^{A}$ | 30.2 (9)$^{A}$ | 22.7 (3) | 38.3 (3) | 29.7 (3) |
| | Spring | Short | 28.2 (6)$^{A}$ | | 14.7 (3) | 41.7 (3) | |
| | Autumn | Long | 29.3 (6)$^{A}$ | 32.8 (9)$^{A}$ | 28.5 (4) | 36.3 (5) | |
| | Spring | Long | 19.8 (6)$^{AB}$ | | 15.8 (4) | 27.9 (2) | |
| | Control | Control | 25.5 (6)$^{A}$ | 23.7 (9)$^{A}$ | | | |
| 8$^{D}$ | Autumn | Short | 10.7 (6)$^{GH}$ | 10.5 (9)$^{DEF}$ | 10.6 (3) | 10.9 (3) | 9.9 (3) |
| | Spring | Short | 10.4 (6)$^{H}$ | | 9.2 (3) | 11.5 (3) | |
| | Autumn | Long | 11.2 (6)$^{FGH}$ | 10.7 (9)$^{DEF}$ | 10.8 (4) | 10.6 (5) | |
| | Spring | Long | 11.7 (6)$^{EFGH}$ | | 11.2 (4) | 12.7 (2) | |
| | Control | Control | 7.1 (6)$^{I}$ | 7.4 (9)$^{G}$ | | | |
| 18$^{B}$ | Autumn | Short | 16.1 (6)$^{CDEF}$ | 16.9 (9)$^{BC}$ | 13.2 (3) | 19.1 (3) | 18.5 (3) |
| | Spring | Short | 16.7 (6)$^{CDE}$ | | 12.3 (3) | 21.2 (3) | 17.0 (2) |
| | Autumn | Long | 17.6 (6)$^{CD}$ | 18.7 (9)$^{B}$ | 14.1 (4) | 22.3 (5) | |
| | Spring | Long | 19.4 (6)$^{BC}$ | | 14.0 (4) | 27.8 (4) | |
| | Control | Control | 16.5 (6)$^{CDEF}$ | 15.6 (9)$^{BCD}$ | | | |
| 19$^{C}$ | Autumn | Short | 12.7 (6)$^{H}$ | 12.1 (9)$^{F}$ | 11.7 (3) | 13.8 (3) | 10.9 (3) |
| | Spring | Short | 14.0 (6)$^{EFGH}$ | | 12.9 (3) | 15.0 (3) | 11.5 (2) |
| | Autumn | Long | 13.2 (6)$^{GH}$ | 11.5 (9)$^{EF}$ | 10.2 (4) | 12.5 (5) | |
| ‘ | Spring | Long | 14.4 (6)$^{DEFGH}$ | | 9.6 (4) | 17.1 (4) | |
| | Control | Control | 15.3 (6)$^{DEFG}$ | 13.9 (9)$^{CDE}$ | | | |

both 2012 and 2013 increased biomass production in 2014 by 11% per annum (Spencer et al., unpublished data). This supports the growth model indication that soil nutrition constrains biomass production and that this might be mitigated with application of fertilizer or biomass ash (Grove et al., 2007).

### 4.2. Frequency of harvest

The short and long frequencies of harvest exhibited high survival and strong coppice vigour with no downward trend in yield after consecutive harvests. Excluding the three failed sites, mortality for the duration of the project, across all treatments, was 4.7%. These results contrast with other studies which reported mortality of up to 100% (Noble and Diggle, 2014) or progressive declines in productivity (Milthorpe et al., 1998; Wildy and Pate, 2002) under more frequent harvest regimes. In our study, coppice mortality following the initial harvests in 2006 and 2007 accounted for 46% of overall mortality. The peak mortality at first harvest appears to be induced by the harvest, because mortality rates in the uncut treatments remained steady. Harvest mortality was mostly confined to smaller trees, and appears to have had little effect on biomass yield. In 2010, there was a slight increase in mortality where many sites received about half their average annual rainfall.

It has been suggested that carbohydrate root reserves in WA flora have an important role in the capacity to resprout (Bell et al., 1996). Wildy and Pate (2002) found that with removal of foliage from *E kochii* every three months, starch was a poor predictor of subsequent biomass production and mortality. They also found the reduction of starch reserves persisted for 12–18 months after harvest. Hence to minimise mortality risk, a minimum 3-year harvest interval was chosen in this study. It was anticipated that this would also deliver sufficient biomass to facilitate development of low cost, high volume harvest systems (Abadi et al., 2012). No trend was observed within sites to suggest a penalty in annual yield under the shorter harvest frequency.

Excluding the three failed sites, the frequency of harvest resulted in sustainable biomass production, at higher yielding sites. Site 1 displayed large variation between cycles peaking at cycle two with 38.3 Mg ha$^{-1}$ yr$^{-1}$ (dry weight) with lower yields at cycles one and

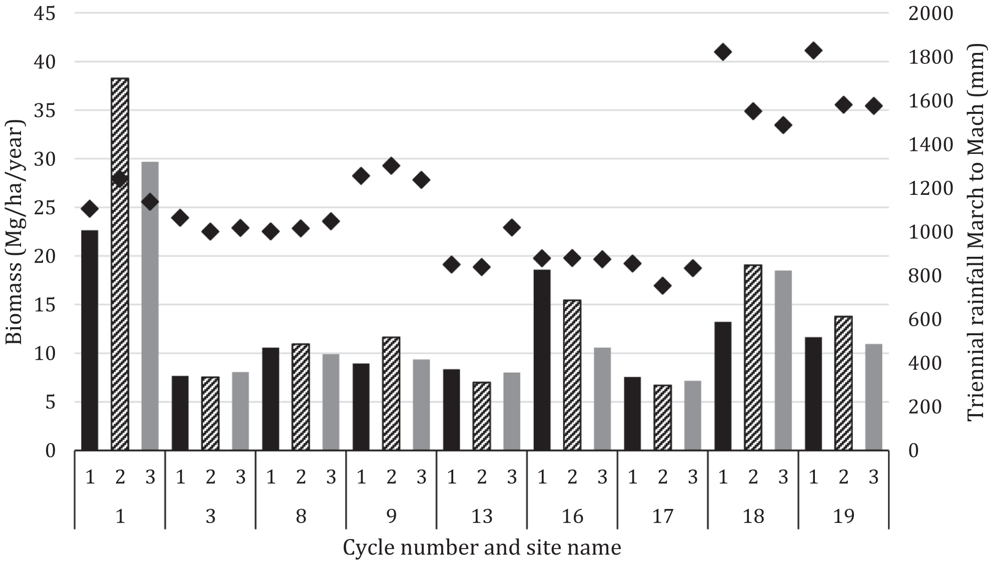


**Fig. 5.** Annualised dry biomass yield (Mg ha$^{-1}$ year$^{-1}$) for all sites under three 3-year harvest cycle regimes. Solid black bars represent cycle one, pattern bars cycle two and grey bars cycle 3. Total rainfall for each 3-year cycle from autumn to autumn (March to March) is represent by diamonds. Site 3 was removed in 2014 and the last 3-year rotation was limited to 2-years of data.

three. Site 16 showed progressive declines in yield for the 3-year autumn harvest coppice treatment from 18.6 to 10.6 Mg ha$^{-1}$ yr$^{-1}$. There was no progressive decline in rainfall over this period and no other site displayed a declining yield trend (Fig. 5). Site 16 is just upslope from the boundary between the sandplain landform and the in-situ weathered profiles over the igneous basement. These junctions are often the site of discharge from perched sandplain aquifers, that are readily depleted with localised tree planting (George, 1990). Investigations of this site reported in Bennett et al. (2005) and Pracilio et al. (2006) support the conclusion that the sandplain aquifer had been exhausted by the extensive mallee planting of which Site 16 was part. These sites would be optimally managed by matching plantation area to the discharge volume of the aquifer.

There was a species response to harvest with $E_{koc}$ seemingly the least tolerant, especially following spring harvests. At each $E_{koc}$ site, the uncut treatments yielded more biomass than the coppice treatments. In contrast, most $E_{pol}$ and $E_{lox}$ sites yielded more biomass under coppice regimes, however, this was not consistent between sites. The juxtaposition of an $E_{pol}$ belt adjacent to Site 16, an $E_{koc}$ belt, provided an opportunity to observe the consequence of the drying out of the shallow aquifer on two different mallee species (Bennett et al., 2005; Pracilio et al., 2006). While $E_{koc}$ showed progressive decline in harvest yield, $E_{pol}$ suffered extensive drought death. This vulnerability of $E_{pol}$ to the hotter/drier climate of the northern wheatbelt (evaporation > 2500 mm and rainfall < 350 mm) has been widely observed in farmer plantings. $E_{pol}$ is native to central Victoria and southern NSW (evaporation 1600 mm and rainfall 480 mm) and in the absence of sufficient water it is prone to mortality (J. Bartle, D. Huxtable and B. Spencer personal observations). In contrast, $E_{koc}$ is a native species in this region and although it showed a decline in biomass at Site 16, no widespread death was recorded. When comparing the two species drought responses, $E_{pol}$ increases root growth thereby capturing more annual rainfall whereas $E_{koc}$ reduces leaf area (Brooksbank, 2011). These adaptations seem to favour $E_{koc}$ in the northern wheatbelt, where this species can survive without groundwater supply but increase growth rate by ten-fold when additional groundwater is available (Brooksbank et al., 2011). However, without additional ground water, the zone of transition for $E_{pol}$ selection lies where the aridity index (ratio of annual rainfall to annual evaporation) is about 0.2. Similar contrasting drought responses and growth rates have been found in other tree species in this region (Mitchell et al., 2013).

### 4.3. Season of harvest

Large-scale markets for mallee biomass will require nearly continuous supply of biomass to minimise storage costs (Abadi et al., 2012); hence harvest may need to occur at any time of the year. The seasonal rainfall and temperature cycles in the south west of WA are likely to influence the response of mallee crops to harvest. It has been shown that below ground carbohydrate reserves in unharvested mallee are lower in summer than spring (Wildy and Pate, 2002) and lower survival rates are observed with autumn harvests (Milthorpe et al., 1994; Noble, 1982; Noble and Diggle, 2014). Thus, it was hypothesised that the autumn harvest would produce less biomass and higher mortality than spring harvest. Our results showed no consistent adverse impacts of autumn harvest, refuting our hypothesis. Our study had longer harvest intervals than the works cited above and the 3-year harvest cycle appears to avoid depleting stand vigour. However, this study found differential species responses to season of harvest. $E_{koc}$ sites yielded more biomass with autumn treatments and this could be attributed to shorter rainfall period in the northern wheatbelt where all the $E_{koc}$ sites were located. In contrast, the $E_{lox}$ sites produced more biomass with spring harvests. The failed $E_{lox}$ sites (6, 7 and 14) did produce less biomass with higher mortality following spring harvests compared to the uncompromised trials. These sites were all in the northern wheatbelt where only one $E_{lox}$ site survived (Site 17). More research is needed to establish the northern range of $E_{lox}$ for production of biomass, especially on sub-optimal sites. There was no seasonal effect of biomass production at the $E_{pol}$ sites.

Without the stress of harvest, it was expected that uncut treatments would exhibit higher survival and this was generally observed. With respect to harvested mallee, our results indicate that season had a greater effect on mortality than frequency of harvest; $E_{koc}$ sites exhibited higher spring losses whereas greater losses of $E_{lox}$ occurred in autumn, consistent with previous studies (Noble, 1982; Noble and Diggle, 2014). Native herbivorous spring beetles (*Liparetrus* spp.) were observed at sites in the north-eastern wheatbelt, where all of the $E_{koc}$ sites were located, and herbivory of emergent coppice foliage may have contributed to higher spring mortality. The four $E_{pol}$ sites exhibited low mortality whereas Milthorpe et al. (1994) found elevated mortality from of $E_{pol}$ from autumn harvests but this was attributed mainly to waterlogging after a flood event. It appears that mortality reflected seasonal factors, especially at a species-level.

### 4.4. Biomass yield

Caution should be taken when directly comparing the biomass yield between sites with different planting configurations. The mallee plantings selected for this experiment were chosen from established plantings reflecting the landholder's site selection, planting design and management. Selection focussed on stands that were seen as adequately representative of overall performance and fell within the narrow-belt specification of planting row number and within-row spacing. It was necessary to estimate actual yield of standing biomass for belts ranging from 2 to 6 planting rows. When comparing the yield between 2-row belt and belts with more than 2-rows, Mendham et al. (2012) and Peck et al. (2012) doubled the yield of the outer row of sites containing more than 2-rows because the inner rows had on average only 65–70% the biomass of outer rows. However, subsequent spacing trial data suggests that the outer row biomass from treatments containing more than 2-rows have about 90% of the biomass of a 2-row belt (B. Spencer *et al.*, unpublished data). Nevertheless, a tree in a 2-row belt will have greater access to water and nutrient resources and will not be subjected to the additional competition imposed by sites containing more than 2-rows. Hence the biomass yield estimates for 2-row belts will be inflated when compared to sites of more than 2-rows.

In this study, biomass is reported as dry weights, with fresh mallee biomass, depending on size and site, varying between 36% and 48% in water content. Most mallee studies report fresh biomass, making direct comparisons difficult. Mallee growth rates, reported as dry mass or adjusted to 40% moisture, are in the range of 1–25 Mg ha$^{-1}$ yr$^{-1}$ (Bennett et al., 2015; Brooksbank, 2012; Carter and White, 2009; Grove et al., 2007; Pracilio et al., 2006; Sudmeyer and Hall, 2015). For most of our sites, the growth rates were within the above range, although at some sites considerably higher rates were observed. These sites may reflect the age of plantings. The project was initiated in 2006 on established plantings that were then between five and 12-years of age and all mallee plantings were older than any of those referred to above.

This study confirms that $E_{pol}$ is capable of high biomass production in the high rainfall/low evaporation south-western wheatbelt region. $E_{lox}$ is well adapted to the central and eastern wheatbelt on mid to lower slopes, but three sites failed due to the presence of either silcrete hardpan or shallow saline groundwater. $E_{koc}$ sites were restricted to upper slope sandplain soils in the northern wheatbelt, but can deliver reasonable yields (3–16 Mg ha$^{-1}$ yr$^{-1}$), although only one study site was subjected to the 3- and 4-year harvest regime. It is important to match species to site types as they have different tolerances to salinity and water requirements (Brooksbank, 2011). There is potential for siting plantations of $E_{lox}$ on the transition zone from saline valley floor to lower slopes and for $E_{koc}$ on sand plain seepage areas in the central and northern wheatbelt regions. Productivity of all three species is constrained by shallow saline groundwater and by the scattered occurrence of silcrete hardpan.

### 4.5. Capacity to coppice

Mortality at harvest has been reported for other species of eucalypts grown for biomass. The 4.7% mortality in this project compares to other studies where 99% of *E. camaldulensis* and *E. dunnii* coppiced within 1-year post-harvest (Grunwald and Karschon, 1974; Li et al., 2012). In contrast, mortality over 25% has been recorded for *E. globulus* at 15 months post-harvest (Whittock et al., 2003) and *E. grandis* 20 months post-harvest (Little and Toit, 2003). However, these mallee data were for multiple harvest cycles over a longer time period than the other studies.

The effect of genetic variation in coppicing has not been reported for these three mallee species. However, there is evidence from other eucalypts that it may be important. Whittock et al. (2003) for instance, found significant variation between the coppicing of sub-races of *E. globulus* post-harvest and Borzak et al. (2016) found, for the same species, that larger lignotubers exhibited lower mortality and faster growth. Our results indicate a spike in coppice mortality after the first harvest that did not occur in subsequent harvests. Establishing the heritability of coppicing vigour of families or sub-races could potentially reduce post-harvest mortality and increase production.

### 4.6. Economic considerations of harvest

There are three economic questions that need to be addressed before harvesting: the market for biomass products; the cost of harvest; and the competition imposed by mallee belts on adjacent crops and pasture. The components of biomass (leaf, twig, bark, wood) change with the species and the size of the uncut or coppiced mallee, providing scope to manipulate biomass composition by varying the frequency of harvest. The component allometric regressions demonstrate more frequent harvests increase the percentage of leaf while reducing the wood fraction. Depending on prices for the components, and available markets for biofuels and extractives such as cineole (Davis, 2002), manipulating frequency of harvest could be important for economic optimisation. There are boundaries to harvest frequency due to mortality risk if too frequent and harvester capability if too infrequent. Cycle duration will influence the competition that mallee belts impose on adjacent crop or pasture. Sudmeyer et al. (2012a) demonstrated greater competition on adjacent annual crops occurs with larger mallee, and that competition is reduced after harvest for up to five years. However, the ultimate assessment of economic viability will need to include a holistic assessment of all on- and off-farm benefits that have been a major motivation for development of a woody perennial crop (Bartle and Abadi, 2009).

### 4.7. Conclusion

This study reports that dry weight biomass yields ranged from 2.2 to 32.8 $\mathrm{Mg\,ha^{-1}\,yr^{-1}}$. All three species planted at sites not limited by saline water or silcrete hardpan consistently produced woody biomass that could be used for a range of products and services. The frequencies of harvest imposed did not compromise biomass production and except for three sites, mortality was generally low and independent of frequency of harvest. There was a species response to season of harvest where $E_{lox}$ performed better following spring harvests whereas $E_{koc}$ had higher mortality with lower yields. $E_{pol}$ was unaffected by season and frequency of harvest imposed.

This study confirms that $E_{pol}$ is a high biomass producer provided it is planted in higher rainfall and lower evaporation regions. $E_{lox}$ is an adaptable species and can tolerate moderate salinity, but planting too close to saline water tables reduces productivity and may result in high mortality after harvest. $E_{koc}$ sites were restricted to sandplain soils in the northern wheatbelt, but can deliver reasonable yields (3–16 $\mathrm{Mg\,ha^{-1}\,yr^{-1}}$), although only one study site was subjected to the 3- and 4-year harvest regime. Mallee belt plantings will span multiple soil types and it is inevitable that yield variability will occur. It is important to match species to site types as they have different tolerances. Frequency of harvest will affect biomass component composition and hence utility and value. Soil factors including pH, $EC_e$, shallow saline water tables and soil nutrition were found to strongly influence biomass production. These results have implications for initial site selection for mallee plantings and management decisions regarding timing of harvest.

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## Appendix A. Supplementary material

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# Chapter 3

## Number of rows and within-row spacing affects biomass yield, mallee form and size

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# Planting configuration affects productivity, tree form and survival of mallee eucalypt in farm forestry plantings

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**Abstract** Mallee eucalypts have been extensively planted in the Western Australia wheatbelt for salinity mitigation and as a short-rotation coppice crop for the production of cineole and bioenergy feedstocks. Mallee has been planted in wide-spaced narrow belts (2–6 rows) within annual crops and pasture, but optimal planting configurations have not been determined. Here, we assess the biomass yield responses of *Eucalyptus loxophleba* ssp. *lissophloia* and *E. polybractea* to; four row treatments (1, 2, 4 and 6 row belts) and five within-row spacing treatments (1, 1.5, 2, 3 and 4 m). Thirteen years after planting, the row effects on plot-level biomass productivity of *E. loxophleba* ranged from 4.3 to 21.2 Mg ha$^{-1}$ year$^{-1}$. For *E. polybractea*, both row number and within-row spacing affected yield, which ranged from 2.7 to 18.8 Mg ha$^{-1}$ year$^{-1}$. For both species, the highest growth rates were observed in the one-row belts with shorter (< 3 m) within-row spacing. Within the belts, reductions of growth rate were observed with additional rows, due to increased competition and significant suppression of internal rows; and with wider within-row spacing, due to lower initial planting density. However, when including the area between belts, wider belts generated more biomass. For both species, average tree size decreased with additional rows and shorter within-row spacing. For both species, the number of stems per tree increased with wider within-row spacing, and also for *E. polybractea,* with fewer rows. The substantial variation in productivity, tree size and form found in these results will affect harvestability and ultimately the economic viability of future mallee plantings.

**Keywords** Agroforestry · Alley farming · Tree belt design · Bioenergy crops · Oil mallee · Spacing trial

## Introduction

Over the last three decades research has been undertaken to develop woody perennial crops to complement annual crops and pastures in the Western Australian (WA) wheatbelt. Economically viable perennial crops could help mitigate dryland salinity (Olsen et al. 2004; Bartle et al. 2007; Bartle and Abadi 2010). Lefroy and Stirzaker (1999) examined tree crop planting options for salinity management and concluded that integrated plantings would be preferred to segregated or rotated tree crop systems. In this case integrated plantings would take the form of wide-

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spaced narrow belts within the existing annual crop/pasture farming system.

Mallee eucalypts (hereafter referred to as mallee) are small, multi-stemmed lignotuberous trees. Mallee were selected as the most prospective woody perennials for crop development due to their ability to coppice after regular, short-cycle harvest. Some 300 native mallee species occur across the inland, lower annual rainfall (200–500 mm) regions of the southern states of Australia (Nicolle 2006). Mallee attracted commercial interest from the early years of European settlement in Australia as a source of eucalyptus oil (extracted from the leaf by steam distillation). Species with leaf oil consisting predominantly of 1,8-cineole (hereafter referred to as cineole) were particularly favoured (Davis 2002). There are a few current operations in Australia extracting eucalyptus oil from mallee species, from both native and cultivated stands, on coppice harvest cycles of 1–5 years. Historic markets for cineole focussed on non-prescription medical uses but recent work has shown promise for industrial scale use (Barton and Tjandra 1989; Davis 2002; Soh and Stachowiak 2002; Leita et al. 2010). High total oil, cineole-rich mallee species have been selected to suit the full range of edaphic and climatic conditions in the WA wheatbelt. Two of these are the subject of this work, *Eucalyptus polybractea* R.T Baker, native to New South Wales and Victoria, and *Eucalyptus loxophleba* Benth. subsp. *lissophloia* LAS Johnson & KD Hill, from WA. Both of these species readily coppice after harvest (Eastham et al. 1993; Wildy et al. 2000a; Spencer et al. 2019). Recent interest in carbon sequestration by agroforestry systems to combat climate change (Harrison and Gassner 2020) gave strong impetus to develop mallee for its carbon offset and bioenergy potential (Wu et al. 2008; Abadi et al. 2012; Yu et al. 2015). Biofuels became a major research area with a particular focus on conversion to fuels by pyrolysis (O'Connell et al. 2007; Garcia-Perez et al. 2008; Wu et al. 2009; McGrath et al. 2017).

Integration of mallee into the wheatbelt farms has potential direct commercial returns. Other on-farm and regional benefits may also be substantial: hydrological control reducing salinity and waterlogging (Rundle and Rundle 2002; Silberstein et al. 2002; Ellis et al. 2006; Robinson et al. 2006); stock shelter and wind erosion control (Bird et al. 1992; Sudmeyer and Scott 2002a, b; Baker et al. 2018) and biodiversity benefits (Smith 2009). However, mallee have extensive root systems and while their deep root penetration is beneficial (Nulsen et al. 1986; Robinson et al. 2006), their lateral roots spread well beyond the planted belts, creating a wide competition zone with the adjacent annual crops and pastures (Sudmeyer et al. 2012). Economic analyses have been undertaken to help define the full range of costs and benefits (Cooper et al. 2006; Abadi et al. 2012).

The number of rows in a belt, plant spacing within the rows, and harvest frequency will all affect biomass yield and composition. In agroforestry plantings, shorter within-row spacing leads to smaller trees but greater yield (Karim and Savill 1991; Dagar et al. 2016). This within-row tree spacing effect has also been demonstrated in plantation forestry prior to canopy closure (Niemistö 1995; DeBell and Harrington 2002; Pinkard and Neilsen 2003; West and Smith 2019). The most common planting configuration for mallee has been 4-row belts, 40–100 m apart, with 2 m between rows and 1.5 m within-row spacing (URS 2008; Bartle 2009). The area between belts is commonly called the alley. A study across eight sites in WA with more than 2-rows, found yield reduction in internal rows of 60% for unharvested belts; and for harvested belts inner row suppression of up to 80% (Huxtable et al. 2012). Evidence that 1- or 2-row belts may better utilise the land occupied indicates the need to better define the yield characteristics of these narrow belts (Prasad et al. 2010; Paula et al. 2013).

This study presents the results of two mallee spacing experiments consisting of four different numbers of rows, and five within-row spacing treatments. The aim is to determine:

(1) The planting configuration that maximises mallee productivity by testing total biomass response to planting configurations; and
(2) The effect of planting configuration on survival and tree form.

## Methodology

### Study site and species

The experiments were established at two sites north of the town of Narrogin (32.93°S, 117.18°E, altitude 290–310 m) in the Western Australian wheatbelt. The wheatbelt has mild wet winters and hot dry summers. Annual average rainfall (1986–2015) for Narrogin was 447 mm, annual evaporation 1566 mm, average daily maximum temperature was 22.7 °C and average daily minimum temperature was 9.8 °C (Jeffrey et al. 2001).

The experimental sites were selected considering suitability of soil types to the two selected mallee species: *E. polybractea* and *E. loxophleba* subsp. *lissophloia*, which are widely planted in the Western Australian wheat belt. These species will be hereafter referred to as $E_{pol}$ and $E_{lox}$ respectively. Each site consisted of only one of the two species. Both sites have similar landscape position and soil type, i.e. shallow valley floor landform in the Eastern Darling Range Zone and depositional profiles having duplex soils with deep grey sandy surface soil horizon to 1 m over sandy clay (Moore 2001). Both sites were cleared of native vegetation several decades ago and converted to an agriculture based on well-fertilised annual crop/pasture rotations.

Both experiments were established in winter 2000. Prior to planting, weed control was carried out using glyphosate and simazine. Seedlings were planted into soil that had been ripped to a depth of 50 cm and riplines were 2 m apart.

## Experimental design

Both experiments had a split plot design with four replicates and random allocation of main plots within each replicate, and sub-plots within the main plots (Fig. 1). The belt row configuration was the main plot treatment with four levels: 1, 2, 4 or 6 row belts. The distance between rows was maintained at 2 m as this is the minimum spacing required for a single row harvester to access internal rows. The main plots were divided into five within-row spacing treatments of 1, 1.5, 2, 3 or 4 m. At each main plot boundary there was a six-tree buffer while there was a three-tree buffer between the sub-plots. The larger buffer was used between the main plots as it represented a change in both tree-spacing and number of row treatments. Each sub-plot consisted of 12 trees distributed between the number of rows prescribed.

Two analyses were performed: firstly, to compare the productivity of each treatment on the area the mallee plots physically occupy; and secondly, to compare the productivity of each treatment including the alley area to determine mallee productivity of the entire paddock. These two approaches were used as both have limitations, the first analysis does not account for the area of influence the mallee belt has on the immediately adjacent agricultural land (called the competition zone) and the second approach does not account for the additional area foregone to agriculture that the wider belts occupy.

In the first analysis, to standardise the plot area of each treatment, the outer edge of the plot was calculated as half the internal distance between rows, as used by Paul et al. (2013a). Hence, the 2 m inter-row space had 1 m added to each side to derive plot area. The 1-row treatment was also allocated a 1 m edge to derive area. Consequently, the 1-row treatment is twice the length and half the width of the 2-row treatment; analogously the 1-row treatment was six times the length and one sixth the width of the 6-row treatment. This method allocates equivalent plot areas to different row treatments with the same spacing treatment. For instance, for a 1 m within-row spacing, the 1-row belt of 12 trees has a plot area of 24 $m^2$, 12 trees × 2 $m^2$ (1 $m^2$ each side of the belt) and the 6-row belt at 1 m within-row spacing also has a plot area of 24 $m^2$ (2 trees along the belt × 2 m between row × 5 internal rows plus 2 external tree × 2 rows × 1 $m^2$ for the external edge). However, plot area is modified by the within-row spacing treatments (Table 1).

In the second analysis, the alley area was included to calculate mallee productivity over the entire paddock. Alley widths at both experiments were approximately 50 m apart. The plot area, for instance, for a 1 m within-row spacing, the 1-row belt of 12 trees has a plot area of 0.06 ha (12 m × 50 m) whereas the 6-row belt at the same spacing has a plot area of 0.01 ha (2 m × 50 m) (Table 1).

## Estimating dry mass of trees

Diameters of each stem were measured in the winter of 2013 with a diameter tape at approximately 10 cm above ground level. All stems over 10 mm were measured. Fibrous bark, buttressing and swelling associated with low branching was avoided by slightly raising or lowering the measurement height. For multiple stemmed trees, the Equivalent Diameter (EDRC) method of Chojnacky and Milton (2008) was used to provide a single diameter:

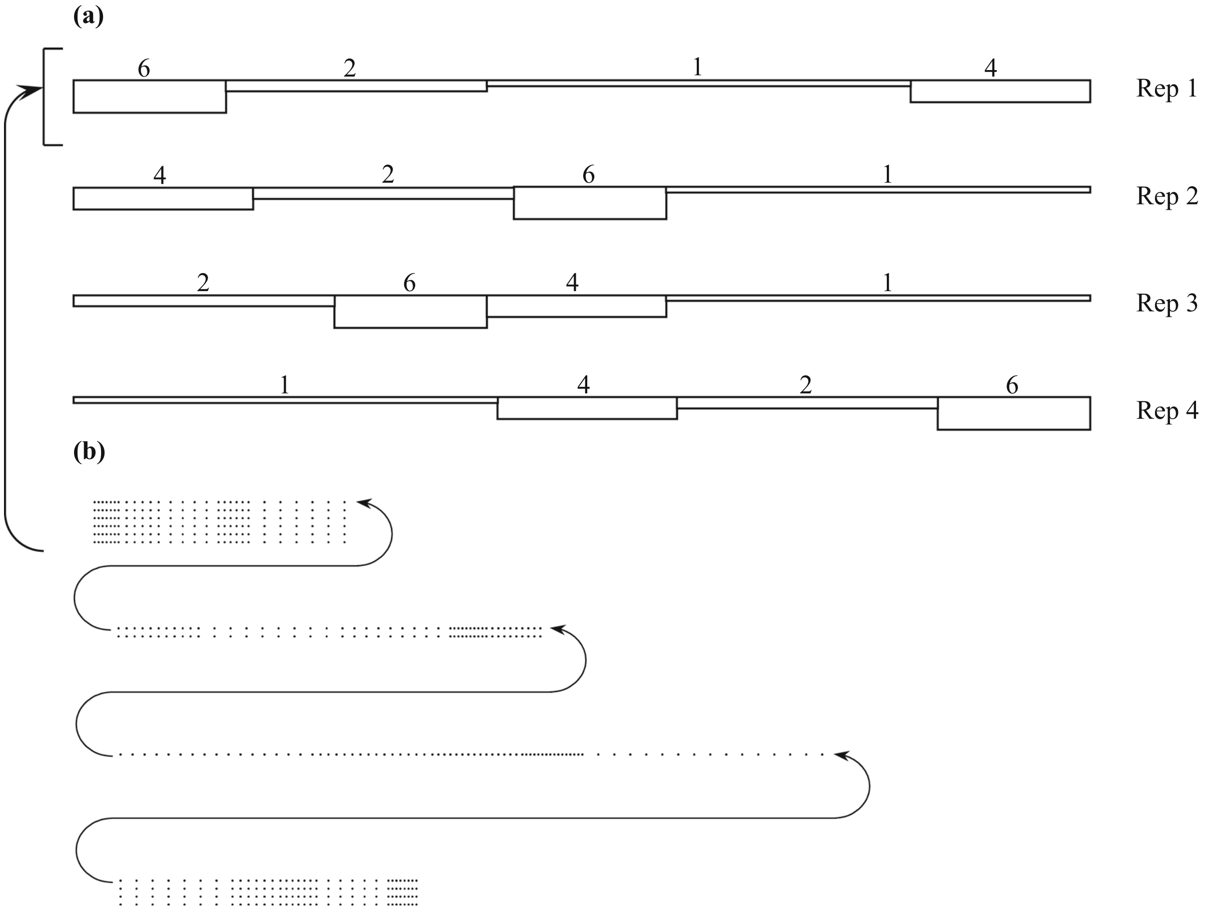

**Fig. 1** A schematic of the trial design and planting configuration: **a** fully randomised allocation of main plot treatments (number of rows) within each replicate, and **b** further randomised arrangement of the subplot (within-row spacing) treatments within the whole plot factor using replicate 1 as an example

**Table 1** Plot areas and stocking density (trees ha$^{-1}$) of within-row spacing treatments (m) for the plot-level scenario and the plot areas for each row-treatment and within-row spacing for the paddock-level scenario

| Within row spacing (m) | Plot-level scenario | | Paddock-level scenario | | | |
|---|---|---|---|---|---|---|
| | Plot area (ha) | Trees ha$^{-1}$ | Row treatment and plot area (ha) | | | |
| | | | 1-row belt | 2-row belt | 4-row belt | 6-row belt |
| 1 | 0.0024 | 5000 | 0.060 | 0.030 | 0.015 | 0.010 |
| 1.5 | 0.0036 | 3333 | 0.090 | 0.045 | 0.023 | 0.015 |
| 2 | 0.0048 | 2500 | 0.120 | 0.060 | 0.030 | 0.020 |
| 3 | 0.0072 | 1667 | 0.180 | 0.090 | 0.045 | 0.030 |
| 4 | 0.0096 | 1250 | 0.240 | 0.120 | 0.060 | 0.040 |

The numbers refer to each replicate at the two experimental sites. Note that for the plot-level scenario, at a given within-row spacing, the plot area is the same for all four different row spacing treatments; see text in the Methods section for details

$$EDRC = \sqrt{\sum_{i=1}^{n} \mathrm{drc}_i^2} \qquad (1)$$

where *drc* equals the diameter of each stem and *n* equals the number of stems of each tree.

Mallee allometric equations developed by Spencer et al. (2019) were used to estimate dry biomass in a two-step process; first converting EDRC to above ground fresh biomass, then partitioning fresh biomass into oven dry wood, bark, twig and leaf. These data were then summed to estimate the dry biomass of the tree which was used to calculate standing dry biomass for each treatment and plot- and paddock-level scenarios. Other mallee eucalypt allometric equations were assessed; these include Paul et al. (2013b) which did not cover suitable size range for stem diameter, while the continental-scale multi-stemmed equation published Paul et al. (2016) underestimated biomass when compared to the species-specific equations generated by Spencer et al. (2019).

### Statistical model

Treatment effects were evaluated by sites using mixed linear models using REML to estimate variance components in SAS 9.4 (SAS 2017) with the following formula:

$$\mathrm{y}_{ijk} = \mathrm{r}_i + \mathrm{o}_j + \mathrm{s}_k + \mathrm{o}_j \mathrm{x}\ \mathrm{s}_k + \mathrm{o}_j(\mathrm{r}_i) + \mathrm{e}_{ijk} \qquad (2)$$

where y is the trait of interest (dry biomass $\mathrm{ha}^{-1}$, number of stems or survival), $\mathrm{r}_i$ is the replicate effect, $\mathrm{o}_j$ is the row treatment, $\mathrm{s}_k$ is the spacing treatment, $\mathrm{o}_j$ x $\mathrm{s}_k$ is the interaction between the row and the spacing treatments, and $\mathrm{e}_{ijk}$ is the residual error. Replicate and replicate nested with the main plot (row treatment) were specified as random effects. The proportion of trees that survived were analysed following arcsine transformation. Tree biomass was natural-log transformed to reduce heteroscedasticity and heterogeneity of variance. Prior to measurement, a fire had burnt one replicate of the 6-row treatment at the $\mathrm{E}_{\mathrm{pol}}$ site. Additionally, at the $\mathrm{E}_{\mathrm{lox}}$ site, two subplots (4 and 3 m spacing) of one replicate of the 1-row treatment had high mortality and the remaining trees had been damaged by termites modifying the growth form of the trees. The burnt and termite affected plot data were treated as missing observations in analysis.

## Results

### Planting configuration on mallee survival

Tree survival, averaged across treatments, was 86% (range: 69–94%) at the $\mathrm{E}_{\mathrm{lox}}$ site while at the $\mathrm{E}_{\mathrm{pol}}$ site it was 89% (range: 78–96%) (Fig. 2a). Significant differences in survival were observed at the $\mathrm{E}_{\mathrm{lox}}$ site ($p < 0.05$) for the row treatments (Table 2), where there was 78% survival for the 1-row belts compared to above 86% for the other row treatments. No differences in survival were observed between treatments at the $\mathrm{E}_{\mathrm{pol}}$ site.

### Planting configuration affects productivity of mallee in agroforestry systems

For the plot-level scenario, across both experiments, the 1-row treatment produced significantly more biomass per unit area than the other row-treatments (Fig. 2b). Table 2 summarises the significance of the main- and sub-plot results at both sites. The number of row treatment had a highly significant effect on biomass production which ranged from 4.3–21.2 Mg $\mathrm{ha}^{-1}$ $\mathrm{year}^{-1}$ at the $\mathrm{E}_{\mathrm{lox}}$ site ($p < 0.0001$) and 2.7–18.8 Mg $\mathrm{ha}^{-1}$ $\mathrm{year}^{-1}$ at the $\mathrm{E}_{\mathrm{pol}}$ site ($p < 0.001$). There was a yield reduction with additional rows, with the highest yielding 1-row treatment producing more than twice the biomass of any of the 4 and 6-row treatments. The within-row spacing treatment was also highly significant at the $\mathrm{E}_{\mathrm{pol}}$ site ($p < 0.0001$) where the 1 m within-row spacing yield exceeded the other within-row spacing treatments. Although not significant, a similar trend was observed at the $\mathrm{E}_{\mathrm{lox}}$ site except for the 1-row treatment. Across both sites and most row-treatments, the 3 and 4 m within-row spacing treatments consistently produced the least biomass. The interaction between row treatment and within-row spacing was not significant (Table 2).

For the paddock-level scenario, productivity ranged from 0.65–1.56 Mg $\mathrm{ha}^{-1}$ $\mathrm{year}^{-1}$ at the $\mathrm{E}_{\mathrm{lox}}$ site and from 0.43–1.86 Mg $\mathrm{ha}^{-1}$ $\mathrm{year}^{-1}$ at the $\mathrm{E}_{\mathrm{pol}}$ site with most biomass being generated at the wide belts (4- or 6-rows) with short within-row spacing (Fig. 2c). These wider belts produced significantly more biomass ($p < 0.01$) than the 1- or 2-row treatments, with the 6-row belt, averaged across within-row spacing treatments, producing almost double the biomass of

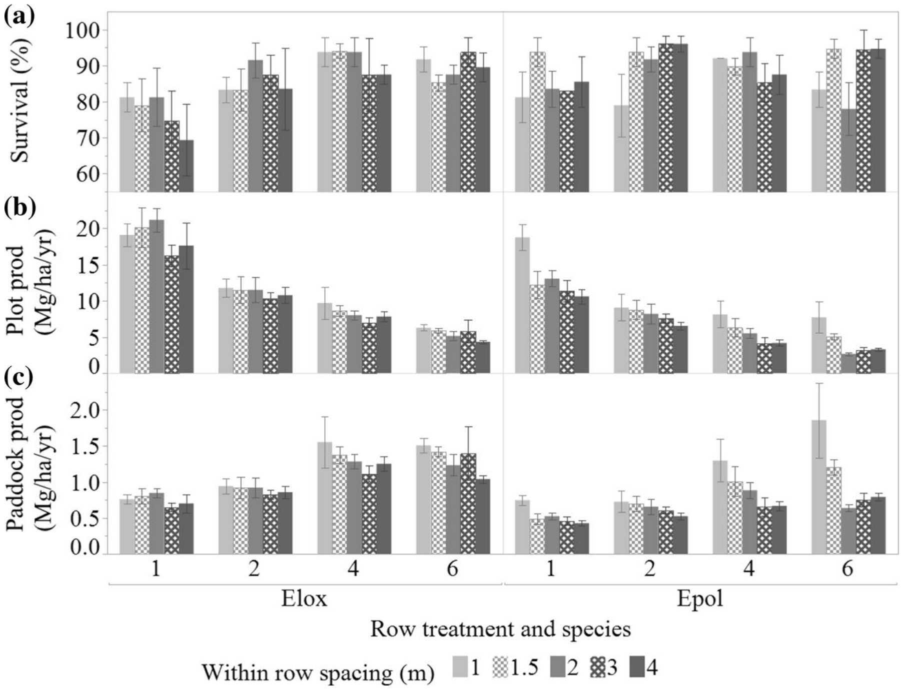

**Fig. 2** Effect of number of rows (1, 2, 4 and 6) and within-row spacing treatments on: **a** mallee survival; **b** plot-level productivity (Plot prod Mg $ha^{-1}$ $year^{-1}$) which includes only the area occupied by mallee; and **c** paddock-level productivity (Paddock prod Mg $ha^{-1}$ $year^{-1}$) which includes the alley area between mallee belts. All graphics refer to the *Eucalyptus loxophleba* subsp. *lissophloia* ($E_{lox}$) and *E. polybractea* ($E_{pol}$) sites near Narrogin, Western Australia. Error bars represent ± one standard error (n = 3–4)

the 1-row belt at both sites. The within-row spacing treatments were highly significant at the $E_{pol}$ site ($p < 0.0001$) where, averaged across row-treatments, the 1 m within-row spacing belt yielded nearly twice the biomass compared to the 3 and 4 m within-row treatments. Analogous to the plot-level analysis, a similar trend occurred at the $E_{lox}$ site, but was not significant. The interaction between row treatment and within-row spacing was also not significant.

The biomass production of the 4- and 6-row treatments were further analysed and there was a difference ($p < 0.0001$) in biomass production between the external and internal rows (Fig. 3). At both sites there were interactions ($p < 0.005$) between external and internal row biomass and the within-row spacing treatments, driven by the higher yields of the external rows at shorter within-row spacing. The short within-row spacing outperformed the wider spacing at the $E_{pol}$ site ($p < 0.01$) while at the $E_{lox}$ site, the 4-row treatment yielded nearly 2 Mg $ha^{-1}$ $year^{-1}$ more than the 6-row treatment ($p < 0.05$). At most within-row spacing treatments, there was at least a doubling, but up to a five-fold difference in biomass production of the external rows compared to the internal rows. This was much more pronounced for the higher density within-row spacing treatments.

## Planting configuration affects individual tree size and number of stems

Average tree biomass varied significantly for both row number treatment and within-row spacing treatment (Table 2). Generally, for all row-number treatments and species, tree size increased with increasing within-row spacing such that the largest trees were observed in the 4 m within-row spacing (Fig. 4a). However, for $E_{pol}$ the 1- and 1.5 m within-row spacing treatments had similar productivity. For instance, at the $E_{lox}$ site, the 1-row treatment had the smallest mallee at the 1 m within-row spacing and averaged 61 kg per tree, while at the 4 m within-row spacing, average tree size increased to 264 kg. The magnitude of difference between the within-row spacing treatments was generally two to four-fold greater at the 4 m spacing

**Table 2** Linear mixed model analyses of arcsine-transformed survival, dry mallee productivity (Mg $ha^{-1}$ $yr^{-1}$) of both plot and paddock scenarios, natural log transformed average dry tree biomass and the number of stems per mallee

| Effect | *Eucalyptus loxophleba* site | | | | | *Eucalyptus polybractea* site | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | Survival | Productivity (plot) | Productivity (paddock) | ln (tree biomass) | Number of stems | Survival | Productivity (plot) | Productivity (paddock) | ln (tree biomass) | Number of stems |
| Fixed effects | *F* (ndf, ddf) | *F* (ndf, ddf) | *F* (ndf, ddf) | *F* (ndf, ddf) | *F* (ndf, ddf) | *F* (ndf, ddf) | *F* (ndf, ddf) | *F* (ndf, ddf) | *F* (ndf, ddf) | *F* (ndf, ddf) |
| Row | 4.1 (3,9)* | 30.1 (3,9) *** | 10.3 (3,9)* | 57.4 (3,9)*** | 3.7 (3,9) | 0.8 (3,8) | 20.1 (3,8)*** | 8.3 (3,8)* | 30.6 (3,8)*** | 12.3 (3,8)** |
| Spacing | 0.7 (4,46) | 1.9 (4,46) | 2.2 (4,46) | 61.3 (4,46)*** | 6.2 (4,46)*** | 2.0 (4,43) | 14.5 (4,43)*** | 15.5 (4,46)*** | 28.5 (4,43)*** | 5.8 (4,43)*** |
| Row × spacing | 0.7 (12,46) | 0.6 (12,46) | 0.5 (12,46) | 0.5 (12,46) | 0.8 (12,46) | 1.2 (12,43) | 1.5 (12,43) | 2.0 (12,43) | 1.5 (12,43) | 0.7 (12,43) |

*F*-values and numerator and denominator degrees of freedom in parentheses (ndf, ddf), for the fixed effects (row treatment, within-row spacing treatment and their interaction) for the *Eucalyptus loxophleba* subsp. *lissophloia* and *E. polybractea* spacing experiments near Narrogin, Western Australia. Significant test results are denoted as: * = $P < 0.05$; ** = $P < 0.001$; *** = $P < 0.0001$)

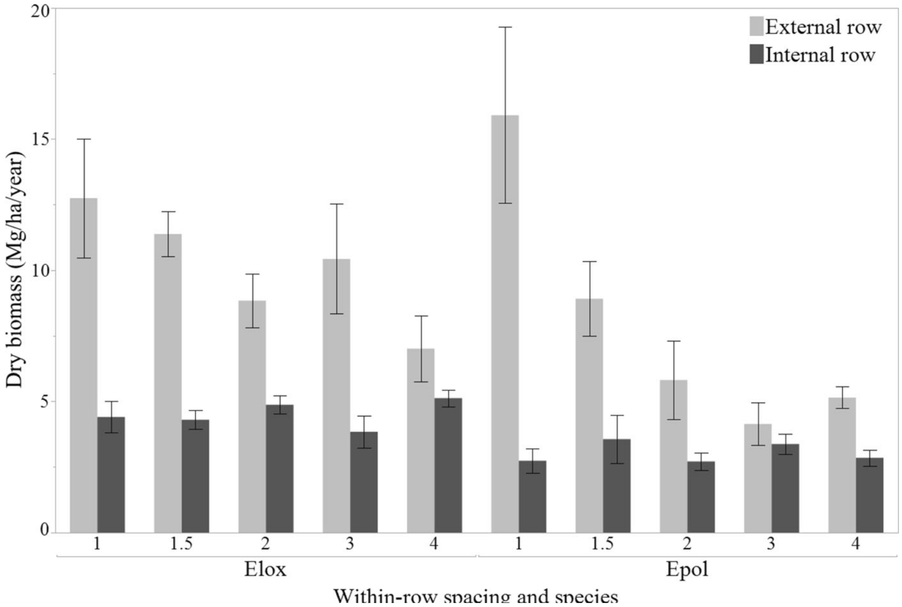

**Fig. 3** Yield responses of the internal and external rows of the combined 4- and 6-row treatments and within-row spacing treatments of *Eucalyptus loxophleba* subsp. *lissophloia* ($E_{lox}$) and *E. polybractea* ($E_{pol}$) at two sites near Narrogin, Western Australia. Error bars represent ± one standard error (n = 3–4)

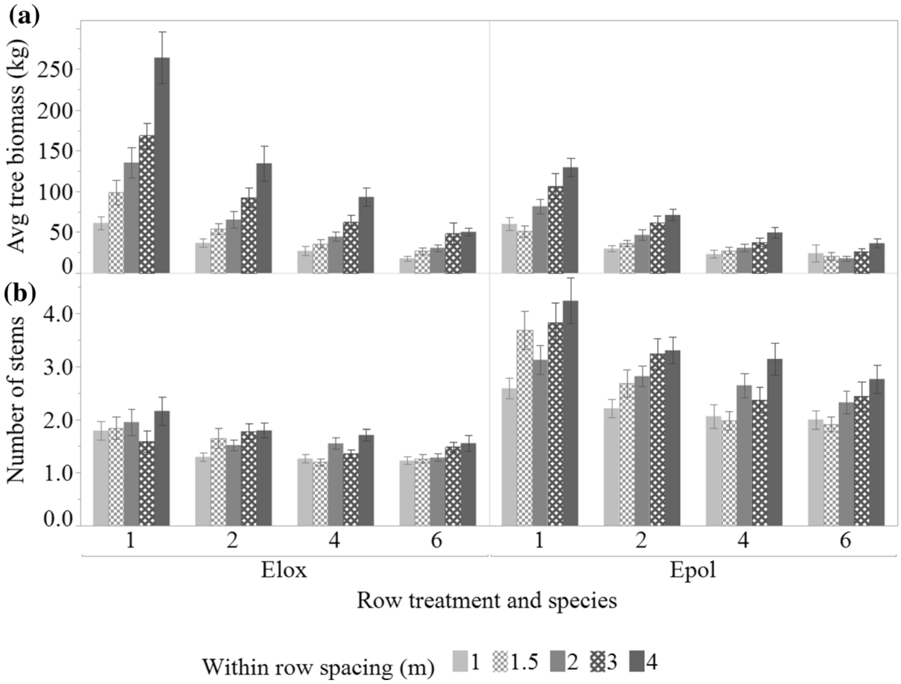

**Fig. 4** Effect of number of rows (1, 2, 4 and 6) and within-row spacing treatments on: **a** mallee size (kg dry biomass per tree); and **b** the number of stems per tree. Each graphic refers to the *Eucalyptus loxophleba* subsp. *lissophloia* ($E_{lox}$) and *E. polybractea* ($E_{pol}$) sites near Narrogin, Western Australia. Error bars represent ± one standard error (n = 3–4)

compared to the 1 m spacing. This difference was less pronounced at the $E_{pol}$ site especially for the 6-row treatment. Trees on average were also largest in the 1-row treatment and smallest in the 6-row treatment although there was no statistical difference between the 4- and 6-row treatments at the $E_{pol}$ site. The average tree biomass of the 1-row belt was three times the biomass of the 6-row treatment at both sites.

On average, $E_{pol}$ had more stems per mallee compared to the $E_{lox}$ site (2.8 vs 1.6 stems). The number of stems was significantly affected by within-row spacing at both sites (Table 2), with fewer stems per mallee at the denser within-row spacing treatments (Fig. 4b). At the $E_{lox}$ site, this ranged from 1.4 stems at the 1 m within-row spacing to 1.8 stem at the 4 m within-row spacing; the corresponding figures for the same treatments at the $E_{pol}$ site were 2.2 and 3.4 stems. The number of stems also varied significantly between the row treatments but only at the $E_{pol}$ site. The 1-row treatment averaged 3.5 stems per mallee, which decreased to 2.3 stems per mallee in the 6-row treatment. This trend, although not significant, was also apparent at the $E_{lox}$ site.

## Discussion

Understanding the impact of planting configuration and tree belt design on productivity of tree crops may facilitate their optimal integration into farming systems. To help develop this knowledge we examined effects of planting configuration on productivity of two commonly planted mallee species within the Western Australian wheat belt. Our results revealed that the design of a mallee belt exerts significant impacts on several key attributes including productivity, tree size and form (stem number), and tree mortality. These are discussed below.

### Biomass production

Productivity of the plot-level scenario of $E_{lox}$ and $E_{pol}$ in this study ranged from 2.7 to 21.2 Mg ha$^{-1}$ yr$^{-1}$. These results are mostly within the range observed for unharvested mallee productivity study from 19 sites in the Western Australian wheatbelt (Spencer et al. 2019). This study considers the impact of spacing configuration on productivity and found the productivity of the 1-row $E_{lox}$ (> 20 Mg ha$^{-1}$ year$^{-1}$ over 13 years) is the highest yield we have observed for this species. Biomass production per plot area, was affected by both the row treatment and within-row spacing. In this study, the 1-row treatment had significantly faster growth rates than the other treatments and productivity penalties were observed with additional rows and also with wider spacing.

The yield responses from the paddock-level scenario, in which wider belts produced more biomass than the narrower belts, were contrary to the plot level results. This was, however, expected: a 6-row belt from external stump to stump physically occupies 10 m, whereas a 2-row belt occupies only 2 m, which is a considerable difference with 50 m alley widths. This land is completely foregone to agriculture. The narrower-belts also have faster growth rates per tree. Competition imposed from unharvested mature mallee on the immediately adjacent agriculture has been found to extend a further 14 m from mature mallee belts (Sudmeyer et al. 2012), however, it is unknown if belt width will impact competition extent.

A common finding between the plot- and paddock-level scenarios was that shorter within-row spacing treatments were generally more productive. In plantation forestry, Binkley (2004) hypothesised that prior to canopy closure suppression of growth through tree dominance is low and resource supply is high for all trees. At this stage the increase in biomass production is a function of stocking rate. As competition between trees begins, growth rate slows, with earlier onset of competition in higher density plantings, where less competitive individuals are suppressed. The application of this concept to narrow belts indicates that competition will lead to conspicuous asymmetry in size between trees, described as phase two of the Binkley (2004) model. This was indeed observed in these two spacing experiments where clear asymmetry was observed in 4- and 6-row treatment, especially comparing the external with internal rows.

The lower productivity observed from the internal rows of the 4- and 6-row treatments was caused by the suppression of growth rates from the external rows. This production penalty has been observed elsewhere for mallee and other species (Ritson 2006; Prasad et al. 2010; Huxtable et al. 2012; Paula et al. 2013) and is driven by the trees in the external rows having greater access to the additional resources especially light, nutrients and water.

The most likely reason for the slower growth rates of many planting configurations is the lack of available water. In the Western Australian wheatbelt, the annual potential evaporation (PET) can be up to five-fold the annual rainfall (at Narrogin annual PET is three and a half times the annual rainfall) and water has been shown to be a major limiting resource for mallee belts. Rainfall has not been shown to be a predictor of mallee productivity (Spencer et al. 2019) probably because other water sources are available. For instance, Bennett et al. (2015) demonstrated by intercepting surface run-off by tree belts with small bunds, there was a 35% increase in biomass production. Mallee with access to fresh groundwater have shown up to ten times the biomass accumulation compared to those without access to groundwater (Wildy et al. 2004; Brooksbank et al. 2011). Access to these additional water sources are likely to benefit exterior trees with fewer rows and wider within-row spacing.

Work on other species in higher rainfall and lower insolation environments indicate that shading can limit tree growth (Long and Smith 1984; Righi et al. 2016; Pommerening and Sánchez Meador 2018). Wildy and Pate (2002) found that shaded *E. kochii* coppice produced less biomass than unshaded coppice in the first year post-harvest. Shading could be a factor in mallee belts especially because the larger external trees may shade the smaller internal-row trees during winter when radiation is lower and water is more readily available. However, if shading was limiting growth, the internal trees from the denser within-row spacing treatments would be less productive than the internal trees of the wider within-row spacing. This was not observed at these two sites, where there was a reduction in productivity of external trees with wider spacing, but the internal trees remained similarly suppressed (Fig. 4). Indeed, eucalypts tend to be crown-shy thus making shading due to crown dominance unlikely in even aged plantings (Lane-Poole 1936; Schönau and Coetzee 1989).

Competition for nutrients is another factor that could affect productivity under different planting configurations. Both spacing trials were located on fertilised annual cropping paddocks and trees from external rows, narrower belts or wider spacing would have greater access to additional nutrients. Indeed, in plantation forestry, soil nutrition may vary considerably in small areas across a site resulting in varied growth rates (Thomson 1986; Phillips and Marion 2004). Recently, we showed soil organic carbon and nitrogen ($NO_3^-$ and $NH_4^+$) were correlated with mallee biomass productivity in a multi-site long term study (Spencer et al. 2019). Organic carbon is probably a surrogate for nutrient supply and water availability in sandy soils (Doran and Smith 1987; Loveland and Webb 2003). The nitrogen correlation was, however, limited to frequently harvested treatments where biomass removal has been shown to deplete soil nitrogen stores (Grove et al. 2007; Yu et al. 2015). However, in this study, neither spacing trial had been harvested.

For both mallee species, the plot-level growth rate per tree of the 1-row belt was markedly higher than for the 2-row belt even at double the within-row spacing. Similar but smaller responses have been observed elsewhere but on much younger trees (Prasad et al. 2010; Paula et al. 2013). The process involved in this highly divergent response is unclear, but it suggests that competition between rows is more pronounced than competition within rows. A likely explanation is that trees in multiple row belts are subject to the additional competition of the neighbouring row. Within a few years of planting, root systems will overlap, competition for resources within the belt area will strengthen and roots will grow into the adjacent agricultural land to acquire water and nutrients. This lateral root growth has been observed with crop suppression in the alley of unharvested mallee where there was a reduction in crop and pasture yield by 36% between 2 and 20 m from the mallee belts compared to open paddock yields in the Western Australian wheatbelt (Sudmeyer et al. 2012). Such suppression of adjacent crops from agroforestry plantings have been widely observed in other countries (Rao et al. 1991; Prasad et al. 2010; Dagar et al. 2016; Oliveira et al. 2016).

Two metres between rows was generally viewed as the minimum distance for a harvester to access multiple row belts. This planting configuration was found to reduce mallee productivity compared to 1-row belts. Single-row belts may also reduce establishment costs and decrease the paddock area allocated to mallee while still achieving enhanced water use and some degree of salinity control. However, 2-row belts, compared to 1-row belts, may provide greater capacity to consume excess water and will be less porous, providing better stock shelter and wind erosion control. If the between-row spacing of 2 m was increased, this would reduce the penalty of the additional row and minimise the productivity difference between the 2- and 1-row belts.

### Tree size and form

As the within-row spacing increases, the average tree size increases. Average tree size is also affected by mortality, that is, if mortality is high in a plot, the average tree size of survivors also increases. This was observed in the 1-row $E_{lox}$ treatment where the average mallee biomass for the 4 m within-row spacing is more than fourfold as large as the 1 m within-row spacing. This difference was due to both the increased spacing and higher mortality at the 4 m within-row spacing. Mortality at larger within-row spacing will make available additional space and resources resulting in larger mallee than mortality at shorter spacing. In these experiments, there was a large range of whole-tree biomass across spacing treatments. The smallest trees were in the 4- and 6-row treatments at shorter within-row spacing. This divergence in mallee size will affect harvestability and proportions of the biomass components.

Maximum tree size, tree form and production per kilometre of row and belt are factors in the harvest viability of a mallee belt system. Mallee is difficult to harvest, having high wood density (Ilic et al. 2000) and multiple stems. Poplar, willow, sugar cane or forage harvesters are not suitable for harvesting mallee with large stem diameters (Giles and Harris 2003; Abadi et al. 2012), but traditional forestry harvesters have been used (Spinelli et al. 2014) and a prototype single-row chipper-harvester to improve harvesting efficiency has been developed and tested (Bartle 2009; Goss et al. 2014). Traditional forest harvesting equipment is more efficient with larger, taller trees. The chipper-harvester, being a continuously moving, integrated cutting-and-chipping operation, is mostly influenced by yield per kilometre of row, provided tree size range is below about 150 kg per tree. By varying the speed of the harvester, maximum efficiency can be maintained over a range of tree sizes, but overall harvest and transport (forwarding) efficiency is improved with high yields per kilometre of row (Abadi et al. 2012). Tree form is less significant for the chipper harvester than it is for traditional forest harvesting and chipping, but an upright form is easier to handle.

In the current work, the number of stems per tree is used as a proxy for upright form, and the number of stems increased for both species and with wider spacing. This response is similar to that observed in eucalypt forestry trials where branch size is inversely proportional to stocking rates (Neilsen and Gerrand 1999; Gerrand and Neilsen 2000; Henskens et al. 2001). Mallee belt design can therefore aim to use shorter within-row spacing to increase yield, reduce tree size and stem number. Concentrating biomass into fewer rows will reduce the total amount of biomass produced but may result in increased harvest efficiency for a chipper harvester because the biomass will be concentrated into fewer rows. In contrast, narrow belts would likely increase costs using traditional forestry equipment because additional travel distance would be required to process less biomass.

The strategy of maximising biomass while minimising tree size with shorter within-row spacing will alter the component partitioning of biomass with increased stemwood in larger trees (Paul et al. 2017). Foliar cineole, has greater economic value than wood, twig and bark (Barton 2000; Davis 2002). Currently, for leaf oil production, whole trees are harvested in the paddock with the oil extracted via hydro-distillation or steam distillation (Wildy et al. 2000b; Babu and Singh 2009). Both traditional forestry harvesting equipment and single-row chipper-harvester process whole tree biomass on-site ready for transport. This material can then be delivered to a processing plant where the leaf material would be separated from the other fractions and cineole extracted (Enecon 2001). The results from our study suggest there is scope to maximise leaf production by producing smaller mallee, without reducing mallee productivity. Where cineole production is a major objective, leaf biomass yield can be favoured by shorter within-row spacing. If a larger proportion of wood fraction is preferred then 1-row belts with larger within-row spacing can be used, but this may require conventional forestry harvesting equipment.

## Conclusion

The two species in this experiment showed broadly similar production responses to both row-number and within-row spacing treatments. Single row belts with shorter within-row spacing have faster growth rates per tree than any other configuration, particularly for $E_{pol}$. However, wider belts generate more biomass but the internal rows display considerable suppression with reduced productivity and occupy more land. Closer within row spacing will favour leaf biomass production. If wood biomass is the target product, narrow belts with wider spacing should be considered.

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**Code availability** The SAS code generated during analysis from the current study are available from the corresponding author on reasonable request.

**Compliance with ethical standards**

**Conflicts of interest** Authors declare that there are no conflicts of interest.

**Availability of data and material** The datasets generated during and/or analysed during the current study are available from the corresponding author on reasonable request.

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# Chapter 4

## Flowering phenology and genetic parameters with biomass and foliar cineole

Spencer B, Mazanec R, Abadi A, Gibberd M, Zerihun A (2020). Flowering phenology in a Eucalyptus loxophleba seed orchard, heritability and genetic correlation with biomass production and cineole: breeding strategy implications. Scientific Reports 10:15303 https://doi:10.1038/s41598-020-72346-3 www.nature.com/scientificreports

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# Flowering phenology in a *Eucalyptus loxophleba* seed orchard, heritability and genetic correlation with biomass production and cineole: breeding strategy implications

**Beren Spencer[1,2]✉, Richard Mazanec[2], Amir Abadi[1], Mark Gibberd[1] & Ayalsew Zerihun[1]**

**Reproductive synchronicity within a seed orchard facilitates gene exchange and reduces self-fertilisation. Here we assessed key flowering traits, biomass and foliar 1,8-cineole concentrations of *Eucalyptus loxophleba* (subsp. *lissophloia* and *gratiae*) in an open-pollinated seed orchard. Monthly flowering observations were made on 1142 trees from 60 families and nine provenances across 2 years. The percentage of trees flowering in both years was similar at 87%. There were differences between provenances and families within provenances for flowering traits, biomass and 1,8-cineole and interactions between provenances and year for flowering traits. Heritability of start and end flowering, and 1,8-cineole were high to moderate ($\hat{h}^2 = 0.75–0.45$) and duration of flowering, propensity to flower and biomass estimates were moderate to low ($\hat{h}^2 = 0.31–0.10$). Genetic and phenotypic correlations between flowering traits were high ($r_g = 0.96–0.63$ and $r_p = 0.93–0.34$) except between duration and end of flowering. The correlations were weaker between flowering traits and biomass or 1,8-cineole. 'Dual flowering', when trees underwent two reproductive cycles in a year, was responsible for out-of-phase flowering and those with low biomass and 1,8-cineole concentration should be removed from the breeding programme to hasten selection for desirable traits.**

In southwest Western Australia, as elsewhere in Australia, the large scale conversion of native vegetation to agricultural land was followed by extensive salinity problems[1,2]. In the early 1990s, a research program was initiated to identify multi-purpose perennial crops that could mitigate the salinity problems while providing additional economic benefit when integrated with the annual crop based farming systems of the region[3]. The research identified mallee eucalypts as the preferred candidates due to their capacity to coppice after short-cycle harvest[4–7]. While the initial selection of mallee species focussed on foliar 1,8-cineole concentration (hereafter referred to as cineole), for large-scale bio-renewable feedstock[8–11], subsequently, biomass yield emerged as an additional selection criterion as the opportunity for carbon sequestration and bioenergy became prospective during the late 1990s[12–14].

Two species, *Eucalyptus loxophleba* subsp. *Lissophloia* L.A.S. Johnson & K.D. Hill, hereafter referred to as $E_{liss}$, and *E. loxophleba* subsp. *gratiae* Brooker, hereafter referred to as $E_{grat}$ were selected for development given their high concentrations of foliar cineole, fast growth rates and prominence in native woodlands of the southwest of Western Australia[15,16]. Successful development of improved tree crops, with multiple desirable traits, presents a challenge with limited understanding of key genetic parameters, such as heritability, genetic and phenotypic correlations. Thus, in 1993 a breeding programme was initiated; and by 2002, 11 *E. loxophleba* trials had been established using progeny from a total of 78 parents selected in the wild for high cineole content[17,18]. The trials

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were assessed for foliar cineole concentration and biomass then thinned to produce genetically improved seed for agroforestry plantings. This study assessed one of these open-pollinated seed orchards for key flowering traits, cineole and biomass yield with the aim of facilitating production of improved seed to enhance the potential commercial viability of these species as tree crops.

The timing of key reproductive events is crucial for outcrossing, and in eucalypts barriers that restrict pollen dispersal can result in self-pollination and reduced fitness[19–22]. Open-pollinated seed orchards, without any reproductive isolation could facilitate panmixia resulting in panmictic equilibrium. A shift from panmictic equilibrium (e.g. due to asynchrony in flowering time) may lead to unbalanced contributions of genetic material from individual parents to the next generation and reproductive isolation[23]. Several studies have shown that time of anthesis in eucalyptus is under genetic control, resulting in different flowering times between provenances and families[24–27]. Therefore, understanding the reproductive phenology of a seed orchard can enable culling reproductively isolated trees, families or provenances thereby increasing the overall level of outcrossing in the orchard.

Synchronicity of flowering within a seed orchard also enables transmission of favourable genes or traits in a breeding programme. For instance, foliar cineole concentration for $E_{liss}$ has a high narrow sense heritability of $0.53 \pm 0.07$[18]. Fully exploiting traits with high heritability relies on outcrossing (synchronised flowering) within a seed orchard. Padovan et al.[28] identified 12 single nucleotide polymorphisms for *E. loxophleba* that are associated with terpene production including two for cineole. However, when selecting for a breeding trait, unfavourable genetic correlation between traits could compromise gains for the second trait. For instance, with *E. camaldulensis*, there is a moderate negative correlation between total monoterpenes (including cineole) and biomass[29]. Understanding the correlations between key flowering traits, biomass and foliar cineole concentration will reduce unfavourable bias from breeding selections.

The flowering phenology of several eucalypt species are documented, for example *E. regnans*[30] and *E. globulus*[24], but only general observations of flowering periods in natural populations have been made for $E_{liss}$ and $E_{grat}$. Records of the timing of flowering activity for $E_{liss}$ indicate some variation including September to February[31] and August to October[32]. Similarly, $E_{grat}$ has been documented to flower between September and December[31] and from October to November[32]. Bell[33] monitored flowering in three $E_{liss}$ families in a seed orchard near Collie, Western Australia, where peak flowering occurred from November to December, however some individuals flowered as early as June/July. Observations in $E_{liss}$ and $E_{grat}$ seed orchards and in wild populations indicated that flowering can be observed at almost any time of the year (B. Spencer and W. O'Sullivan personal observations).

The timing and duration of anthesis for a species determines its capacity to breed within a plantation but also with neighbouring populations of closely related species. Genetic pollution by pollen dispersal has been identified as an important conservation threat that can lead to extinction[34]. This is particularly so in the *Eucalyptus* genus due to highly mobile pollen and weak reproductive barriers[35]. Further, the considerable interest in tree planting on farms has facilitated pollen flow between formerly isolated species resulting in exotic eucalypt hybrids[36,37] and the *E. loxophleba* group is known to be a prolific pollinator. For example, an exotic $E_{liss}$ planting has pollinated over half of a remnant *E. loxophleba* ssp. *supralaevis* population, with the exotic pollen travelling up to 1940 m from its source[38]. *Eucalyptus loxophleba* has been recorded as hybridising with *E. kruseana*[39], *E. wandoo*[40] and *E. absita*[41] where the species naturally co-exist. Additionally, the Western Australian Herbarium database lists hybrids of the various subspecies of *E. loxophleba* with *E. accedens, E. astringens, E. blaxellii, E. erythronema, E. occidentalis, E. orthostemon, E. rudis, E. spathulata* and *E. victrix*[42].

This work examines the flowering phenology of $E_{liss}$ and $E_{grat}$ in a single orchard and determines:

- the variation between and within subspecies, provenances and families to identify out of phase provenances and families under a common environment setting; and
- the heritability and genetic parameters of key flowering traits, biomass and foliar cineole.

It is expected that knowledge of genetic control and heritability of the various traits considered here, and culling of those which flower outside of the peak flowering period from the seed orchard will speed up production of seeds with desired traits and support commercial viability of mallee as bioenergy tree corps.

## Methods

### Study site.

The study site was located approximately 200 km south east of Perth near Toolibin Lake (32.88°S, 117.62°E). The average rainfall is approximately 400 mm and falls mainly between May and September.

The seed orchard, containing both $E_{liss}$ and $E_{grat}$ was planted in 1999 and the population contains 60 families from nine provenances and three broad regions encompassing the full natural distribution of these taxa (Table 1, Fig. 1). Here a family is defined as the sexually produced open-pollinated progeny from a wild parent tree and a provenance refers to the progeny of wild trees from a known geographic range. The distribution of $E_{liss}$ is much wider than $E_{grat}$ and to reflect this, seven of the nine provenances were $E_{liss}$ (Fig. 1). The 60 families were planted in six-tree row plots in a randomised row-column design with 20 replicates. The orchard was thinned in 2005 from 7200 trees to 1142 based on breeding values for foliar cineole concentration and above ground biomass. After thinning, all 60 families and nine provenances were still represented in the seed orchard with the number of trees retained in each family ranging from 14 to 22.

### Floral assessments.

Floral assessments were completed approximately every 4 weeks from May 2012 to January 2013 and from February 2014 to January 2015. To carry out these assessments the canopy of each tree was scanned for presence of reproductive activity from the ground using 8 × 40 binoculars in teams of two to reduce assessor bias. In addition, several times a day, observer teams were calibrated with each other to further reduce bias.

| Region | Provenance | No families | No trees assessed | Mean maximum temperature (°C) | Mean minimum temperature (°C) | Mean annual rainfall (mm) | Mean annual pan evaporation (mm) | Elevation (m) |
|---|---|---|---|---|---|---|---|---|
| $E_{grat}$ South | Dumbleyung | 7 | 133 | 22.9 | 9.9 | 340.5 | 1674.1 | 341 |
| $E_{grat}$ South | Lake Grace | 14 | 274 | 23.5 | 10.0 | 330.3 | 1798.4 | 329 |
| $E_{liss}$ East | Coolgardie | 3 | 53 | 25.1 | 11.2 | 306.4 | 2412.7 | 469 |
| $E_{liss}$ East | Goongarrie | 2 | 39 | 26.5 | 12.7 | 275.1 | 2698.4 | 415 |
| $E_{liss}$ East | Norseman | 11 | 217 | 24.6 | 10.4 | 302.4 | 2114.2 | 375 |
| $E_{liss}$ West | Narembeen | 4 | 78 | 24.2 | 10.0 | 370.1 | 1931.5 | 457 |
| $E_{liss}$ West | SouthernCrs | 5 | 90 | 25.4 | 11.4 | 317.6 | 2167.7 | 400 |
| $E_{liss}$ West | Trayning | 11 | 208 | 25.7 | 11.6 | 321.8 | 2235.2 | 378 |
| $E_{liss}$ West | Westonia | 3 | 50 | 25.5 | 11.0 | 335.3 | 2264.2 | 354 |

**Table 1.** Descriptions of regions, provenances, and climate of locations from where the two subspecies of *Eucalyptus loxophleba* for the seed orchard planting were sourced. The number of families in each provenance and individual trees assessed for flowering traits are also described. Climatic data and elevation which were obtained from SILO dataset[43] from 1985 to 2015.

Each tree in the seed orchard was assessed for the three stages of reproduction: immature buds; mature buds; and flowering. Firstly, the immature buds were assessed following the shedding of the involucral bracts. This was separated into three phases: a score of one was given when the involucral bracts had just shed but the buds were still clustered together; a score of two indicated clear separation between the buds; a score of three was given when the operculum was visible and buds were approaching mature size. If a tree had more than one of these phases present during any one assessment, the different phases were recorded. In the 2012 assessment, it was noted that many individual trees had two reproductive events within the year. In the 2014 assessment, where these separate events were observed within a tree, they were recorded separately. These are referred to as reproductive flushes.

After the buds had matured, the bud density was assessed on a scale of one to four. A rank of four was given to trees with a dense crop (mature buds, flowers and capsules post-anthesis) distributed evenly throughout the tree. A score of three was given to trees with an average crop density, or a dense crop unevenly distributed throughout the canopy. A score of two was given to trees with a sparse crop and a score of one to trees with a crop too sparse to accurately assess.

Finally, as flowering matured, the percentage of buds, active flowers and spent flowers were estimated from the full canopy of each tree. In the 2014 assessment, each reproductive flush was recorded separately. Different flushes were discernible because there was a physical gap between the reproductive flushes which were usually separated by vegetative growth and the newer buds were smaller and brighter green. Flowers were determined as active when the operculum had been shed and white or cream anthers were evident. Flowers were classified as spent when the anthers were desiccated, brown or absent.

## Flowering criteria.

Where a bud density score of one was observed, that reproductive flush was eliminated from the analysis. These flushes were sparse and scores fluctuated widely between assessments. This appeared to be due to loss of reproductive structures through natural abortion or predation processes. These flushes also tended to receive inflated flowering scores because flowers are more prominent than buds on a sparse crop. We defined flowering as a phase when at least two percent of the buds in a reproductive phase were in flower with fresh anthers present. A score of 1% was used to represent few extant flowers to assist the team in the following assessment to indicate that the tree had commenced flowering. Additionally, a tree was defined as flowering when more than 5% of its pre-flowering buds had progressed past anthesis between assessments. This was rarely seen during autumn and winter assessments, but was on occasions in spring and early summer, when flowering proceeded more rapidly.

## Biomass assessment.

Two biomass assessments were taken for individuals within this seed orchard. The first assessment was taken pre-thinning in autumn 2004 by measuring the Crown Volume Index (CVI), as described in Spencer et al.[5], of each of the 5372 healthy trees in the orchard. Briefly, CVI is the measurement of the height and two perpendicular crown widths expressed as $m^3$.

The stem basal area of the remaining trees post-thinning (1163 trees) was assessed in February 2014. The stems of each tree were measured with a diameter tape approximately 10 cm above the ground. Loose and fibrous bark was removed and burls and buttressing associated with the lignotubers were avoided. A single diameter estimate was obtained by calculating the Equivalent Diameter near Root Collar (EDRC) as specified in Chojnacky and Milton[44] using the formula:

$$EDRC = \sqrt{\sum_{i=1}^{n} \mathrm{drc}_i^2} \tag{1}$$

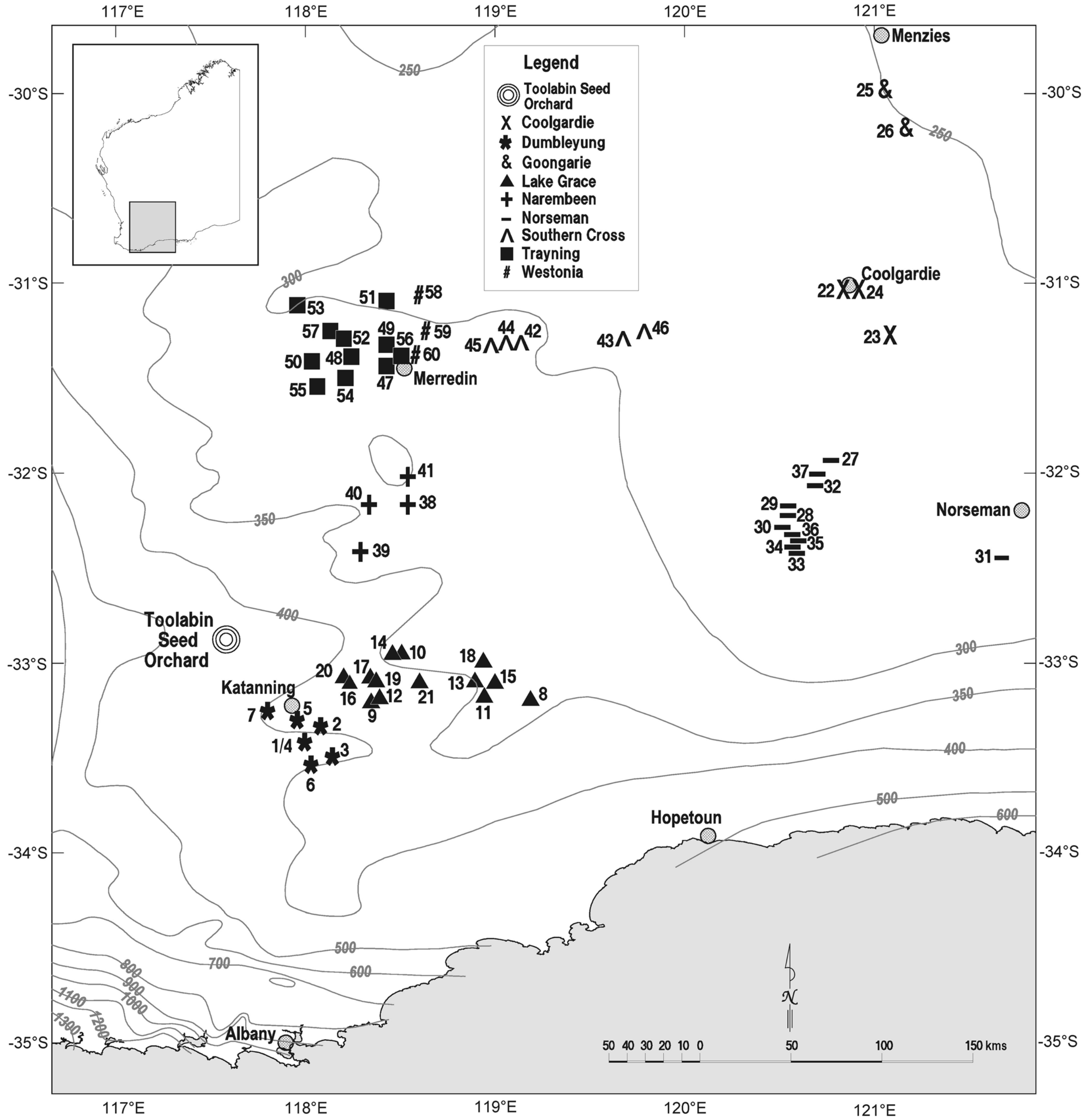

**Figure 1.** The location of the 60 parent trees from the *Eucalyptus loxophleba* seed orchard and their provenances. Circles represent key population centres and large triple circle is the location of the seed orchard. The eastern $E_{liss}$ region comprises Goongarie, Coolgardie and Norseman. The western $E_{liss}$ region comprises Westonia, Trayning, Southern Cross and Narembeen. The Egrat subsp. has two provenances, Lake Grace and Dumbleyung. This map was generated using MicroStation Version 8i. Bentley Systems Incorporated, 2006 https://www.bentley.com/en/products/brands/microstation.

where *n* is number the number of stems *drc* is individual diameter of each stem. General $E_{liss}$ species allometric equations were applied to the natural log of CVI and stem diameter to convert to dry biomass as specified in Spencer et al.[5] using the back-transformation correction methodology as outlined by Baskerville[45].

**Cineole assessment.** Leaf samples were collected in autumn 2004 from 5223 healthy trees in the seed orchard. For each tree, cineole was extracted from 3 g leaf samples in 50 mL ethanol for $\geq 4$ weeks and analysed using HP5890A gas chromatograph as detailed in Mazanec et al.[18].

**Heat sum.** Heat sum was calculated by averaging the maximum and minimum daily temperatures above a base temperature below which an organism will not develop[46]. Climatic data was obtained through SILO from interpolated dataset[43]. We used a base temperature of 5 °C which was determined for *E. globulus*[47] and used in eucalypt flowering studies by Jones et al.[26]. The annual heat sum for the flowering year was calculated from the first day of summer preceding the assessment year.

**Statistical analysis.** The floral traits (start, end, duration of flowering and the number of reproductive flushes), biomass and foliar cineole of each tree were analysed using a series of mixed linear models in SAS 9.4[48]:

$$y_{ijklmn} = r_i + c_j(r_i) + b_k(r_i) + p_l + f_m(p_l) + y_n + p_l.y_n + y_n.f_m(p_l) + e_{ijklmn} \tag{2a}$$

$$y_{ijklm} = r_i + c_j(r_i) + b_k(r_i) + p_l + f_m(p_l) + e_{ijklm} \tag{2b}$$

where $y_{ijklmn}$ *and* $y_{ijklm}$ are the floral, biomass and cineole traits, $r_i$ is the random replicate effect, $c_j(r_i)$ is the random effect column$_j$ nested within replicate$_i$, $b_k(r_i)$ is the random effect row$_k$ nested within replicate$_i$, $p_l$ is the provenance$_l$, $f_m(p_l)$ is the family$_m$ within the provenance$_l$, $y_n$ is the year$_n$ and $e_{ijklmn}$ and $y_{ijklm}$ are the residual errors of the respective models. Interactions were tested between year and provenance ($p_l.y_n$), as well as between year and family nested within provenance ($y_n.f_m(p_l)$; however, the proportion of dual flush trees, cineole and biomass analyses excluded the year factor $y_n$ from the model (Eq. 2b) because these data were not collected over multiple years. Given that the dataset was unbalanced due to mortality and thinning, replicate was treated as a random effect so that inter-block information could be recovered[49]. Biomass data was transformed using natural logarithms to conform to homogeneity assumptions. The proportion of trees that flowered and proportion of dual flush trees were tested with the same model following arcsine transformation. Tukey–Kramer tests were used to determine the differences in the least square means of key traits. Start of flowering week was standardised to compare the 2012 and 2014 assessments. At the first assessment in 2012, 9% of the trees were flowering, and the 2014 data were truncated to match the 9% flowering from the 2012 assessment.

To estimate heritability, the data were analysed with ASReml Version 4.1[50] using a linear mixed model (Eq. 2a). To estimate univariate heritability in flowering traits from 2012 and 2014, Eq. (2b) was used but family within the provenance was a random effect as required to calculate variance for heritability estimates[51]. The heritability analysis for number of flushes and propensity to flower in each year used a binomial model with a logit link. Genetic correlations were estimated from bivariate analysis of traits. Insignificant random effects were removed if they introduced instability to the model and log-likelihood was used to determine the model of best fit.

Eucalypts have a mixed breeding system in open-pollinated seed orchards and self-fertilisation is common[19]. Based on the outcome of the Sampson and Byrne[38] study of a third closely related subspecies in the loxophleba group, *E. loxophleba* subsp. *supralaevis*, it is assumed that $E_{liss}$ and $E_{grat}$ have a mixed mating system. Griffin and Cotterill[20] suggested using a coefficient of relationship of $\rho = 1/1.25$ to compensate for mixed mating and a selfing percentage of 30% for seed sourced from wild populations of *E. regnans*. This approach has been assessed and confirmed appropriate for correcting heritability estimates in an open-pollinated eucalypt progeny trial[52] and subsequently applied to a series of $E_{liss}$ progeny trials[18].

Narrow-sense heritability was calculated from variance components of the individual traits using the formula:

$$\hat{h}^2 = \frac{\sigma_a^2}{\left(\sigma_a^2 + \sigma_\eta^2\right)} \tag{3}$$

where $\hat{h}^2$ is the narrow sense heritability, $\sigma^2_a$ is the additive genetic variance and $\sigma^2_\eta$ residual error component of variance.

Genetic and phenotypic correlations were calculated between all combinations of flowering traits (start, duration and end), dry biomass weight and foliar cineole concentration using the formula:

$$r = \frac{\sigma_{xy}}{\sqrt{\sigma_x^2 * \sigma_y^2}} \tag{4}$$

where r is either $r_g$, the genetic correlation, or $r_p$, the phenotypic correlation. $\sigma_{xy}$ is the additive genetic covariance for the genetic correlations and $\sigma^2_x$ and $\sigma^2_y$ are the additive genetic variances of the two traits. For the phenotypic correlations, $\sigma_{xy}$ is the phenotypic covariance and $\sigma^2_x$ and $\sigma^2_y$ are the phenotypic variances for the traits.

## Results

**General flowering observations.** Of the population of 1142 trees, 87% flowered in each year (Table 2, Fig. 2a). The reproductive activity differed between the subspecies with $E_{grat}$ from the two south provenances having the lowest (79–82%) flowering in both years (Table 2). $E_{liss}$ showed consistent reproductive activity: 91% and 93% from the eastern and western provenances, respectively. From the monthly observations, the population commenced flowering in late summer to early autumn, with peak flowering in spring (Fig. 2a). Flowering was about 2 weeks earlier in 2014 and this trend continued for most of the assessment period. The earlier flowering in 2014 corresponded with a higher heat sum (4515 °C) compared to 2012 (4332 °C).

Tracking of individual reproductive flushes in 2014 revealed that 16% of trees flowered on two separate occasions (Table 2). Two provenances from the $E_{liss}$ east region had the highest proportion of dual flush trees followed by the two $E_{grat}$ provenances. The trees that had dual reproductive phases exhibited two different flowering

| Provenance | Region | % flower 2012 | % flower 2014 | % dual flush 2014 |
|---|---|---|---|---|
| Dumbleyung | $E_{grat}$ south | 81.8 | 80.3 | 17.4 |
| Lake Grace | $E_{grat}$ south | 82.8 | 78.4 | 21.2 |
| Coolgardie | $E_{liss}$ east | 86.8 | 94.3 | 28.3 |
| Goongarrie | $E_{liss}$ east | 84.6 | 94.9 | 5.1 |
| Norseman | $E_{liss}$ east | 93.1 | 89.8 | 34.3 |
| Narembeen | $E_{liss}$ west | 94.9 | 84.6 | 1.3 |
| Southern Cross | $E_{liss}$ west | 83.3 | 91.1 | 4.4 |
| Trayning | $E_{liss}$ west | 88.0 | 95.7 | 1.4 |
| Westonia | $E_{liss}$ west | 92.0 | 92.0 | 8.0 |

**Table 2.** Proportion of individuals flowering (%) for trees derived from each provenance for each assessment year and the proportion of trees with dual reproductive flushes (%) in the 2014 assessments.

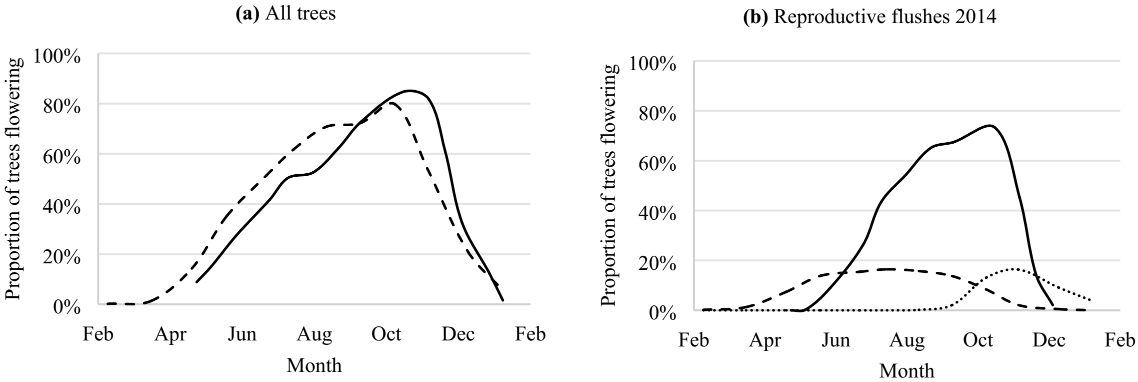

**Figure 2.** Proportion and timing of all trees flowering in the *Eucalyptus loxophleba* seed orchard at monthly intervals: (**a**) across all families and provenances in 2012 (solid line) and 2014 (dashed line); and (**b**) of single flush (black solid line) and the first (dashed line) and second (dotted line) flush of dual flush trees. Panels (**a**) and (**b**) were generated using Microsoft Excel, Version 2016. Microsoft Corporation, 2016 https://www.microsoft.com/en-au/download/office.aspx.

phenologies; the first flush of the dual flush trees had a long flowering duration compared to the second flush (Fig. 2b). In trees with a single flowering flush, the timing of flower initiation affected the duration of flowering. Trees that started flowering in late summer and autumn flowered for over 20 weeks, whereas a flowering phase that commenced in late winter and early spring flowered for under 12 weeks (Supplementary Table S1). In assessing the duration of flowering, flowering phases that commenced after week 38 were omitted because a high proportion had not concluded by the time the study was terminated.

**Effect of provenances on traits.** Flowering start, end, duration, proportion of flowering, proportion of dual flush, foliar cineole content and biomass differed, significantly ($P < 0.0001$) between provenance and family within provenance and between family within provenance for start of flowering ($P < 0.001$) (Table 3). High levels of synchronicity in flowering events occurred within provenances, but there were substantial differences in the timing of flowering between provenances, as shown in (Fig. 3 and Table 4). For all the flowering traits assessed in this study, there were provenance by year interactions. Thus, a few provenances varied substantially between flowering years, with Dumbleyung, Southern Cross and Westonia finishing flowering four weeks later in 2014 compared to 2012. Also, the Norseman and Southern Cross provenances commenced flowering earlier in 2012 than in 2014 while all other provenances started flowering later. There were considerable provenance differences between flowering traits. The Coolgardie provenance commenced flowering earlier than the other provenances in both years, whereas the Goongarrie provenance started flowering later. All provenances finished flowering later in 2012 than 2014. Flowering duration was generally longer in 2014 than in 2012 with about a 4-week difference at Norseman and Southern Cross. Flowering was shortest in both years for $E_{liss}$ provenances from Goongarrie. The other $E_{liss}$ east provenances had some of the longest flowering duration. The $E_{grat}$ provenances were less reproductively active across both years except when compared to Narembeen in 2014 and Southern Cross, Goongarrie and Coolgardie in 2012.

Two of the $E_{liss}$ east provenances, Norseman and Coolgardie, had the highest proportion of dual flush trees followed by the two $E_{grat}$ provenances (Tables 2, 5). All of the $E_{liss}$ west provenances had a consistently low (1–8%) percentage of dual flush trees. The average foliar cineole concentrations for each provenance ranged from 1.6% in Southern Cross to 2.7% of green leaf weight in Coolgardie (Table 5). On average, the $E_{liss}$ east provenances tended to have higher cineole concentration, while the $E_{liss}$ west region contained the two worst performing provenances.

| Effect | Flowering | | | Proportion | | | ln (dry biomass) | |
|---|---|---|---|---|---|---|---|---|
| | Start | End | Duration | Flowering | Dual flush | Foliar cineole | Pre-thinned | Post-thinned |
| Fixed effects | *F* (degree of freedom) or *Z* value | *F* (degree of freedom) or *Z* value | *F* (degree of freedom) or *Z* value | *F* (degree of freedom) or *Z* value | *F* (degree of freedom) or *Z* value | *F* (degree of freedom) or *Z* value | *F* (degree of freedom) or *Z* value | *F* (degree of freedom) or *Z* value |
| Year | 11.1 (1)** | 353.0 (1)*** | 34.2 (1)*** | 0.52 (1) | | | | |
| Prov | 17.2 (8)*** | 57.4 (8)*** | 26.7 (8)*** | 7.38 (8)*** | 20.98 (8)*** | 104.6 (8)*** | 28.6 (8)*** | 11.2 (8)*** |
| Family(Prov) | 3.1 (8)** | 12.1 (51)*** | 4.4 (51)*** | 2.63 (51)*** | 4.29 (51)*** | 22.1 (51)*** | 4.1 (51)*** | 3.3 (51)*** |
| Year*Prov | 7.8 (51)*** | 3.6 (8)** | 2.2 (8)* | 2.51 (8)* | | | | |
| Year*Family(Prov) | 1.0 (51) | 1.9 (51)** | 1.1 (51) | 1.05 (51) | | | | |
| **Random effects** | | | | | | | | |
| Rep | 0.0 | 1.7* | 0.0 | 2.0* | 0.0 | 2.3* | 2.7** | 0.6 |
| Col(Rep) | 2.2* | 2.5** | 1.9** | 1.9** | 0.0 | 2.0 | 4.6*** | 4.0*** |
| Row(Rep) | 4.0*** | 2.3** | 2.9*** | 0.0 | 1.0 | 2.4*** | 4.5*** | 2.9** |
| Residual | 27.9*** | 28.0*** | 41.6*** | 32.1*** | 21.4*** | 49.3*** | 50.1*** | 20.2*** |

**Table 3.** Results of least square means analysis of flowering traits, cineole and biomass. *F*-values and degrees of freedom, in parentheses, for the fixed effects traits (start, end and duration of flowering, proportion flowering, proportion dual flush, foliar cineole concentration and pre- and post-thinning biomass estimates) and the *Z*-values for the random effects (replicate, column nested within replicate and row nested within replicate) at the *Eucalyptus loxophleba* seed orchard. Significance test results for fixed and random effects are denoted as: $*P < 0.05$; $**P < 0.001$; $***P < 0.0001$.

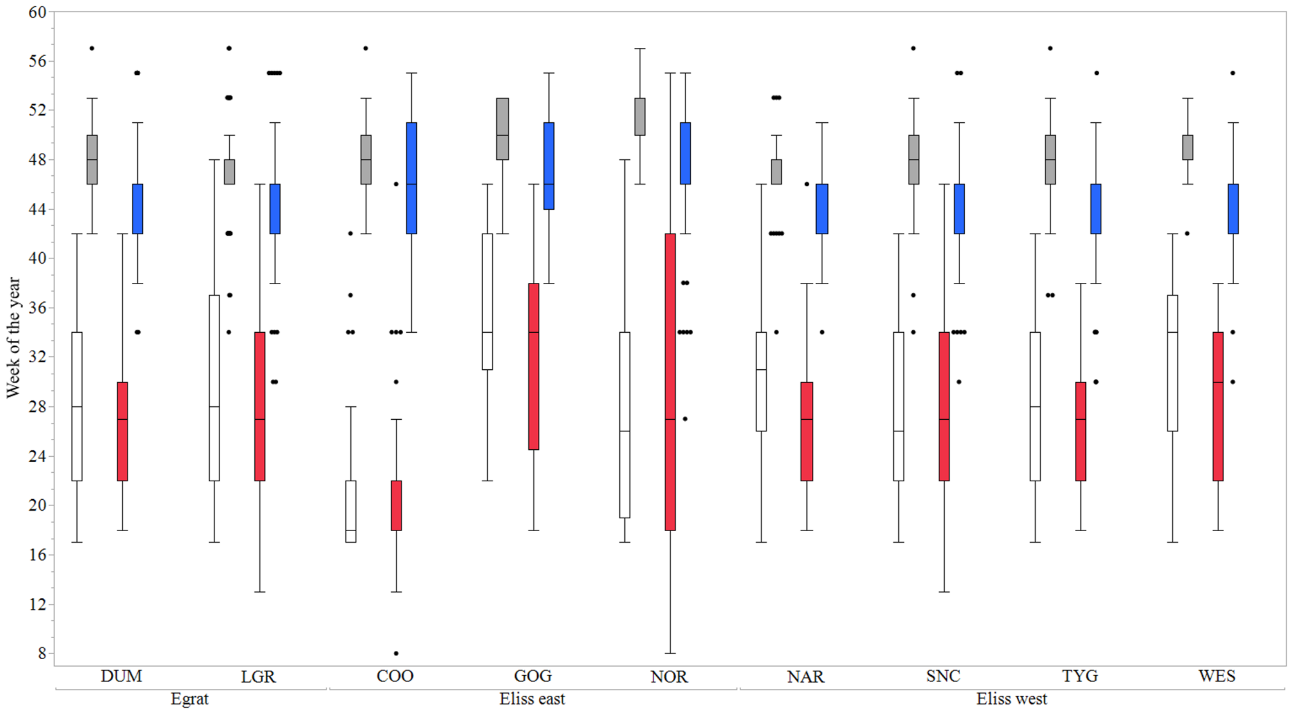

**Figure 3.** Start and end flowering times from 2012 and 2014 of *Eucalyptus loxophleba* provenances. Box plots represent variation in flowering times for 2012 start (no fill) and end (grey fill) and 2014 start (red fill) and end (blue fill). Flowering week is from the first week of January. Line in the middle of each box is the median provenance flowering time and edge of boxes 25th and 75th percentile. Dots represent outliers. Provenances are: *DUM* Dumbleyung, *LGR* Lake Grace, *COO* Coolgardie, *GOG* Goongarrie, *NOR* Norseman, *NAR* Narembeen, *SNC* Southern Cross, *TYG* Trayning, *WES* Westonia. Figure was generated using JMP, Version 14. SAS Institute Inc. 1989–2019 https://www.jmp.com/en_au/home.html.

There was consistency in the biomass of different provenances between first and second measurement (Table 5). For the pre- and post-thinning growth, both $E_{grat}$ provenances were ranked among the top performers along with $E_{liss}$ from Goongarrie. Coolgardie and Westonia were the poorest performers in both assessments. On average, $E_{liss}$ east and $E_{liss}$ west were similar in their biomass ranking.

| | | | Start flowering | | End flowering | | Duration flowering | | Proportion flowering | |
|---|---|---|---|---|---|---|---|---|---|---|
| **Region** | **Prov** | **Year** | **Estimate (SE)** | **Sig** | **Estimate (SE)** | **Sig** | **Estimate (SE)** | **Sig** | **Estimate (SE)** | **Sig** |
| $E_{grat}$ | DUM | 2012 | 28.4 (0.67) | BCD | 47.8 (0.47) | BCD | 27.5 (1.00) | A | 1.29 (0.05) | CD |
| $E_{grat}$ | DUM | 2014 | 26.9 (0.68) | CD | 45.7 (0.45) | DE | 24.7 (0.96) | ABC | 1.26 (0.05) | D |
| $E_{grat}$ | LGR | 2012 | 29.2 (0.48) | BC | 48.1 (0.32) | BC | 20.7 (0.66) | CDEF | 1.30 (0.05) | CD |
| $E_{grat}$ | LGR | 2014 | 27.6 (0.50) | CD | 44.0 (0.32) | E | 18.1 (0.67) | EFG | 1.23 (0.05) | D |
| $E_{liss}$ east | COO | 2012 | 21.4 (1.02) | E | 49.3 (0.55) | AB | 16.0 (1.18) | G | 1.36 (0.08) | ABCD |
| $E_{liss}$ east | COO | 2014 | 21.2 (0.97) | E | 47.4 (0.52) | BCD | 16.2 (1.11) | G | 1.48 (0.08) | AB |
| $E_{liss}$ east | GOG | 2012 | 34.4 (1.21) | A | 47.3 (0.24) | CD | 19.0 (0.48) | EFG | 1.34 (0.09) | ABCD |
| $E_{liss}$ east | GOG | 2014 | 32.2 (1.13) | AB | 44.4 (0.25) | E | 17.7 (0.49) | G | 1.49 (0.09) | AB |
| $E_{liss}$ east | NOR | 2012 | 27.3 (0.50) | CD | 47.3 (0.38) | BCD | 17.9 (0.79) | EFG | 1.47 (0.05) | AB |
| $E_{liss}$ east | NOR | 2014 | 28.8 (0.51) | BCD | 44.3 (0.40) | E | 17.5 (0.84) | EFG | 1.42 (0.05) | AB |
| $E_{liss}$ west | NAR | 2012 | 30.4 (0.80) | ABC | 50.6 (0.25) | A | 24.4 (0.49) | AB | 1.49 (0.07) | AB |
| $E_{liss}$ west | NAR | 2014 | 27.7 (0.86) | BCD | 48.7 (0.25) | B | 20.8 (0.50) | DE | 1.32 (0.07) | BCD |
| $E_{liss}$ west | SNC | 2012 | 26.8 (0.80) | CD | 48.0 (0.38) | BC | 22.2 (0.78) | BCD | 1.33 (0.06) | BCD |
| $E_{liss}$ west | SNC | 2014 | 27.3 (0.76) | CD | 44.6 (0.36) | E | 18.0 (0.75) | EFG | 1.45 (0.06) | AB |
| $E_{liss}$ west | TYG | 2012 | 29.0 (0.52) | BCD | 47.8 (0.26) | BC | 19.8 (0.52) | DEFG | 1.39 (0.05) | ABC |
| $E_{liss}$ west | TYG | 2014 | 26.8 (0.50) | D | 44.3 (0.25) | E | 18.4 (0.50) | FG | 1.50 (0.05) | A |
| $E_{liss}$ west | WES | 2012 | 30.3 (1.03) | ABCD | 48.5 (0.48) | BC | 19.3 (1.01) | DEFG | 1.46 (0.08) | AB |
| $E_{liss}$ west | WES | 2014 | 27.8 (1.03) | BCD | 44.8 (0.48) | E | 18.0 (1.01) | DEFG | 1.46 (0.08) | AB |

**Table 4.** Least square means of flowering traits for provenance by year with the standard error (in parentheses) at the *Eucalyptus loxophleba* seed orchard. Proportion flowering was arcsine-transformed. Tukey's tests were performed to determine the difference between provenances by year interaction, means that are similar have the same letter at α = 0.05 significance level (Sig). Provenances are: *DUM* Dumbleyung, *LGR* Lake Grace, *COO* Coolgardie, *GOG* Goongarrie, *NOR* Norseman, *NAR* Narembeen, *SNC* Southern Cross, *TYG* Trayning, *WES* Westonia.

| | | Proportion dual flush | | Cineole | | Log dry biomass pre-thinning | | Log dry biomass post-thinning | |
|---|---|---|---|---|---|---|---|---|---|
| **Region** | **Prov** | **Estimate (SE)** | **Sig** | **Estimate (SE)** | **Sig** | **Estimate (SE)** | **Sig** | **Estimate (SE)** | **Sig** |
| $E_{grat}$ | DUM | 0.32 (0.05) | BC | 2.1 (0.04) | E | 1.9 (0.05) | A | 3.4 (0.06) | A |
| $E_{grat}$ | LGR | 0.39 (0.04) | B | 2.5 (0.03) | C | 1.9 (0.05) | A | 3.4 (0.05) | A |
| $E_{liss}$ east | COO | 0.44 (0.08) | AB | 2.7 (0.05) | A | 1.5 (0.06) | E | 2.9 (0.09) | C |
| $E_{liss}$ east | GOG | 0.09 (0.09) | D | 2.3 (0.06) | CDE | 1.8 (0.06) | AB | 3.5 (0.10) | A |
| $E_{liss}$ east | NOR | 0.60 (0.04) | A | 2.5 (0.03) | BC | 1.7 (0.05) | BC | 3.1 (0.05) | BC |
| $E_{liss}$ west | NAR | 0.01 (0.07) | D | 2.6 (0.05) | AB | 1.8 (0.06) | BC | 3.2 (0.07) | AB |
| $E_{liss}$ west | SNC | 0.08 (0.06) | D | 1.6 (0.04) | F | 1.6 (0.05) | DE | 3.2 (0.07) | AB |
| $E_{liss}$ west | TYG | 0.02 (0.04) | D | 2.3 (0.03) | D | 1.7 (0.05) | CD | 3.1 (0.05) | BC |
| $E_{liss}$ west | WES | 0.15 (0.08) | CD | 1.8 (0.05) | F | 1.5 (0.06) | DE | 3.0 (0.09) | BC |

**Table 5.** Least square means for proportion dual flush (arcsine transform of proportion that flowered twice), foliar cineole content (% green weight) and biomass estimates of orchard pre-thinned (aged 5) and post-thinned (aged 15) at the *Eucalyptus loxophleba* seed orchard with the standard error in brackets. Tukey's tests were performed to test the difference between provenances, means that are similar have the same letter at α = 0.05 significance level (Sig). Provenances are: *DUM* Dumbleyung, *LGR* Lake Grace, *COO* Coolgardie, *GOG* Goongarrie, *NOR* Norseman, *NAR* Narembeen, *SNC* Southern Cross, *TYG* Trayning, *WES* Westonia.

**Effect of families on traits.** There was substantial variation in flowering times within provenances. There was, for instance, about 3 months difference between the commencements of flowering among families derived from parents with Lake Grace, Norseman or Southern Cross provenance (Fig. 4). There was a spread of eight weeks in end of flowering within the individuals from the Lake Grace provenance and nearly 6 weeks range for Goongarrie and Norseman. There was also substantial overlap of flowering between most families although there are a few early or late families which were reproductively isolated from other families in the seed orchard. For instance, Family 8 (from Lake Grace) ended flowering in week 40 while Family 26 (from Goongarrie) started flowering in week 38 (details of the individual tree start and end of flowering times for families 8 and 26 is included in Supplementary Fig. S1). Generally, the peak flowering times of both of these families were out of phase with the peak flowering periods of the other families. The families that flowered the earliest (22, 8, 27 and 23) all started flowering on the twentieth week of the year whereas the last families to start flowering (46, 36, 37

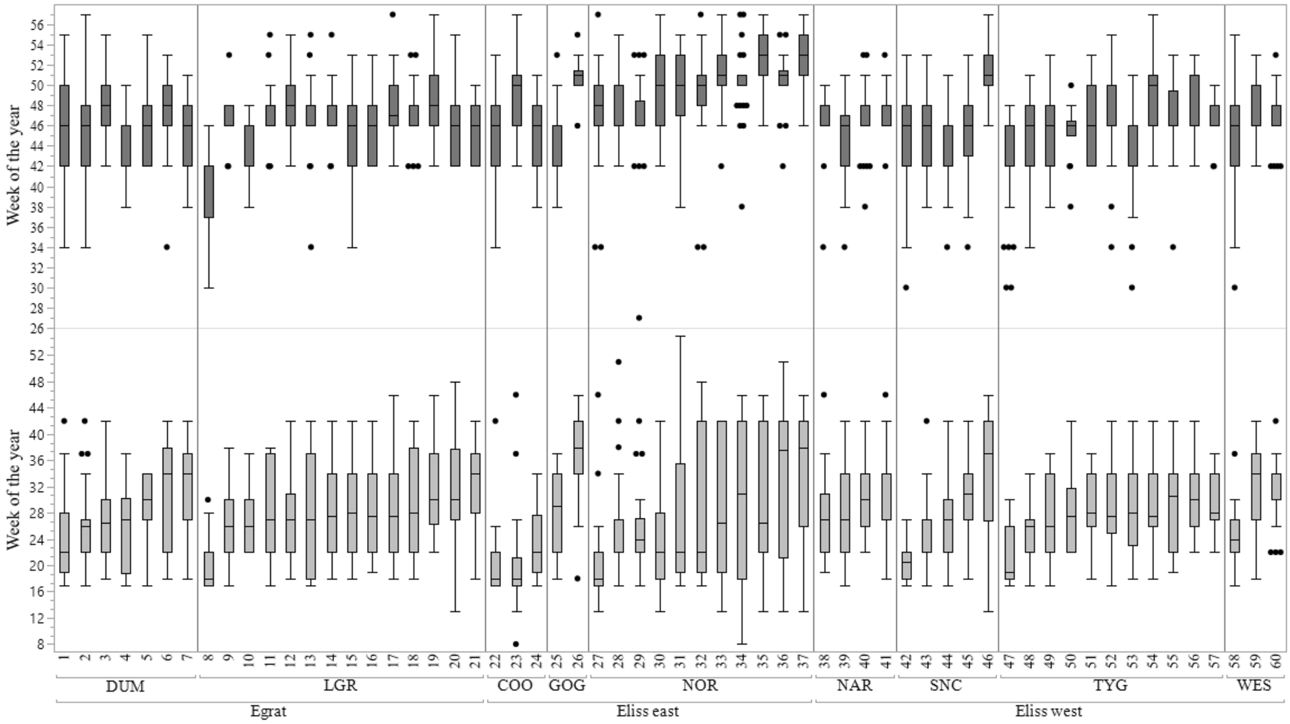

**Figure 4.** The start and end of flowering times (averaged over 2012 and 2014) of *Eucalyptus loxophleba lissophloia* ($E_{liss}$) *E. loxophleba gratea* ($E_{grat}$) families. Light grey box plots represent start flowering week and dark grey boxes plots represent end of flowering times. First week of January is week 1. Line in the middle of each box is the median provenance flowering time and edge of boxes 25th and 75th percentile. Dots represent outliers. *DUM* Dumbleyung, *LGR* Lake Grace, *COO* Coolgardie, *GOG* Goongarrie, *NOR* Norseman, *NAR* Narembeen, *SNC* Southern Cross, *TYG* Trayning, *WES* Westonia. Figure was generated using JMP, Version 14. SAS Institute Inc. 1989–2019 https://www.jmp.com/en_au/home.html.

and 26) started flowering 13–17 weeks later. A similar trend was observed with the end of flowering trait with the earliest families ending flowering in weeks 40–44 (8, 47, 10 and 53) and the latest end flowering families (26, 46, 37 and 35) ended after week 51. Least square means for all flowering traits of each family are detailed in Supplementary Table S2.

Table 6 shows substantial differences in the proportion of trees that flowered, proportion of trees with dual flushes, foliar cineole content and biomass from the families within provenances. The families within provenances that showed wide ranges in the proportion of trees that flowered were from Lake Grace (62–100%), Southern Cross (81–100%), and Dumbleyung (82–100%), while the rest had narrower ranges (90–100%). Similarly, there were families within provenances with considerable variation in dual flush proportions: in families from the Dumbleyung, Lake Grace, Coolgardie and Norseman provenances, the percentage of dual flush trees ranged from < 1 to 69%. For families from Goongarrie, there were no variation (both at 1%), while for the rest of the families within the remaining provenances, trees with dual flush were in the range of 0–7%.

There were two families (46 and 58) that averaged foliar cineole under 0.9% of green leaf weight whereas the top performing families had cineole concentration exceeding 3.0%. Several provenances have tight ranges (little variation among families) while other provenances show wide ranges between families. For example, families within the Narembeen provenance show little variation (2.5–2.7%), whereas those within Westonia are highly variable (0.9–2.6%). Likewise, there is more variation in biomass estimates across both measurements within the Lake Grace provenance compared to Westonia. There was nearly double the average back-transformed biomass from the high yielding families in the pre-thinned assessment (1, 6, 15 and 16 vs 59, 34, 22 and 46) and this difference was more pronounced after thinning (26, 3, 13 and 6 vs 46, 24, 22 and 34).

**Heritability, genetic and phenotypic correlations.** Narrow-sense heritability ranged from high for end of flowering ($\hat{h}^2 = 0.66–0.75$), moderate for start flowering traits ($\hat{h}^2 = 0.45 \pm 0.10$) and foliar cineole content ($\hat{h}^2 = 0.53 \pm 0.09$) and low for duration of flowering and both biomass estimates ($\hat{h}^2 = 0.10–0.33$) (Table 7). Narrow-sense heritability for dual flush flowering was ($\hat{h}^2 = 0.61 \pm 0.16$), propensity to flower in 2012 ($\hat{h}^2 = 0.19 \pm 0.10$) and 2014 ($\hat{h}^2 = 0.24 \pm 0.11$). There were strong genetic correlations between the same flowering traits from different years ($r_g = 0.84–0.96$) but the phenotypic correlations were lower ($r_p = 0.40–0.55$). Genetic correlations were also high between most of the key flowering traits across years except for end of flowering and duration of flowering which generally had high standard errors. The genetic correlations between the start and end of flowering were all positive and above 0.62. The magnitudes of the genetic correlations between start and duration of flowering were also high although these correlations were negative. Phenotypic correlations were generally lower

| Prov | Family | Foliar cineole (SE) | Log dry biomass pre-thinned (SE) | Log dry biomass post-thinned (SE) | Arcsine proportion flowered (SE) | Arcsine proportion dual flush (SE) |
|---|---|---|---|---|---|---|
| DUM | 1 | 1.7 (0.09) | 2.2 (0.08) | 3.4 (0.14) | 1.29 (0.09) | 0.73 (0.14) |
| DUM | 2 | 2.2 (0.10) | 1.8 (0.09) | 3.5 (0.14) | 1.27 (0.09) | 0.33 (0.14) |
| DUM | 3 | 2.0 (0.08) | 2.0 (0.08) | 3.8 (0.14) | 1.28 (0.09) | 0.82 (0.13) |
| DUM | 4 | 2.4 (0.09) | 1.8 (0.09) | 3.3 (0.15) | 1.14 (0.10) | −0.01 (0.16) |
| DUM | 5 | 2.4 (0.08) | 1.9 (0.08) | 3.4 (0.14) | 1.33 (0.09) | 0.08 (0.13) |
| DUM | 6 | 1.7 (0.08) | 2.1 (0.08) | 3.7 (0.14) | 1.46 (0.09) | 0.19 (0.13) |
| DUM | 7 | 2.5 (0.08) | 1.7 (0.08) | 3.0 (0.14) | 1.17 (0.08) | 0.09 (0.13) |
| LGR | 8 | 2.7 (0.09) | 2.0 (0.09) | 3.6 (0.16) | 0.95 (0.09) | −0.01 (0.18) |
| LGR | 9 | 2.3 (0.08) | 1.9 (0.08) | 3.6 (0.14) | 1.54 (0.09) | 0.00 (0.12) |
| LGR | 10 | 2.4 (0.08) | 1.8 (0.08) | 3.4 (0.14) | 1.13 (0.08) | −0.01 (0.15) |
| LGR | 11 | 2.6 (0.08) | 1.8 (0.08) | 2.9 (0.14) | 1.29 (0.09) | 0.63 (0.14) |
| LGR | 12 | 2.9 (0.08) | 1.9 (0.08) | 3.4 (0.14) | 1.39 (0.09) | 0.98 (0.13) |
| LGR | 13 | 2.8 (0.08) | 2.0 (0.08) | 3.8 (0.14) | 1.38 (0.09) | 0.78 (0.13) |
| LGR | 14 | 1.8 (0.09) | 1.7 (0.08) | 2.8 (0.15) | 1.46 (0.09) | 0.58 (0.13) |
| LGR | 15 | 2.8 (0.08) | 2.1 (0.08) | 3.4 (0.14) | 1.21 (0.09) | 0.45 (0.14) |
| LGR | 16 | 2.2 (0.08) | 2.1 (0.08) | 3.6 (0.14) | 1.18 (0.08) | −0.01 (0.13) |
| LGR | 17 | 2.3 (0.08) | 1.9 (0.08) | 3.3 (0.14) | 1.34 (0.08) | 0.52 (0.12) |
| LGR | 18 | 2.7 (0.08) | 1.9 (0.08) | 3.3 (0.14) | 1.21 (0.09) | 0.56 (0.14) |
| LGR | 19 | 2.8 (0.09) | 1.9 (0.09) | 3.4 (0.14) | 1.42 (0.08) | 0.94 (0.12) |
| LGR | 20 | 2.6 (0.08) | 1.8 (0.08) | 3.5 (0.14) | 1.26 (0.09) | 0.08 (0.12) |
| LGR | 21 | 1.4 (0.08) | 1.6 (0.08) | 3.0 (0.15) | 0.91 (0.09) | −0.02 (0.19) |
| COO | 22 | 3.2 (0.10) | 1.4 (0.09) | 2.7 (0.14) | 1.39 (0.09) | 0.37 (0.13) |
| COO | 23 | 2.4 (0.08) | 1.5 (0.08) | 3.2 (0.14) | 1.49 (0.09) | 0.96 (0.12) |
| COO | 24 | 2.5 (0.08) | 1.5 (0.08) | 2.7 (0.16) | 1.38 (0.09) | 0.00 (0.14) |
| GOG | 25 | 2.5 (0.08) | 1.9 (0.08) | 3.2 (0.14) | 1.25 (0.09) | 0.09 (0.12) |
| GOG | 26 | 2.2 (0.08) | 1.8 (0.08) | 3.9 (0.14) | 1.58 (0.09) | 0.09 (0.12) |
| NOR | 27 | 2.7 (0.09) | 1.7 (0.08) | 3.1 (0.14) | 1.45 (0.09) | 0.78 (0.12) |
| NOR | 28 | 2.6 (0.09) | 1.8 (0.08) | 3.6 (0.14) | 1.38 (0.09) | 0.09 (0.13) |
| NOR | 29 | 2.5 (0.08) | 1.9 (0.08) | 3.5 (0.13) | 1.22 (0.08) | 0.22 (0.14) |
| NOR | 30 | 2.5 (0.08) | 1.8 (0.08) | 3.2 (0.15) | 1.53 (0.09) | 0.83 (0.13) |
| NOR | 31 | 2.4 (0.09) | 1.8 (0.09) | 3.1 (0.15) | 1.45 (0.09) | 0.97 (0.13) |
| NOR | 32 | 2.2 (0.08) | 1.7 (0.08) | 2.9 (0.15) | 1.56 (0.09) | 0.52 (0.12) |
| NOR | 33 | 2.4 (0.08) | 1.7 (0.08) | 3.0 (0.14) | 1.49 (0.08) | 0.83 (0.12) |
| NOR | 34 | 2.6 (0.08) | 1.4 (0.08) | 2.6 (0.14) | 1.33 (0.09) | 0.78 (0.13) |
| NOR | 35 | 2.6 (0.08) | 1.7 (0.08) | 2.9 (0.14) | 1.49 (0.09) | 0.60 (0.12) |
| NOR | 36 | 2.5 (0.08) | 1.6 (0.08) | 3.2 (0.14) | 1.50 (0.09) | 0.41 (0.12) |
| NOR | 37 | 2.4 (0.08) | 1.8 (0.08) | 3.1 (0.13) | 1.46 (0.08) | 0.52 (0.12) |
| NAR | 38 | 2.6 (0.08) | 1.7 (0.08) | 3.1 (0.13) | 1.36 (0.09) | −0.01 (0.14) |
| NAR | 39 | 2.7 (0.08) | 1.8 (0.08) | 3.3 (0.14) | 1.52 (0.08) | 0.07 (0.12) |
| NAR | 40 | 2.5 (0.08) | 1.8 (0.08) | 3.1 (0.15) | 1.35 (0.09) | 0.00 (0.14) |
| NAR | 41 | 2.5 (0.08) | 1.8 (0.08) | 3.3 (0.14) | 1.41 (0.09) | −0.01 (0.13) |
| SNC | 42 | 1.5 (0.08) | 1.7 (0.08) | 3.2 (0.14) | 1.40 (0.09) | 0.09 (0.12) |
| SNC | 43 | 2.3 (0.08) | 1.8 (0.08) | 3.4 (0.14) | 1.43 (0.09) | 0.00 (0.12) |
| SNC | 44 | 1.5 (0.08) | 1.7 (0.08) | 3.1 (0.14) | 1.12 (0.09) | 0.11 (0.14) |
| SNC | 45 | 1.9 (0.08) | 1.6 (0.08) | 3.5 (0.15) | 1.40 (0.09) | 0.00 (0.13) |
| SNC | 46 | 0.9 (0.08) | 1.3 (0.08) | 2.8 (0.16) | 1.59 (0.10) | 0.22 (0.14) |
| TYG | 47 | 3.1 (0.09) | 1.6 (0.08) | 2.9 (0.14) | 1.45 (0.09) | 0.00 (0.12) |
| TYG | 48 | 2.0 (0.08) | 1.7 (0.08) | 3.0 (0.14) | 1.58 (0.09) | 0.08 (0.12) |
| TYG | 49 | 2.1 (0.08) | 1.8 (0.08) | 3.4 (0.14) | 1.44 (0.09) | 0.00 (0.12) |
| TYG | 50 | 2.1 (0.09) | 1.7 (0.08) | 3.1 (0.14) | 1.56 (0.09) | −0.01 (0.13) |
| TYG | 51 | 2.0 (0.08) | 1.5 (0.08) | 3.0 (0.17) | 1.51 (0.10) | −0.01 (0.15) |
| TYG | 52 | 2.6 (0.08) | 1.6 (0.08) | 3.3 (0.14) | 1.42 (0.08) | 0.07 (0.12) |
| TYG | 53 | 2.5 (0.09) | 1.7 (0.08) | 3.0 (0.14) | 1.42 (0.09) | 0.00 (0.12) |
| TYG | 54 | 2.3 (0.08) | 1.9 (0.08) | 3.1 (0.14) | 1.34 (0.08) | 0.08 (0.12) |
| TYG | 55 | 2.2 (0.09) | 1.6 (0.08) | 3.3 (0.15) | 1.40 (0.09) | −0.01 (0.13) |
| TYG | 56 | 2.3 (0.09) | 1.6 (0.09) | 3.0 (0.14) | 1.27 (0.09) | 0.00 (0.12) |

Continued

| Prov | Family | Foliar cineole (SE) | Log dry biomass pre-thinned (SE) | Log dry biomass post-thinned (SE) | Arcsine proportion flowered (SE) | Arcsine proportion dual flush (SE) |
|---|---|---|---|---|---|---|
| TYG | 57 | 2.5 (0.08) | 1.7 (0.08) | 3.2 (0.14) | 1.49 (0.09) | − 0.01 (0.12) |
| WES | 58 | 0.9 (0.09) | 1.5 (0.08) | 3.0 (0.16) | 1.38 (0.10) | 0.27 (0.15) |
| WES | 59 | 2.6 (0.09) | 1.5 (0.09) | 3.0 (0.14) | 1.46 (0.09) | 0.19 (0.13) |
| WES | 60 | 1.8 (0.08) | 1.6 (0.08) | 3.0 (0.14) | 1.54 (0.09) | 0.00 (0.12) |

**Table 6.** Least square means for foliar cineole content (% green weight), biomass estimates of pre-thinned (aged 5) and post-thinned (aged 15) orchard, arcsine transformed proportion of flowered and dual flush flowing with the standard error in brackets (SE). *DUM* Dumbleyung, *LGR* Lake Grace, *COO* Coolgardie, *GOG* Goongarrie, *NOR* Norseman, *NAR* Narembeen, *SNC* Southern Cross, *TYG* Trayning, *WES* Westonia.

| | **Genetic correlations** | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | **Start flower 12** | **Start flower 14** | **End flower 12** | **End flower 14** | **Duration flower12** | **Duration flower14** | **Cineole** | **Ln (DBM) pre-thinned** | **Ln(DBM) post-thinned** |
| **Phenotypic correlations** | | | | | | | | | |
| Start flower 12 | **0.446 ± 0.097** | 0.888 ± 0.057 | 0.654 ± 0.109 | 0.739 ± 0.093 | − 0.905 ± 0.034 | − 0.592 ± 0.156 | − 0.099 ± 0.162 | − 0.291 ± 0.173 | − 0.118 ± 0.190 |
| Start flower 14 | *0.549 ± 0.028* | **0.447 ± 0.097** | 0.628 ± 0.115 | 0.797 ± 0.078 | − 0.807 ± 0.086 | − 0.690 ± 0.109 | − 0.276 ± 0.154 | − 0.456 ± 0.157 | − 0.158 ± 0.188 |
| End flower 12 | *0.335 ± 0.041* | *0.336 ± 0.043* | **0.655 ± 0.117** | 0.963 ± 0.030 | − 0.258 ± 0.177 | 0.154 ± 0.206 | − 0.209 ± 0.150 | − 0.211 ± 0.171 | − 0.043 ± 0.184 |
| End flower 14 | *0.309 ± 0.042* | *0.395 ± 0.038* | *0.540 ± 0.035* | **0.746 ± 0.123** | − 0.396 ± 0.159 | − 0.118 ± 0.205 | − 0.244 ± 0.147 | − 0.227 ± 0.168 | − 0.038 ± 0.181 |
| Duration flower12 | *− 0.931 ± 0.006* | *− 0.476 ± 0.031* | *0.017 ± 0.046* | *− 0.136 ± 0.044* | **0.328 ± 0.083** | 0.843 ± 0.114 | − 0.001 ± 0.173 | 0.256 ± 0.185 | 0.126 ± 0.200 |
| Duration flower14 | *− 0.377 ± 0.034* | *− 0.814 ± 0.015* | *− 0.014 ± 0.044* | *0.191 ± 0.040* | *0.400 ± 0.034* | **0.186 ± 0.063** | 0.127 ± 0.194 | 0.493 ± 0.185 | 0.240 ± 0.217 |
| Cineole | *− 0.003 ± 0.048* | *− 0.061 ± 0.048* | *0.226 ± 0.034* | *0.171 ± 0.033* | *− 0.032 ± 0.047* | *0.030 ± 0.046* | **0.526 ± 0.087** | 0.209 ± 0.160 | 0.180 ± 0.167 |
| Ln (DBM) pre-thinned | *− 0.114 ± 0.036* | *− 0.054 ± 0.037* | *0.036 ± 0.037* | *0.125 ± 0.031* | *0.131 ± 0.037* | *0.034 ± 0.038* | *− 0.015 ± 0.02* | **0.096 ± 0.025** | 0.763 ± 0.091 |
| Ln (DBM) post-thinned | *− 0.139 ± 0.042* | *− 0.123 ± 0.041* | *0.037 ± 0.045* | *− 0.048 ± 0.044* | *0.164 ± 0.041* | *0.080 ± 0.041* | *0.047 ± 0.045* | *0.678 ± 0.017* | **0.288 ± 0.075** |

**Table 7.** Heritability, genetic and phenotypic correlations for *Eucalyptus loxophleba* seed orchard. Narrow-sense heritability (± standard errors) of each trait is on the diagonal (in bold), above the diagonal is genetic correlation (± standard errors) and below the diagonal is phenotypic correlations (± standard errors) in italics. Traits are start flowering week in 2012 and 2014, end of flowering week in 2012 and 2014, duration of flowering in 2012 and 2014, foliar cineole concentration by weight, ln (dry biomass) of the pre-thinned orchard as measured in 2004 and ln (dry biomass) of post-thinned seed orchard as measured in 2014.

than genetic correlations, but they had lower standard errors. There were high negative phenotypic correlations between start of flowering 2012 and duration of flowering in 2012 ($r_p = -0.93$) and in 2014 ($r_p = -0.81$).

The correlations were of similar magnitude to the standard errors for many of the biomass and cineole estimates, except between the two biomass estimates. There were however, moderate genetic correlations between flowering duration in 2014 and pre-thinned biomass, and weak genetic correlations between start and end of flowering time in 2014 and foliar cineole concentration. In contrast, there was a weak positive phenotypic correlation between cineole and of end of flowering in 2012 and 2014. There were also weak negative phenotypic correlations between post-thinned biomass and start of flowering. There was a moderate negative genetic correlation between pre-thinned biomass and start of flowering in the 2014 assessment and a corresponding positive weak phenotypic correlation end of flowering for 2014.

## Discussion

Understanding flowering phenology is critical in seed orchard design, as synchronicity of flowering amongst parents is key to maximising outcrossing, genetic quality of the seed, and its ability to deliver traits of commercial interest. The results from this study displayed large variation in flowering phenology within and between subspecies, provenances and families. With the exception of a few families, there was a high level of synchronicity of flowering within the seed orchard but substantial differences between provenances. However, caution should be used when directly comparing the provenance-level data as there are different numbers of families in each provenance; for example, Coolgardie, Goongarrie and Westonia were poorly represented with a small number of families.

The large range of commencement and end of flowering times within provenances may in part be reflected by the percentage of dual-flush trees in each provenance. In most cases, dual flush trees started flowering earlier (i.e. the first flush) in the year than their single flush contemporaries, resulting in greater flowering duration. These were the most of the out-of-phase trees in the orchard. We estimate that only a small proportion (< 4%) of the total buds in the orchard flowered out of phase with the single flush trees. Early dual flush trees which were active before the main flush of the orchard will have a greatly reduced opportunity to outcross with other trees in the seed orchard.

After the $E_{liss}$ eastern provenances the southern $E_{grat}$ provenances had the second highest proportion of dual flush trees. There is no evidence that the two regions exhibiting high rates of dual flush flowering is due to their relatedness. Investigation on the chloroplast DNA has revealed that $E_{grat}$ is more closely related to the neighbouring $E_{liss}$ western and *E. loxophleba* subsp. *loxophleba* than to $E_{liss}$ eastern provenances[53]. However, the high heritability of the dual flush trait ($\hat{h}^2 = 0.61 \pm 0.15$) suggests that selections could be made to reduce its prevalence in breeding populations. A large percentage of these dual flush trees were $E_{grat}$, and based on this alone, it may be appropriate to separate $E_{grat}$ into a separate breeding population. The taxonomic split between $E_{liss}$ and $E_{grat}$ is contentious[54,55], however, a breeding population comprised of $E_{grat}$ alone would allow more out-crossing of these dual flush trees especially at the start and end of the annual reproductive cycles. Considering $E_{liss}$ has been shown to have weak reproductive barriers[38–42], it would allow the subspecies to be planted in their natural range thereby minimising genetic 'pollution' of other subspecies.

The dual flush $E_{liss}$ trees, most of which originate from eastern provenances, flowered out of phase with the single flush $E_{liss}$. Many of these families also performed poorly in biomass assessments. For example, the Coolgardie provenance (families 22, 23 and 24) ranked last for biomass in the two measurements of this trial, flowered considerably earlier than any other family and nearly 30% of the trees flowered twice. Family 22 had the highest foliar cineole concentration, but ranked second and third last for biomass and was the earliest family to flower. Some families in the Norseman provenance, in contrast, included seven of the latest ten flowering families with a high proportion of dual flush trees and below average biomass. High cineole families may be kept in the seed orchard to maintain cineole levels, but seed should not be collected from these families for biomass plantings and they should be carefully considered before inclusion in future breeding programmes. Families with a high proportion of dual flush trees, low cineole and biomass rank, could be removed from the breeding populations. The other $E_{liss}$ eastern provenance, Goongarrie, consisted of two families, both with a low proportion of dual flush trees. Family 26 was the fourth latest to flower, but this family ranked highest in biomass in the second assessment and family 25 flowered during peak time and ranked well for both biomass and cineole. Thus, although $E_{liss}$ east provenances were on average poor performers in the seed orchard, some families from the eastern provenances may be candidates for next-generation seed or clonal orchards.

Mazanec et al.[18] reports on the variation of biomass from different provenances from three large $E_{liss}$ progeny trials across southern Australia. Each trial consisted of nine provenances with at least 13 families. It was found that the progeny from Norseman ranked last for biomass at two of the three trials and the progeny from Cardunia Rocks, the other eastern provenance, performed average or below at the three trials. These trials were measured at age three and gives further support for the poor biomass performance from the $E_{liss}$ eastern range. However, short-duration studies can at times fail to indicate longer-term performance and further measurements of these trial should reveal more about the top biomass producing $E_{liss}$ provenances.

This study shows that, with the exception of a few families, there is a considerable proportion of trees flowering in the seed orchard throughout most months the year. However, the amount of reproductive activity is uneven; the rate of anthesis is much more pronounced in late winter and spring when it is common for 30–50% of buds to have commenced flowering in a 1-month period. This contrasts with other studies of southern *Eucalyptus* species. For example, warmer temperatures are known to trigger key developmental stages in *E. nitens* including bud initiation and growth[56] and heat-sum has been found to be the main driver of anthesis time in *E. globulus* and *E. nitens*[26,57]. With only 2 years of data, the effect that heat sum had on the timing of anthesis for *E. loxophleba* is impossible to determine. The year of greater heat did correspond with earlier flowering and shorter flowering duration, both expected outcomes in the heat sum model. However, $E_{liss}$ and $E_{grat,}$ in contrast to *E. nitens* and *E. globulus*, are adapted to arid conditions.

Moisture availability and rainfall events have been identified as important factors affecting anthesis for plants in arid environments[58–62] and that is a likely cause of slow reproductive development of $E_{liss}$ and $E_{grat}$ during late summer and autumn. Furthermore, Friedel et al.[59] demonstrated that soil moisture was a predictor of flowering for 46 arid-zone species although flowering events lagged between 1 and 9 months after rainfall. This is consistent with the flowering of $E_{liss}$ and $E_{grat}$ in this study where there were no significant rainfall events in 2012 until May and in 2014 until April. $E_{liss}$ and $E_{grat}$ are well adapted to drought and appear to exhibit drought-induced dormancy of the crown.

The plastic nature of the species recorded in the arid zone of Australia by Friedel et al.[59], Davies[61] and others suggest that eastern provenances of $E_{liss}$ may be more strongly adapted to rainfall-induced growth and reproduction than western provenances. The multiple reproductive events observed in this trial may be a function of that adaptation. It is possible that trees from the low rainfall eastern provenances were responding to the higher rainfall experienced near Lake Toolibin. However, the two most southerly provenances ($E_{grat}$) also had a high percentage (20%) of dual flushes trees and are the two closest provenances to Lake Toolibin. $E_{grat}$ was less reproductively active, suggesting that these provenances did not benefit from the slightly higher rainfall of the study site when compared to their natural range. In contrast, western and eastern $E_{liss}$ provenances displayed high levels of reproductive activity at a site with higher rainfall and lower evaporation compared to their natural distribution. There was however, no trend in northern provenances to flower earlier than the more southerly $E_{grat}$ provenances, a trend recorded for *E. marginata*[63] and for *Corymbia citriodora* subsp. *variegata* and *C. maculata*[27]. However, the opposite trend has been observed in *E. globulus* where Victorian provenances flowered later than the more southerly eastern-coast Tasmanian provenances[26].

Biomass generally has a lower heritability than either flowering traits or cineole production and this was observed in this trial. The pre-thinned biomass heritability was low ($\hat{h}^2 = 0.10 \pm 0.03$) but increased after thinning to $0.29 \pm 0.08$ suggesting that estimates of additive genetic variance for this trait were biased upwards as a result. The difference between the two estimates suggests that some degree of bias was introduced as a result of selective thinning. We have no way of knowing if the bias extends to the flowering traits; and therefore, some caution should be used in interpretation of these results. However, Mazanec et al.[18] found narrow-sense heritably at three $E_{liss}$ progeny trials of 0.19 ± 0.05, 0.13 ± 0.04 and 0.25 ± 0.05 at Monarto (South Australia), Condobolin (New South Wales) and Brookton (Western Australia) respectively. Stem diameter, an accurate estimator of biomass, has been found to be moderately heritable in other eucalyptus species including *E. nitens* $\hat{h}^2 = 0.18–0.19$[64,65], and *E. cladocalyx* $\hat{h}^2 = 0.14$[66] and $\hat{h}^2 = 0.30$[67].

Heritability of flowering traits in this study were under moderate to strong genetic control. These results are consistent with results from other studies in eucalypts. Jones et al.[26] reported high broad-sense heritability for peak anthesis time ($\hat{H}^2 = 0.78$) in an *E. globulus* clonal seed orchard but found weak heritability for duration of flowering ($\hat{H}^2 = 0.09$) with the low heritability attributed to the correlation between duration of flowering and flower abundance[68]. For the same species, Gore and Potts[25] found narrow-sense heritability over 0.64 for start, peak and end of flowering after a single year of assessment. Flowering intensity of *E. cladocalyx* has been recorded as $\hat{h}^2 = 0.48$[69] and $\hat{h}^2 = 0.52$[66]. The number of reproductive flushes was under strong genetic control ($\hat{h}^2 = 0.61 \pm 0.16$) but the propensity to flower in 2012 and 2014 were quite low $\hat{h}^2 = 0.19 \pm 0.10$ and $\hat{h}^2 = 0.24 \pm 0.11$ respectively, although this is much higher than the $\hat{h}^2 = 0.06 \pm 0.05$ reported for *E. globulus*[26].

Foliar cineole concentration was observed to be under strong genetic control ($\hat{h}^2 = 0.53 \pm 0.09$). Mazanec et al.[18] found similar narrow-sense heritability at an $E_{liss}$ progeny trial in Brookton, Western Australia, of over 1700 trees from 126 families ($\hat{h}^2 = 0.53 \pm 0.07$) and from an $E_{grat}$ progeny trial of 90 families of ($\hat{h}^2 = 0.50 \pm 0.08$)[70]. Similar heritability was found in *E. camaldulensis*[29]. Heritability as high as $\hat{h}^2 = 0.83$ was found in a small *E. kochii* seed orchard[71]. In the current study, the parent trees had been tested for foliar cineole concentration prior to selection with seed only sourced from elite individuals (foliar cineole concentration > 2.5%) so selection was biased to high cineole individuals. This is because at that time, in the genetic selection of this species, cineole was considered likely to be the major product for commercial planting of these species[8,11]. The two more recent $E_{liss}$ and $E_{grat}$ progeny trials mentioned above were not subject to pre-selection for foliar cineole but the heritability results were similar suggesting that pre-selection for high cineole did not influence estimates of additive variance in this trial.

The strong genetic and phenotypic correlations between duration and start of flowering have been found in other species, for instance, *Lythrum salicaria* ($r_p = -0.92$)[72]. The correlations for $E_{liss}$ are much higher when comparing within years than across years and may be attributable to the annual variation in dual flush flowering which exhibit longer duration of flowering. It is unknown if the trees that flowered twice in 2014 also did so in 2012 and although the heritability for this trait is high, another assessment following individual reproductive flushes would be useful. Furthermore, assessment of the performance of progeny from the first-and second-flush of the dual flush trees and single flush trees could indicate the degree of inbreeding or selfing.

Genetic correlations suggest that selection for high biomass and cineole may result in selection of early flowering trees. For example, if selection is biased toward biomass then the low genetic correlation for that trait with flowering time will allow greater freedom in selection of trees with later flowering times. The orchard was initially conservatively thinned, in most cases an individual from each family was retained from each replicate. Poor biomass producing $E_{liss}$ east families with high levels of dual flush flowering could be eliminated from the orchard which would increase flowering synchronicity without negatively impact biomass production.

## Conclusion

This study has shown that timing of anthesis is strongly influenced by genetic factors. Genetically, there is a large amount of variation with broad-scale differences among provenances and families. Most flowering traits, along with cineole, were moderately to strongly heritable whereas biomass heritability was low. At the start and end of the annual flowering cycle, dual flush trees were reproductively isolated and because the trait is strongly heritable, this could result in greater flowering asymmetry in progeny collected from the dual flush trees. Dual flush flowering was evident in the $E_{grat}$ provenances. For this reason and because of the potential spread of genetic material to native stands through pollen dispersal, $E_{grat}$ should be treated as a separate breeding population for use within its natural range. The $E_{liss}$ east provenances included families with the highest proportion of dual flush trees and the poorest biomass yield. These families should be eliminated from the general breeding programme. Due to the moderate to negligible genetic correlations between flowering traits, biomass and foliar cineole concentrations, selections should be based on biomass and cineole. Families with a high proportion of dual flush trees and otherwise desirable characteristics should be further studied to determine if this was a one-off event or if it is a recurring phenomenon. Opportunistic use of available soil moisture may be responsible for dual flush flowering in eastern provenances, however, the 2-week divergence in flowering time between assessment years may be driven by heat-sum.

## Data availability

The flowering key traits (start, end and duration of flowering), biomass estimates and foliar cineole data from this manuscript has not been achieved, but prior to publication it will be published on CSIRO data access portal https://data.csiro.au/dap/discoveryService with a DOI number.

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## Author contributions

R.M. conceived the idea of the experiment. B.S. carried out the experiment and drafted the manuscript. R.M. and A.Z. commented on the statistical models. R.M., A.Z., A.A. and M.G. contributed to the final version of the manuscript.

## Competing interests

The authors declare no competing interests.

## Additional information

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# Chapter 5

## Genetic parameters of sapling coppice biomass

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ORIGINAL ARTICLE

# Sapling and coppice biomass heritabilities and potential gains from *Eucalyptus polybractea* progeny trials

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## Abstract

*Eucalyptus polybractea* has been planted as a short-rotation coppice crop for bioenergy in Western Australia. Historical breeding selections were based on sapling biomass and despite a long history as a coppice crop, the genetic parameters of coppicing are unknown. Here, we assessed sapling biomass at ages 3 and 6 from three progeny trials across southern Australia. After the second sapling assessment, all trees were harvested. Coppice biomass was assessed 3.5 years later. Mortality following harvest was between 1 and 2%. Additive genetic variance for the 6-sapling estimate at one site was not significant. Sapling heritabilities were between 0.06 and 0.36 at 3 years, and 0.18 and 0.20 at 6 years. The heritability for the coppice biomass was between 0.07 and 0.17. Within-site genetic and phenotypic correlations were strong between all biomass assessments. Cross-site correlations were not different from unity. Selections based on net breeding values revealed positive gains in sapling and coppice biomass. Lower or negative gains were estimated if 3-year sapling selections were applied to the coppice assessments (−7.1% to 3.4%) with useful families culled. Positive gains were obtained if 6-year sapling selections were applied to the coppice assessment (6.4% to 9.3%) but these were lower than those obtained by applying coppice selections to the coppice assessment (8.4% to 14.8%). Removal of poor performing families and families that displayed fast sapling growth rates but under-performed as coppice will benefit potential coppice production. These results indicate that selections should be made using coppice data.

**Keywords** Coppice biomass · *Eucalyptus polybractea* · Net breeding values · Heritability · Sapling biomass · Genetic and phenotypic correlations · Selection simulation

## Introduction

A suite of mallee *Eucalyptus* sp. were selected for short-rotation coppice crops in Western Australia (WA) for essential oils, bioenergy and biofuel feedstocks (Bartle and Abadi 2010; Davis 2002; McGrath et al. 2016). Species selections were based on the ability to coppice vigorously after harvest with high concentration of foliar 1,8-cineole (Hobbs et al. 2009). One of the successful candidates for development was *Eucalyptus polybractea* R.T. Baker which has been harvested on a 2-year cycle in Victoria and New South Wales (NSW) for high-grade eucalyptus oil production for over a century (Davis 2002; Goodger et al. 2007). Agroforestry trials in WA have demonstrated that *E. polybractea* is capable of high productivity when grown on sites with acidic deep soil profiles with low salinity (Spencer et al. 2019; Wildy et al. 2000). In 1993 an *E. polybractea* breeding programme was initiated with three small progeny trials (Bartle et al. 1998). Additional trials were added in later years and included progeny from 100 families using seed from wild parents with cineole concentrations of >2.5% (Mazanec et al. in press).

In order to maximise the potential of *E. polybractea* for cineole production, breeding focused on gains in foliar 1,8-cineole concentration for the high-grade eucalyptus oil market (Boland et al. 1991; Coppen 2002; Davis 2002). The market for eucalyptus oil with high levels of 1,8-cineole did not materialise and the potential for biomass for bioenergy (Abdullah and Wu 2009; McGrath et al. 2016; Wu et al. 2008) emerged as a greater potential avenue for profit (Bartle and Abadi

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2010). Breeding decisions for *E. polybractea* on biomass performance have historically been based on assessment of sapling performance although the relationship between sapling and coppice biomass is unknown. Studies in other eucalypt species have revealed a wide range of genetic correlations between sapling and coppice biomass ranging from weak (Whittock et al. 2003) to very strong (Amâncio et al. 2020; Li et al. 2012). This variation in genetic correlations may be underpinned by the genetic control of lignotuber development, which has been found to be responsible for the varying degrees of success of coppicing (Bortoloto et al. 2020; Walters et al. 2005; Whittock et al. 2003).

The mallee species developed as coppice crops in WA, in contrast, show very high rates of reshooting after harvest (Eastham et al. 1993; Milthorpe et al. 1998; Spencer et al. 2019) suggesting that lignotuber development in these species is less variable. For example, resprouting from the lignotubers, *E. kochii*, does not appear to be limited by either the number of meristematic foci or starch reserves, but rather a loss of fine root material and reduction of thickening of structural rootstock following too frequent harvesting (Wildy and Pate 2002). Subsequent work across four trial-plantings has demonstrated that for *E. polybractea*, a rotation length of 3 years is appropriate for sustainable biomass production (Spencer et al. 2019). However, greater gains in biomass production may be realised if a breeding programme is focused on coppice productivity following harvest.

The heritability of desirable traits underpins the accuracy of selection of elite individuals or families for genetic gain. If multiple traits are of interest, it is of critical importance to understand the genetic relationships between them, as selection for one trait may result in correlated gains or losses in another (Isik et al. 2017). For instance, Milthorpe et al. (1998) observed that selection of individuals based on foliar eucalyptus oil concentration alone may reduce total oil production, and for two subspecies of *E. loxophleba*, weak negative correlations have been observed between biomass and foliar cineole concentrations (Mazanec et al. 2020; Mazanec et al. 2017). Currently, there are no published estimates of genetic parameters for *E. polybractea* pertaining to coppice biomass. Knowledge of the genetic correlations between sapling and coppice biomass production at different ages as well as between subsequent coppice cycles is essential for determining the optimal time for selection for biomass production. Eliminating inferior genotypes will improve both biomass and 1,8-cineole yields and will assist in the economic viability of any future *E. polybractea* industry.

This paper reports the results of a study investigating (1) the heritability of coppicing success and biomass production, (2) the correlations between sapling and coppice biomass across three progeny trials established across southern Australia and (3) the optimal timing of selection through simulations of trial thinning for conversion to seed orchards and establishment of clonal orchards.

## Methods

### Study sites

Three progeny trials were planted in 2009 at the Condobolin Agricultural Research Station in NSW (33.07° S, 147.24° E), Drummartin in Victoria (36.44° S, 114.43° E) and at the Newdegate Research Station in WA (33.12° S, 118.82° E). All sites have mild winters and hot summers with Drummartin and Newdegate receiving most of their rainfall in the winter months while rainfall at Condobolin is more evenly distributed throughout the year (Table 1). Climate and elevation data were sourced from the Australian Bureau of Meteorology stations located at the Condobolin and Newdegate Research Stations and Drummartin town site. The soil at Newdegate is duplex with yellow sand over clay, Drummartin is a duplex with grey-brown loam over sodic red clay while at Condobolin, the soil is gradational red and brown earths (Mazanec et al. in press).

Each of the three progeny trials consisted of the same 66 open-pollinated families. All parent trees were randomly selected from native stands in the region of West Wyalong in NSW. The trials were planted in a Latinised row-column design with families randomly assigned to four-tree row plots. There were six replicates at Condobolin and Drummartin. However, space constraints at the Newdegate trial required a configuration with four replicates and five-tree row plots. Planting spacing of 1.5 m was applied within each row, and rows were 3 m apart. Each trial had a two-tree buffer surrounding the entire trial using the same species with unknown progeny.

In the spring of 2016, each trial, including buffer trees, was cut as close to ground level as possible using chainsaws.

**Table 1** Average climatic conditions during the trial period (2009 to 2020) at the three progeny trial locations. Climatic and elevation data were obtained from Bureau of Meteorology (SILO 2020)

| Trial location | Annual rainfall (mm) | Annual evaporation (mm) | Maximum temperature (°C) | Minimum temperature (°C) | Elevation (m) |
|---|---|---|---|---|---|
| Condobolin | 491 | 2245 | 27.8 | 11.9 | 195 |
| Drummartin | 471 | 1720 | 25.2 | 10.0 | 115 |
| Newdegate | 359 | 1904 | 26.3 | 9.6 | 320 |

## Biomass assessments

Each trial was measured twice before harvest, once when the trials were in their third year (spring 2012 to autumn 2013) and again in spring 2016 when the saplings were about 6 years. Coppice post-harvest was measured in autumn 2020. For the initial sapling and coppice assessments, the height and crown widths (in two perpendicular directions) of each mallee were measured to the closed 5 cm using a surveyor's staff. The Crown Volume Index (CVI) was calculated as the product of those three measurements as described in Spencer et al. (2019). At the initial measurement at Drummartin, both stem basal area (SBA) and CVI were measured. The SBA method was used in the second assessment (2016) where the stems of each mallee were measured at 10 cm above ground level with a diameter tape. Where there were multiple stems, each diameter was converted cross-sectional stem areas and was summed to give the total cross-sectional stem areas as described by Huxtable et al. (2012).

After the 3-year sapling measurement, between 46 and 53 buffer trees, representative of the size of the trial trees, were measured and then destructively sampled. After the second biomass assessment, an additional 40 trees were destructively sampled from each trial. Linear regressions relating SBA or CVI to above-ground biomass were used to estimate above-ground tree biomass. The regression parameters for the initial assessment are detailed in Mazanec et al. (in press). For the coppice assessment, species-specific *E. polybractea* coppice biomass allometric equations were used to estimate above-ground green biomass (Spencer et al. 2019).

## Data analysis

Allometric equations between SBA and biomass were developed using SAS Proc Reg on natural log transformed data (SAS 2017). The equations took the form:

$$\ln(y) = \ln(a) + b\ln(x) + \varepsilon \qquad (1)$$

where $y$ is the above-ground green biomass, $x$ is the SBA, $a$ is the intercept, $b$ is the slope and $\varepsilon$ denotes the model residuals. To remove bias inherent to back-transforming from logarithmic to original scale, the Baskerville (1972) correction was applied.

Analysis of heritability and genetic parameters was conducted using ASReml 4.1 (Gilmour et al. 2015) using the following linear mixed model:

$$\boldsymbol{Y} = \boldsymbol{Xb} + \boldsymbol{Zu} + \boldsymbol{e} \qquad (2)$$

where $\boldsymbol{Y}$ is the phenotypic vector of observation, $\boldsymbol{X}$ is the fixed effects design matrix, $\boldsymbol{b}$ is the vector of fixed effects, $\boldsymbol{Z}$ is the random effects design matrix, $\boldsymbol{u}$ is the vector of the random effects and $\boldsymbol{e}$ is the vector residual error. Terms in $\boldsymbol{b}$ included the intercept and population effects while $\boldsymbol{u}$ included replicate, long column, row within replicate, column within replicate, plot and family effects.

Within the framework of the linear mixed model, we conducted three classes of analysis, as detailed in the following sections.

## Univariate family model analyses

Univariate analyses, for each trial, were conducted to determine requirements for transformation prior to estimation of the fixed and random effects symbolised in $\boldsymbol{b}$ and $\boldsymbol{u}$ of Eq. 2. Terms in $\boldsymbol{b}$ included the intercept and population effects and terms in $\boldsymbol{u}$ included replicate, long column, row within replicate, column within replicate, plot and family effects. Residual plots were examined for heteroscedasticity. Mean-variance relationships and potential transformations were assessed using the slope of the log(absolute residual) on log(predicted value) as outlined by Gilmour et al. (2015). All biomass data required transformation. The 3-year sapling biomass was transformed to $x^{0.3}$ where $x$ = 3-year-old sampling biomass (Mazanec et al. in press), the 6-year-old sapling biomass was subjected to $y^{0.0}$ where $y$ = 6-year-old sapling biomass while coppice biomass was transformed using $z^{0.25}$ where $z$ = coppice biomass. Univariate genetic parameters of the presences/absence of coppice post-harvest were analysed using the logit link function in ASReml with the same model specifications.

## Univariate individual tree model

Once appropriate transformations were determined and applied, a univariate individual tree model was used to estimate additive genetic variances for biomass from each trial. Cases with significant additive genetic variance provided initial estimates of heritability and served as checks for subsequent analysis. Griffin and Cotterill (1988) observed that mixed mating systems in open-pollinated eucalypts may result in inflated heritability estimates. They suggested the use of a coefficient of relationship of $\rho$ = 1/2.5 when estimating heritability to adjust for selfing rates of about 30%. Bush et al. (2011) affirmed that this methodology was applicable in first-generation open-pollinated eucalypts. Recently, Kainer et al. (2018) used the above coefficient of relationship when estimating heritability in *E. polybractea*. In our analysis, we also applied a selfing rate of 30% in ASReml to appropriately adjust additive variance estimate. Terms in $\boldsymbol{u}$ and $\boldsymbol{b}$ were the same between the family and individual tree models with the exception that the family term was replace by an individual tree random additive effect. The additive genetic variance between relatives was modelled via the numerator relationship matrix (Henderson 1976). The significance of individual variance components was checked using a one-tailed log likelihood ratio test with 0.5 degrees for freedom (Gilmour et al. 2015).

### Bivariate analyses

For this model, measurements of trees in different years were treated as different traits for the purpose of estimating genetic correlations. Terms in ***b*** and in ***u*** were the same as the univariate individual tree model, nested within trait. Estimated variances and covariances were used to calculate genetic correlations between the various biomass assessments. The genetic correlations were checked for significant deviation from zero and unity. To test for significant difference from zero, the correlation was constrained to zero and a two-tailed log likelihood ratio test with 1 degree of freedom was used. To test for significant difference from unity, the correlation was constrained to one and a one-tailed log-likelihood ratio test with 0.5 degrees of freedom was used.

### Univariate cross-site analyses

For this model, terms in ***b*** included intercept and the site effects while terms in ***u*** were identical to the univariate individual tree model nested within site. Design effects and site variances were assumed independent between sites and genetic variances were assumed heterogenous. This model was used to estimate reported narrow-sense heritability for each site and cross-site genetic correlation. Cross-site genetic correlations were estimated on a pairwise basis and then checked for significant variation from zero and unity as described above.

An expanded version of the cross-site model was used to include family effects and site by family interaction effects as fixed terms in ***b***. Design elements in ***u*** were as described above. This model was used to generate best linear unbiased estimates (BLUEs) for populations and families.

Narrow-sense heritability was estimated using the following formula:

$$\widehat{h}^2 = \frac{\sigma_a^2}{\left(\sigma_a^2 + \sigma_p^2 + \sigma_e^2\right)} \tag{3}$$

where $\hat{h}^2$ is the narrow-sense heritability, $\sigma^2{}_a$ is the additive variance, $\sigma^2{}_p$ is the plot variance and $\sigma^2{}_e$ is the error variance.

Genetic and phenotypic correlations were calculated using the following formula:

$$r = \frac{\sigma_{12}^2}{\sqrt{\sigma_1^2 * \sigma_2^2}} \tag{4}$$

where $r$ represents either the genetic correlation ($r_g$) or the phenotypic correlation coefficient ($r_p$) and $\sigma^2{}_{12}$, $\sigma^2{}_1$ and $\sigma^2{}_2$ represent either the additive genetic covariance and additive genetic variances or phenotypic correlation and phenotypic variances, respectively.

### Net breeding values

Simulations of thinning trials for conversion to seed orchards were compared between years and between sapling and coppice on the basis of biomass. In order to evaluate the effect of selection at different sapling ages and between sapling and coppice, net breeding values (NBVs) for the three biomass assessments were estimated for each individual tree at each trial. For each assessment, a selection scheme using the NBV was applied to each trial for each sapling and coppice measurement, which simulated thinning the trials for conversion to seed orchards. In this process, the single individual with the highest NBV in a family plot was selected for retention if its NBV was above the mean NBV for the trial under consideration. If no trees in a plot satisfied that criterion, then no trees were retained in that plot. An additional selection scenario was performed to simulate selection for a clone orchard which included very high selection intensities of the best ten unrelated individuals at each trial.

For individual sites, NBV included the intercept, site, population and additive effects. Selection was conducted on the back-transformed scale and genetic gains were calculated using the percentage difference between the back-transformed mean NBV of the selected trees and of mean NBV of the trial.

To observe the impact on genetic gain of selection at the sapling stage, gain was recalculated using NBVs for coppice on the 3- and 6-year saplings' assessment and compared to the gain estimated when selecting for only coppice. Estimated genetic gains apply to redeployment of seedlings on the same site at which selections were made (Mazanec et al. in press).

Preliminary analysis indicated that additive variance for the 6-year sapling estimates at Drummartin was not significant ($P > 0.05$); therefore, heritabilities, genetic correlations and NBVs pertaining to that year were not estimated.

## Results

### Estimation of biomass

The allometric relationship (Eq. 1) between stem basal area and above-ground fresh biomass for the 6-year saplings was highly significant for each trial ($P < 0.0001$) (Table 2). Equations for 3-year sapling assessment are detailed in Mazanec et al. (in press), and for coppice biomass estimates, the species-specific *E. polybractea* equation was used from Spencer et al. (2019).

**Table 2** Statistics of the site-specific allometric models that were used to estimate above-ground fresh biomass from stem basal area at the three progeny trials. Biomass ranges of sample trees included in the models are also shown. MSE is mean square error and $R^2$ is the coefficient of determination

| Location | Biomass range (kg) | Slope | Intercept | MSE | $R^2$ |
|---|---|---|---|---|---|
| Newdegate | 0.2–119.1 | 1.195 | −1.709 | 0.025 | 0.969 |
| Drummartin | 2.4–126.5 | 1.124 | −1.415 | 0.032 | 0.974 |
| Condobolin | 0.5–84.0 | 1.105 | −1.182 | 0.048 | 0.966 |

## Trial biomass estimates

The back-transformed mean tree biomass at each of the three biomass assessments is given in Table 3. Growth rates were highest at the first assessment at Condobolin with a significant location effect ($P < 0.001$) and slowest growth at the Newdegate trial, but there appeared to be reversal of growth rates so that no significant differences were evident at the second measurement. Significant differences emerged again ($P < 0.001$) following the coppice assessment with faster growth rates at the Drummartin trial while rates at the other two sites were similar.

## Mortality post-harvest

Post-harvest survival was very high across the three trials, ranging from 97.9% at Condobolin to 99.5% at Newdegate (Table 4). Across the 66 families, this ranged from 100% survival for 36 families to 94% for the worst performing family (Table S1). Due to the very high survival at each trial and the resulting insignificant additive variance for coppicing ability, heritability of survival post-harvest could not be estimated for each trial.

The untransformed phenotypic mean weight of the 6-year sapling estimates at Newdegate, Drummartin and Condobolin was 31.5, 28.2 and 28.7 kg while the corresponding phenotypic means for saplings that failed to coppice were 10.7, 15.3 and 11.4 kg at each trial, respectively.

**Table 3** Back-transformed best linear unbiased estimates (BLUEs) of average tree biomass (kg) across the three *E. polybractea* progeny trials at each biomass assessment

| Location | 3-year sapling | 6-year sapling | Coppice |
|---|---|---|---|
| Newdegate | 5.2 | 26.5 | 11.8 |
| Drummartin | 8.5 | 22.9 | 26.0 |
| Condobolin | 11.5 | 24.5 | 11.0 |

**Table 4** The number of saplings and coppice at each trial and the proportion that coppiced after harvest

| Location | No. of saplings | No. of coppice | % coppiced |
|---|---|---|---|
| Newdegate | 1117 | 1111 | 99.5 |
| Drummartin | 1290 | 1281 | 99.3 |
| Condobolin | 1362 | 1334 | 97.9 |

## Effect of population on biomass performance

Ranking of biomass performances of the populations was highly variable with significant population effects across the three assessments ($P < 0.001$); however, the population by site interaction was not significant ($P > 0.05$) at each of the three assessments. Removal of three populations that were confounded with family (single family population) did not change the significance of any factor. For the 3-year sapling estimates, populations from West Yalgogrin, Tallimba Rd West and Charcoal Tank Road performed strongly ranking within the top 3 populations at two of the three trials (Table 5). In the second assessment, West Yalgogrin, Tallimba Rd West, Tallimba Rd East and Winters Lane all ranked in the top three at two of the three trials. Across both sapling assessments, West Mid-West Highway, West Wyalong Town and Kerrs Lane performed poorly, with Kerrs Lane performing last at five of the six assessments.

In assessment of biomass performances for the coppice, Charcoal Tank Road ranked in the top three at all three trials, Tallimba Road East at two trials. West Wyalong Town and Kerrs Lane were poor performers with Kerrs Lane ranking last at two trials. When the single parent trees were removed, Winters Lane ranked in the top two at all three trials while West Yalgogrin ranked top two at Newdegate and Condobolin and West Mid-West Highway ranked second at Drummartin.

## Effect of family on biomass performance

Consistent with the population rankings, there was substantial family variation across assessments exemplified by significant family effect ($P < 0.001$) across all three assessments. There was also a significant family by site interaction effect in the first sapling assessments ($P < 0.01$) but this became non-significant in the 6-year sapling and coppice assessments ($P > 0.05$). Removal for the three families confounded with populations did not change the significance of any factor. Further assessment of the performance of the individual families is detailed in selection section below and the family estimates are provided in Tables S2, S3 and S4.

**Table 5** BLUEs of biomass for each population at the three progeny trials with standard errors

| Location | Population | Parent trees | 3-year sapling | 6-year sapling | Coppice |
|---|---|---|---|---|---|
| Newdegate | Charcoal Tank Rd | 1 | 1.686 ± 0.095 | 3.268 ± 0.164 | 1.869 ± 0.092 |
| | Kerrs Lane | 2 | 1.520 ± 0.072 | 3.214 ± 0.115 | 1.840 ± 0.074 |
| | Tallimba Rd East | 1 | 1.586 ± 0.089 | 3.323 ± 0.147 | 1.805 ± 0.086 |
| | Tallimba Rd West | 1 | 1.712 ± 0.089 | 3.582 ± 0.155 | 1.962 ± 0.089 |
| | West Mid-West Hwy | 4 | 1.598 ± 0.057 | 3.053 ± 0.079 | 1.759 ± 0.062 |
| | West Yalgogrin | 4 | 1.666 ± 0.058 | 3.305 ± 0.081 | 1.873 ± 0.063 |
| | Winters Lane | 24 | 1.652 ± 0.045 | 3.325 ± 0.047 | 1.866 ± 0.054 |
| | West Wyalong Town | 29 | 1.629 ± 0.044 | 3.262 ± 0.046 | 1.857 ± 0.054 |
| Average | | | 1.631 ± 0.069 | 3.279 ± 0.041 | 1.856 ± 0.053 |
| Drummartin | Charcoal Tank Rd | 1 | 1.934 ± 0.101 | 3.272 ± 0.158 | 2.313 ± 0.085 |
| | Kerrs Lane | 2 | 1.764 ± 0.071 | 2.962 ± 0.109 | 2.085 ± 0.065 |
| | Tallimba Rd East | 1 | 1.882 ± 0.098 | 3.109 ± 0.154 | 2.192 ± 0.083 |
| | Tallimba Rd West | 1 | 1.780 ± 0.100 | 3.042 ± 0.162 | 2.223 ± 0.086 |
| | West Mid-West Hwy | 4 | 1.849 ± 0.053 | 2.993 ± 0.081 | 2.246 ± 0.053 |
| | West Yalgogrin | 4 | 2.003 ± 0.055 | 3.282 ± 0.084 | 2.189 ± 0.054 |
| | Winters Lane | 24 | 1.945 ± 0.029 | 3.215 ± 0.042 | 2.306 ± 0.042 |
| | West Wyalong Town | 29 | 1.867 ± 0.028 | 3.076 ± 0.040 | 2.241 ± 0.041 |
| Average | | | 1.878 ± 0.067 | 3.133 ± 0.034 | 2.257 ± 0.040 |
| Condobolin | Charcoal Tank Rd | 1 | 1.959 ± 0.086 | 3.074 ± 0.123 | 1.835 ± 0.065 |
| | Kerrs Lane | 2 | 1.869 ± 0.063 | 3.052 ± 0.087 | 1.736 ± 0.052 |
| | Tallimba Rd East | 1 | 2.108 ± 0.086 | 3.297 ± 0.120 | 1.849 ± 0.065 |
| | Tallimba Rd West | 1 | 2.190 ± 0.094 | 3.292 ± 0.137 | 1.922 ± 0.071 |
| | West Mid-West Hwy | 4 | 2.067 ± 0.049 | 3.144 ± 0.062 | 1.815 ± 0.044 |
| | West Yalgogrin | 4 | 2.068 ± 0.049 | 3.282 ± 0.063 | 1.827 ± 0.044 |
| | Winters Lane | 24 | 2.087 ± 0.032 | 3.196 ± 0.031 | 1.827 ± 0.036 |
| | West Wyalong Town | 29 | 2.093 ± 0.031 | 3.204 ± 0.029 | 1.816 ± 0.036 |
| Average | | | 2.055 ± 0.061 | 3.198 ± 0.024 | 1.821 ± 0.035 |

## Genetic parameters

Biomass heritabilities were highest at the Condobolin trial with $h^2$ of 0.32 ± 0.08 for the 3-year sapling assessment, reduced to 0.18 ± 0.05 for the 6-year sapling assessment and 0.17 ± 0.05 for the coppice (Table 6). At Newdegate, heritabilities were lower for all three biomass assessments with the additive variance for coppice only marginally significant ($P$ = 0.0497). At Drummartin, heritability for coppice was 0.07 ± 0.04 but additive variance was only marginally significant ($P$ = 0.0497) for the 3-year sapling assessment and not significant for the 6-year sapling measurement ($P > 0.05$).

Within-site genetic correlations were high and significant ($P < 0.005$) between biomass assessments at the Newdegate and Condobolin trials with low standard errors ranging from 0.65 ± 0.12 to 1.07 ± 0.17 (Table 6). These were significantly different from unity between all assessments at Condobolin and between the two sapling assessments at Newdegate ($P < 0.05$) but not significant between the coppice and sapling assessments ($P > 0.05$). At both trials, the lowest genetic correlations were between the 2016 sapling and the 2020 coppice estimate. Very high genetic correlations were estimated for the 3- and 6-year sapling estimate (2013 and 2016) and the first sapling and the coppice estimate (2013 and 2020). At Drummartin, a weak genetic correlation was estimated for the 3-year sapling and coppice assessments ($r_g$ = 0.47 ± 0.31) due to the weak additive variance and was significantly different from unity ($P < 0.01$).

In contrast to the genetic correlations, phenotypic correlations were lowest between the 3-year sapling measurement and the coppice measurement which ranged from $r_p$ = 0.67 ± 0.02 to 0.73 ± 0.02 (Table 6). The strengths of the correlations were stronger between the two sapling measurements ($r_p$ = 0.79 ± 0.01 to 0.88 ± 0.01). The phenotypic correlations were stronger between coppice and 6-year saplings ($r_p$ = 0.75 ± 0.01 to 0.80 ± 0.01) than between coppice and 3-year saplings ($r_p$ = 0.67 ± 0.02 to 0.73 ± 0.02).

Cross-site genetic correlations were very high between Newdegate and Condobolin (0.82 ± 0.15 to 0.94 ± 0.13) with low standard errors for both sapling estimates and were very high for the coppice estimate but with higher standard errors (0.93 ± 0.31) (Table 7). The 3-year sapling genetic correlation

**Table 6** Heritabilities, genetic and phenotypic correlations ± standard errors for the 3- and 6-year sapling biomass assessments (2013 and 2016) and coppice assessment (2020) from the three mallee progeny trials. Heritabilities on the diagonal (in bold) with genetic and phenotypic correlations respectively above and below the diagonal entries. For cases with non-significant additive genetic variance, heritability and genetic correlations are denoted ns (non-significant)

| | Newdegate | | | Drummartin | | | Condobolin | | |
|---|---|---|---|---|---|---|---|---|---|
| | 2013 | 2016 | 2020 | 2013 | 2016 | 2020 | 2013 | 2016 | 2020 |
| 2013 | **0.20 ± 0.07** | 0.84 ± 0.09 | 1.07 ± 0.17 | **0.06 ± 0.03** | ns | 0.47 ± 0.31 | **0.32 ± 0.08** | 0.93 ± 0.04 | 0.83 ± 0.08 |
| 2016 | 0.79 ± 0.01 | **0.18 ± 0.06** | 0.79 ± 0.16 | 0.88 ± 0.01 | **ns** | ns | 0.87 ± 0.01 | **0.18 ± 0.05** | 0.65 ± 0.12 |
| 2020 | 0.68 ± 0.02 | 0.78 ± 0.01 | **0.08 ± 0.05** | 0.67 ± 0.02 | 0.80 ± 0.01 | **0.07 ± 0.04** | 0.73 ± 0.02 | 0.75 ± 0.01 | **0.17 ± 0.05** |

between Drummartin and Condobolin was also very high with similar standard errors (1.03 ± 0.37). The other coppice cross-site correlations were inflated well above one, due to weak additive variances used for these estimates and should be used with caution. All genetic correlations were not significantly different from unity ($P > 0.05$).

## Estimated gains following selection

Condobolin displayed the highest estimated gains for the 3-year sapling and coppice assessments with 24.1% and 14.8%, respectively (Table 8). Gains were similar for 3- and 6-year sapling at Newdegate with 17.2% to 17.7%, respectively and for 6-year sapling at Condobolin (16.5%). Gains were substantially lower for coppice at Newdegate (9.5%) and Drummartin (8.4%) which also had the lowest 3-year sapling gain of 11.5%. When applying the 3-year selections to the 6-year assessment, gains were reduced by about 13% at Condobolin and 3% at Newdegate. However, simulating the gains if 3-year sapling selections were applied to coppice, negative gains were observed for both sites (−6.2% and −7.1%) with positive but small gains at Drummartin. Repeating the simulation and assuming selection had been conducted in 6-year saplings immediately prior to harvest, positive gains were observed for coppice (6.4% to 9.3%) but these were smaller gains at each site than when selection was conducted and applied to the coppice (8.4% to 14.8%).

With fewer trees selected for clonal selection, gains were higher than for the orchard thinning scenario, with gains for saplings (24.6% to 65.3%) and coppice (18.9% to 37.2%) assessments. Similar trends were also observed when applying selections to other assessments, but the magnitude of gains was larger. When 3-year sapling selections were applied to coppice, gains were reduced by 10.7% at Drummartin, 36.0% at Newdegate and 51.4% at Condobolin when compared to coppice selections applied to coppice.

Comparison of the number of individuals selected from each family post-selection (Tables S5, S6 and S7) revealed four broad performance categories. These were (1) elite families across all assessments, (2) poor performing families across all assessments, (3) strong sapling but poor coppice performers and (4) weak sapling but strong coppice performers. By far, the most numerous category was elite families with 27 families at Newdegate and Drummartin and 25 families at Condobolin maintaining their full complement of possible selections across all assessments (e.g. families 7, 11 and 14). In contrast, selection completely eliminated seven families at Newdegate and five at Condobolin (e.g. families 30, 58, 59 and 66). At Newdegate and Condobolin, families 10 and 61 had the full sapling complement selected (or one eliminated) but no coppice selected, whereas families 17 and 54 performed poorly as saplings yet well after the coppice assessment. At Newdegate, family 48 was eliminated for sapling

**Table 7** Heritabilities, cross-site and genetic correlations ± standard errors for the 3- and 6-year sapling biomass assessments (2013 and 2016) and coppice assessment (2020) from the three mallee progeny trials. Heritabilities on the diagonal (in bold); genetic correlations off-diagonal. ns denotes non-significant additive variance. Trial names Newde, Drum and Condo refer to Newdegate, Drummartin and Condobolin, respectively

| | 2013 | | | 2016 | | | 2020 | | |
|---|---|---|---|---|---|---|---|---|---|
| | Newde | Drum | Condo | Newde | Drum | Condo | Newde | Drum | Condo |
| Newde | **0.19 ± 0.07** | 1.11 ± 0.37 | 0.82 ± 0.15 | **0.18 ± 0.06** | ns | 0.94 ± 0.13 | **0.07 ± 0.05** | 1.45 ± 0.49 | 0.93 ± 0.31 |
| Drum | | **0.05 ± 0.03** | 1.03 ± 0.32 | | **ns** | ns | | **0.07 ± 0.04** | 1.21 ± 0.25 |
| Condo | | | **0.32 ± 0.08** | | | **0.21 ± 0.06** | | | **0.19 ± 0.05** |

**Table 8** Gains made from selection to produce a thinned seed orchard or clonal orchard using data from the three biomass assessments: 3- and 6-year sapling and coppice at Newdegate and Condobolin, and the 3-year sapling and coppice assessment at Drummartin. Gains were calculated from the selections made in 3-year sapling assessments and applied to the 6-year sapling assessment. Gains were also calculated for coppice biomass assuming selections had been conducted in both 3-year and 6-year saplings. The number of trees selected for thinning at Newdegate (*n* = 171–185), Condobolin (*n* = 262–284) and Drummartin (*n* = 238–240), and for clonal population *n* = 10

| Trial | Selection | 3-year sapling gain (%) | 6-year sapling gain (%) | Coppice gain (%) | 6-year gain using 3-year selections (%) | Coppice gain using 3-year selections (%) | Coppice gain using 6-year selections (%) |
|---|---|---|---|---|---|---|---|
| Newdegate | Thinning | 17.2 | 17.7 | 9.5 | 13.8 | −6.2 | 6.4 |
| Condobolin | | 24.1 | 16.5 | 14.8 | 11.3 | −7.1 | 9.3 |
| Drummartin | | 11.5 | | 8.4 | | 3.4 | |
| Newdegate | Clonal | 40.4 | 41.3 | 22.6 | 26.9 | −13.4 | 15.4 |
| Condobolin | | 65.3 | 40.6 | 37.2 | 35.3 | −14.2 | 19.8 |
| Drummartin | | 24.6 | | 18.9 | | 8.2 | |

selections yet the full complement of coppice was selected, and this also occurred with family 25 at Condobolin.

A similar trend held for the clonal selections (Table 8): there were four families that were selected seven or more times out of eight possible selection opportunities (families 11, 19, 41 and 46) (Table S8). Twenty-two families were only selected as sapling whereas eight families were only selected as coppice. Out of the 66 families present in the progeny trials, 36 were not represented for clonal propagation.

## Discussion

This study shows that across the three trials, almost all *E. polybractea* saplings coppiced following harvest. From the populations used in this study, coppicing seems to be a widespread adaptation suggesting that coppicing is strongly linked to the species fitness. There were some large differences in the performances of certain families when comparing the sapling to the coppice assessments. Heritabilities were weak to moderate for all biomass assessments and reduced from first sapling assessment to coppice assessment at Condobolin and Newdegate but increased at Drummartin. Strong within-site phenotypic correlations were observed at all trials with high genetic correlations at the trials with significant additive variance. Within years, cross-site genetic correlations were strong between the trials. Gains were highest for the sapling biomass and reduced for coppice, but if used as a short-rotation coppice crop, selection should be done on the coppice assessment.

Considerable variation of growth rates was observed across the three trials. Condobolin experienced the fastest growth before the 3-year sapling assessment with the slowest growth at Newdegate. Part of elevated growth rates at Condobolin may be explained by the manual watering post-planting, whereas Newdegate receiving about 900 mm less rainfall over the 3 years than the other two sites. However, by the 6-year sapling assessment, tree sizes across the three trials were similar suggesting that the Condobolin may have experienced competition-related growth suppression. The average size of the coppice at Drummartin was more than double that of the other two sites, but over the period, Drummartin received an additional 100 mm of rainfall with the lowest evaporation rates.

## Survival

Previous work has reported that survival from trials at multiple locations in WA, Victoria and NSW and under varying numbers of harvests was generally high, ranging from 90 to 99% (Goodger et al. 2007; Milthorpe et al. 1998; Milthorpe et al. 1994; Spencer et al. 2019; Wildy et al. 2000). In our study, survival was at the higher range recorded for this species. Various factors, including family (Whittock et al. 2003), environment (Spencer et al. 2019; Wildy et al. 2000), harvest timing and frequency (Spencer et al. 2019; Wildy and Pate 2002) can contribute to mortality across studies. The seed for the relatively low (90%) survival reported in Goodger et al. (2007) was from central Victoria whereas in our study, the families were collected from parent trees located in a disjunct south-central NSW population, several hundred kilometres to the northeast. It is possible that the origin of parent trees (environmental adaptation) contributes to resprouting capacity; however, it is more likely that the age at first harvest is responsible for the difference in survival with the harvest age between 1 and 3.5 years old from the trials with lower survival (Goodger et al. 2007; Milthorpe et al. 1998; Milthorpe et al. 1994; Wildy et al. 2000). In this present experiment, the mallee were 6 years old and between 5- and 10-year-old trees at first harvest in Spencer et al. (2019). We found no evidence that population differences contributed to survival following harvest.

We found that larger *E. polybractea* saplings were more likely to coppice than smaller saplings. In contrast, Wildy et al. (2000) found for the same species that coppice success is not related to the size of the sapling prior to harvest. These divergent results may be explained by the age and planting configuration in our study with older trees subjected to block planting configuration compared to alley planting used by Wildy et al. (2000). Both of these factors increase competition between trees suppressing smaller trees, which were less likely to reshoot. Whittock et al. (2003) found in agreement with our study, for *E. globulus*, that coppice success was determined by the size of sapling prior to harvest ($r_g = 0.61$). For *E. kochii*, Wildy and Pate (2002) found that coppice success is not due to starch reserves or the number of meristematic foci in the lignotubers.

## Genetic parameters

Heritability of sapling biomass is generally low to moderate for semi-arid mallee eucalypt species (Mazanec et al. 2020; Mazanec et al. 2017; Spencer et al. 2020); however, nothing has been published regarding the heritability of coppice production of *E. polybractea*. The heritability of resprouting following harvest for other eucalypt species has been estimated. For example, the diameter at breast height of *E. dunnii* coppice has been recorded as $h^2 = 0.42 \pm 0.17$ (Li et al. 2012) and a range of $h^2 = 0.33$ to 0.57 for *E. grandis* (Reddy and Rockwood 1989). Much lower heritabilities ($h^2 = 0.16 \pm 0.05$) were found for coppice height for *E. globulus* in Tasmania which is closer to the range found in the present study (Whittock et al. 2003).

At Newdegate, the two heritability estimates for saplings were stable but lower for coppice. At Condobolin, there was a large drop between the first and second sapling heritability estimates and a further, but smaller, drop in the coppice cycle. At Drummartin, the heritability of coppice was higher than for the two sapling assessments. The reason for the divergent trends in heritability between sites is unclear and may be driven by the different environmental conditions at the three trials and contrasts with the findings from Osorio et al. (2001) who found for *E. grandis* that heritabilities of tree volume and mean annual increments increased from 3 years old until 6 years old (harvest age). It was, however, expected that there would be divergence of performance in a mallee between the sapling and coppice assessments, on the basis of genetic control of lignotuber development in other eucalypt species (Bortoloto et al. 2020; Walters et al. 2005; Whittock et al. 2003).

Heritability of below 0.1 renders selection of good genotypes unreliable (Cotterill and Dean 1990) which is of concern for selection of coppice at Newdegate and Drummartin. The timing of measurement may influence heritability estimates. The coppice were below the size likely to be profitable for harvest especially at Newdegate and Condobolin where the average coppice was under 12 kg (Spinelli et al. 2014). These trials should be assessed immediately prior to harvest of the first coppice cycle as differentiation between families may increase with age. If heritability estimates are not improved, then it may be necessary to use backward selection as this enables calculation of parental breeding values with high accuracy (Falconer and Mackay 1996; Isik et al. 2017).

Across all trials, there were moderate to high positive phenotypic correlations between the three biomass estimations indicating that bigger saplings, either at age 3 or 6 years, will produce bigger coppice. This is contrary to the finding from the selection scenarios that show that selection for 3-year-old saplings will result in a negative gain for coppice production. However, across all trials, the phenotypic correlations were weakest between the first sapling and the coppice estimates. These correlations were similar to the Pearson's correlation found between sapling and coppice biomass for *E. polybractea* of $r = 0.73$ which reduced to $r = 0.66$ between two coppice cycles (Goodger et al. 2007). This indicates that, if selecting for coppice biomass using sapling data, selections are best done on older saplings, which are less likely to be affected by nursery or planting effects.

Across the three *E. polybractea* trials, within-site genetic correlations revealed similarities and differences when compared to the phenotypic correlations. Excluding the Drummartin trial (due to non-significant or weak sapling additive variance), strong and positive genetic correlations were estimated between the sapling (3 and 6 years) and the coppice biomass estimates indicating that similar genes are associated with sapling and coppice growth (Falconer and Mackay 1996). However, contrary to the phenotypic correlations, there were stronger genetic correlations between the first sapling estimate and the coppice estimate at Newdegate (1.07 ± 0.17) and Condobolin (0.86 ± 0.07) than between the second sapling assessment and coppice (Newdegate (0.79 ± 0.14) and Condobolin (0.67 ± 0.11)) but these were either within or close to the margins of error. Similar magnitude of genetic correlations have been found for *E. dunnii* in China (Li et al. 2012). Contrary to these findings, Whittock et al. (2003) found much weaker correlations between diameter of sapling and the height of coppice ($r_g = 0.12$). Such divergence may arise from the substantial difference between subraces of *E. globulus* in lignotuber size and development which led to a much lower proportion of coppice success across the species distribution (Whittock et al. 2003). Similar results were observed by Walters et al. (2005) for *E. obliqua* who found a provenance response to lignotuber size. *E. obliqua* and *E. globulus* are able to regenerate from seed or coppicing from lignotubers, but it seems that certain provenances favour either method. Mallee eucalypts invest in more below-ground biomass than non-mallee species, with *E. polybractea* having root to shoot ratio of 0.61 (Brooksbank and Goodwin in press) which is substantially higher than *E. globulus* with 0.29 to 0.30 (Fabião et al. 1995; Resh et al. 2003). This additional investment in below-ground biomass allows almost all *E. polybractea* saplings to reshoot, unlike *E. obliqua* and *E. globulus*.

There is some debate as to whether selections should be done on first (sapling) or second (coppice) rotations for coppice crops. A study of five experiments of mixed eucalypt species in Brazil demonstrated that selection before the first rotation was appropriate (Amâncio et al. 2020). This is contrasted with the results from a small *E. polybractea* trial that suggests that selection for biomass at first coppice rotation is better than when done at saplings or after the second coppice rotation (Goodger et al. 2007). Studies with the aim of quantifying the best rotation for biomass selection of coppice crops are rare, but our results are in agreement that second rotation data should be used.

At Drummartin, the additive variance became significant after harvest suggesting that the genes associated with coppicing were more strongly expressed than for either of the sapling assessments that either had very weak or non-significant additive variance. Low additive variance resulting in low or no heritability suggests there is little differentiation between families. The higher differentiation between families post-coppicing at Drummartin seems to be an anomaly and was not repeated at the other two trials. This suggests that another factor is operating and may be explained, in part, by the genetic control of lignotuber development in some eucalypts species (Bortoloto et al. 2020; Walters et al. 2005; Whittock et al. 2003). Walters et al. (2005) demonstrated in a nursery experiment that *E. obliqua* with larger lignotubers had higher concentrations of carbohydrates and after decapitation, produced more above-ground biomass. If the same mechanism was responsible for coppice regrowth for *E. polybractea*, the higher heritability of coppice growth at Drummartin may actually be indirect measure of the heritability of lignotuber development. However, why this trend was observed at Drummartin and not the other trials is unclear; possibly peculiar site conditions triggered different genes which give rise to more detectable additive variance.

### Genotype by environment interactions

The genotype by environment cross-site analysis for this study was partly compromised due to non-significant additive variance for the 6-year sapling assessments at Drummartin and the weakly significant additive variance for the coppice assessment at Newdegate. This resulted in inflated estimates of cross-site genetic correlations for Drummartin with the other trials, especially for the coppice assessments. Being limited to two sites with significant additive variance for all three biomass assessments is not ideal for cross-site analysis and these results should be taken with some caution until confirmed on more sites or further assessments of coppice biomass. However, genetic correlations between Newdegate and Condobolin trials were positive and very strong ($r_g = 0.80$ to 0.94) for all three assessments. There is conjecture from different studies regarding the threshold of cross-site genetic correlations necessary for separate trials into different breeding programmes to maximise biomass gains (Li et al. 2017). However, Robertson (1959) suggested a threshold of <0.80 to indicate practical significance of G × E interaction, whereas Xie (2003) suggested a threshold of 0.70. Our results suggest that G × E between the Newdegate and Condobolin trials, although having quite different soils and climates, is of little practical significance.

It is clear that the sapling phase in a commercial mallee plantation forms only a small fraction of the productive life of the trees, with the vast majority of biomass produced in successive coppice cycles (Bartle and Abadi 2010; Davis 2002; Spencer et al. 2019). The results of this study combined with that of Goodger et al. (2007) indicate that maximum gains in biomass production in the first coppice cycle are likely to be achieved by selecting seed orchard parents after the first coppice cycle. Some caution is required and additional measurements in subsequent coppice cycles are required to confirm this result. These findings dictate that considerable time is required to establish an improved seed orchard suitable for coppice production from wild *E. polybractea* parents, with first thinning at a minimum of 5–6 years then a further 3–4 years for coppice assessment, then an additional 3–4 years to produce usable seed. However, this process may be hastened by thinning the three progeny trials into seed orchards based on coppice selections, or by producing elite clonal orchards using coppice shoots (Goodger et al. 2008) which would hasten improved seed for a biomass industries.

## Conclusions

This study found that *E. polybractea* coppices vigorously after harvesting with very low mortality. Due to the uneven number of families in each population, little is gained from further analysis, but generally, populations that performed well did so across the three trials and the three assessments. Inspection of the performance of the individual families revealed that certain families seem to be disposed to either sapling or coppice production. However, more than half the families were stable across the three biomass assessments, with consistently high or low yields.

We found similar genetic parameters to other studies of mallee, with low to moderate heritabilities of biomass estimates. However, this study is unique because it includes biomass estimates of both sapling and coppice. Strong within-site genetic and phenotypic correlations were established between sapling and coppice biomass estimates across two of the three trials suggesting that selections of superior saplings or coppice would result in biomass gains. The cross-sites genetic correlations, where additive variance was significant, revealed that G × E was likely to be of little practical significance. If *E. polybractea* is to be used for long-term short-rotation coppice crops, selection for breeding may best be conducted in the first or subsequent coppice cycles. Assessment of additional coppice cycles is necessary to confirm at what point selection is optimal. Due to the low additive variance for the coppice assessments at Drummartin and Newdegate, a further harvest and reassessment of biomass after 3–4 years of growth may yield improved estimates of genetic parameters at these sites with the potential for further biomass gains. These trials may be thinned to produce improved seed orchards or elite individuals could be used for explant sources for clonal orchards. This will result in enhanced biomass production of *E. polybractea* which will benefit commercial plantations.

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**Author contribution** R.M. established the trials. B.S. and R.M. conceived the idea of the experiment. B.S. carried out the experiment and drafted the manuscript. B.S. and R.M. completed the statistical analysis. R.M., A.Z., A.A. and M.G. contributed to the final version of the manuscript.

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**Data availability** Most data are available in supplementary material. Where data is not available, data will be made available on reasonable request.

**Code availability** ASReml code will be made available on reasonable request.

## Declarations

**Conflict of interest** The authors declare no competing interests.

**Data archiving statement** Biomass estimates from this manuscript have not been currently achieved, but prior to publication, these data will be published on CSIRO data access portal with a DOI number https://data.csiro.au/dap/home?execution=e1s1.

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# Chapter 6

## Economic determinants of mallee viability

Spencer B, Abadi A, Bartle J, Sudmeyer R, Van Gent S, Gibberd M, Zerihun A (2021). Determinants of the economic viability of mallee eucalypts as a short rotation coppice crop integrated into farming systems of Western Australia. GCB Bioenergy 13:242-256 doi: https://doi.org/10.1111/gcbb.12775

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ORIGINAL RESEARCH

GCB-BIOENERGY BIOPRODUCTS FOR A SUSTAINABLE BIOECONOMY Open Access WILEY

# Determinants of the economic viability of mallee eucalypts as a short rotation coppice crop integrated into farming systems of Western Australia

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## Abstract

Mallee eucalypts are being developed as a short rotation coppice crop for integration into agricultural systems in the south-west of Western Australia. These have potential for biomass production for bioenergy, eucalyptus oil and generating carbon credits and to help control the extensive occurrence of dryland salinity. Some 12,000 ha of mallee planting has been undertaken since 1994, mostly in the form of wide-spaced, narrow belts within the annual agricultural system. Production and market data were used to estimate levelized costs (LC) of mallee biomass production under different harvest regimes across 11 sites from 2006 to 2012. We found LC ranged from AUD40 to AUD257 fresh $\text{Mg}^{-1}$. LC was most strongly determined by mallee production, followed by the crop/pasture rotation decisions of the landholder. Mallee harvest regime had minor impact on LC. Crop and pasture yield loss due to competition from the mallee belts accounted for 38% of costs, harvesting biomass was 32%, opportunity cost of the land occupied by the mallee belts was 16% while establishment and maintenance costs accounted for 14% of the costs. When income from carbon sequestered in mallee root biomass was included, the LC dropped by an average of 11% at the current Australian price of AUD15 $\text{Mg}^{-1}$ $CO_2$ equivalent ($CO_2$e). The income from carbon sequestered in root biomass alone is unlikely to make mallee agroforestry economically viable. Hence, income from harvested biomass in the form of feedstocks for industry or carbon credits is necessary to make mallee agroforestry commercially attractive. LC for unharvested mallee belts ranged from AUD33 to AUD237 $\text{Mg}^{-1}$. Where above- and below-ground biomass is converted to $CO_2$e at AUD15 $\text{Mg}^{-1}$, the LC drops to AUD11–AUD64, with three of 11 sites likely to be profitable. These three sites were characterized by high biomass production with low agricultural gross margins.

KEYWORDS

agroforestry, alley cropping, carbon sequestration, competition zone, levelized cost, oil mallee, tree-crop competition

# 1 | INTRODUCTION

Integration of mallee eucalypts—which are lignotuberous Eucalyptus spp. with multi-stemmed growth form—into the dryland farming systems in the wheatbelt of Western Australia (WA) could help address several land degradation issues, in particular the on-farm impacts of dryland salinity and its adverse downstream consequences for water resources, conservation and infrastructure (Bartle et al., 2007; Clarke et al., 2002; George, 1990). Since the early 1990s, widespread test planting of mallee was undertaken with some 1,000 farmers establishing mallee belts on more than 12,700 ha of land (Bartle & Abadi, 2010; URS, 2008). However, the use of revegetation for salinity mitigation is contentious (George et al., 1999) and benefits will take decades to be realized and require extensive planting as part of an integrated farming system. Hence, mallee cropping must also generate an economic return to make it viable.

Selected mallee species have long been used for small-scale production of eucalyptus oil (Davis, 2002). Its major constituent, 1,8-cineole, has potential for large-scale markets in biofuels and industrial products (Barton & Tjandra, 1989; Mewalal et al., 2017; Soh & Stachowiak, 2002). Mallee biomass also has potential as bioenergy and biofuel feedstock (Barron & Zil, 2006; Garcia-Perez et al., 2008; McGrath et al., 2016; Wu et al., 2010; Yu et al., 2009) and biochar (Abdullah & Wu, 2009; Ding et al., 2016). More recently, the Australian Government Carbon Credits (Carbon Farming Initiative) Act 2011 provides opportunities for mallee plantings to generate revenue. In the United States, alley cropping has been estimated to have the potential to mitigate 82 Tg of $CO_2e$ per year (Fargione et al., 2018). In Australia, there are vast untapped agricultural areas with potential to mitigate $CO_2e$ using perennial crops, of which mallee is a strong candidate (Hobbs et al., 2009).

To date, efforts have focussed on assessing the utility of mallee agroforestry and optimization of design and production (Mendham et al., 2012; Peck et al., 2012). Lefroy and Stirzaker (1999) proposed that widely dispersed belts of woody perennials were likely to be the most effective planting configuration for groundwater management. Mallee agroforestry plantings typically consist of belts of mallee with two to six rows separated by 40–100 m wide alleys of conventional crops and pasture (URS, 2008). Narrow belts (fewer rows of mallee) provide greater biomass productivity per unit of land occupied by the belt compared to wider belts or block plantings (Noorduijn et al., 2009; Paul et al., 2013; Spencer et al., 2020). However, narrower belts increase the area of interaction between mallee and the adjacent crop/pasture for a given area planted to mallee. Productivity of crops and pasture within 20 metres of the mallee belts is suppressed due to competition for water (Robinson et al., 2006; Sudmeyer et al., 2012; Sudmeyer & Hall, 2015). For this reason, Sudmeyer and Hall (2015) proposed segregation of mallee from agriculture to reduce the competition loss.

Due to the prevalence of wide-spaced belt planting (URS, 2008), and to facilitate further adoption, the direct and/or indirect economic benefits of mallee production need to be quantified. Past economic studies have had limited long-term experimental data and have used simulation modelling of mallee belt growth and the interaction of belts with crops/pastures to estimate the likely costs and benefits of integrating mallee into the farming systems (Abadi et al., 2012; Bartle & Abadi, 2010). Using this modelling approach, Bartle and Abadi (2010) found that mallee agroforestry (harvested at year 5 and then every 3 years), when compared to agriculture, became profitable after 12 years at a selling price of AUD45 per fresh Mg. Subsequently, Abadi et al. (2012) modelled the economics of a mallee biomass production system and suggested that the cost of production was in the range of AUD53–AUD70 per Mg of fresh biomass with co-benefits valued at between AUD2 and AUD15 $Mg^{-1}$.

This paper considers the economic viability of mallee in an agroforestry system using a decadal experiment providing yield data from mallee belts with six harvesting treatments across 19 sites (Spencer et al., 2019) and crop and pasture yields measured adjacent to the belts over 6 years (Sudmeyer et al., 2012). These data sets provide a unique opportunity to assess the economic viability of mallee using experimental data obtained from operational short rotation coppice systems with real-world management by farmers (Hauk et al., 2014). The aim of this study is to determine break-even prices of mallee biomass compared to conventional agriculture using levelized cost (LC) analysis. LC has been widely used to compare types of energy production (Edenhofer et al., 2012) and also been utilized in calculating the production cost of bioenergy and biofuel crops (Abadi et al., 2016; El Kasmioui & Ceulemans, 2013). LC is useful where the costs of production are known, but there is no active trading in local markets for the product (Peirson et al., 2002). Four scenarios are explored: (a) income generated from agriculture alone, (b) income from harvested above-ground mallee biomass, (c) income from harvested above-ground biomass plus carbon sequestered in below-ground biomass, and (d) income from carbon sequestered in unharvested above- and below-ground biomass. Scenarios b, c and d included the costs associated with reduced agricultural production alongside the mallee belts. Sensitivity of the financial returns was assessed by adjusting key variables for a range of assumptions including discount rates, below-ground biomass estimates and carbon price.

# 2 | MATERIALS AND METHODS

## 2.1 | Study sites and species

This study includes 11 of 19 mallee trial sites originally established to determine mallee and agricultural yield from alley farming systems (Spencer et al., 2019; Sudmeyer et al., 2012). Sites were established in 2006 with 5- to 12-year-old mallee belts on privately owned farms in the wheatbelt of WA (Figure 1; Table 1). For continuity, site names remain the same as in Spencer et al. (2019). Sites 6, 7 and 14 were excluded due to low survival and production following the first harvest (Spencer et al., 2019). Sites 2, 9 and 10 were excluded because the alley widths were too narrow (<40 m) to estimate open paddock yield. Sites 11 and 17 were excluded due to incomplete agricultural data sets (Sudmeyer et al., 2012). The belts were either 2, 3, 4 or 6 rows wide and the alley widths were between 48 and 250 m (Table 1). Further detail about the sites is published in two reports (Mendham et al., 2012; Peck et al., 2012).

The WA wheatbelt has a Mediterranean climate with dry hot summers and mild, cool and rainy winters. Mean annual rainfall ranged from 539 mm for the southerly sites to 321 mm for the northern sites (Table 1). The crops and pastures in the wheatbelt of WA are non-irrigated winter-growing annuals. The pastures are typically grazed with self-replacing merino sheep producing wool and meat. Crops and pastures are grown in annual rotations which can generally be characterized as cereal–pasture–pasture; cereal–pasture–cereal; cereal–legume–cereal; cereal–cereal–canola (Harries et al., 2015).

The three mallee species most widely planted by farmers in WA are represented in this study; *Eucalyptus loxophleba* subsp. *lissophloia* L.A.S. Johnson & K.D. Hill, *Eucalyptus polybractea* R. Baker and *Eucalyptus kochii* subsp. *plenissima* C.A. Gardener. These species will hereafter be referred to as $E_{lox}$, $E_{pol}$ and $E_{koc}$ respectively.

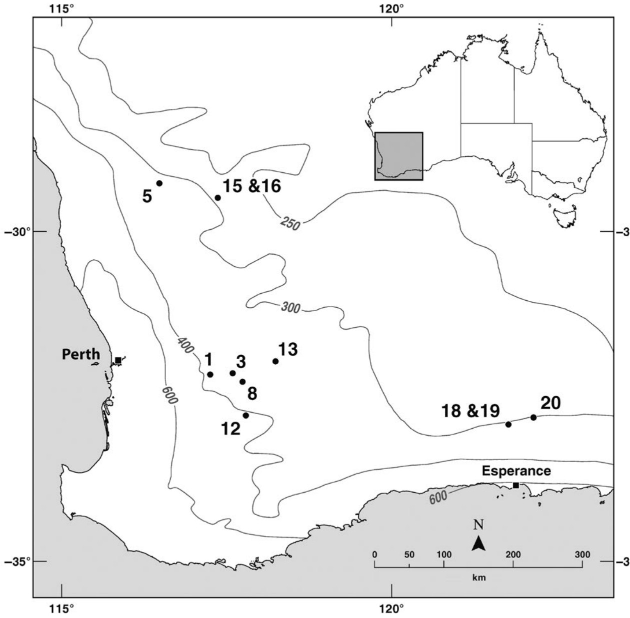

**FIGURE 1** Location of mallee trial sites within the Western Australia wheatbelt; also shown are selected rainfall isohyets (grey line). The site numbers correspond to those in table 1 of Spencer et al. (2019)

## 2.2 | Experimental design

The experimental design at each site was a 2 × 2 factorial, plus unharvested plots, with three replicates. The factors, each with two levels, were frequency of harvest (short vs. long harvest cycles) and season of harvest (spring vs. autumn). Sites 1, 3, 5, 8, 12 and 15 had treatment plots that were 20 m long (along the mallee belt) with a 10 m buffer separating the plots, the remaining sites had 25 m long plots with a 12.5 m buffer. Prior to the establishment of this trial, no mallee had been harvested.

Initially, the frequency of mallee harvest treatments was 3 or 4 years, but at the less productive sites (12, 15 and 20), the second harvest was delayed to avoid the risk of high mallee mortality. At these sites, harvest frequency was extended to 6 years (Table 1).

Crop and pasture were grown in the alley adjacent to each mallee belt in rotations determined by the individual farmer at each site. Each year from 2006 to 2011, the yield of the crop or pasture was determined by harvesting plots parallel to and 2, 4, 6, 8, 12, 16, 20, 24 and 30 m from the mallee belt for each treatment replicate (Sudmeyer et al., 2012). For pasture paddocks, yield was assessed each year in September and is indicative of relative growth as a function of distance from mallee belts, not total annual pasture yield.

Above-ground mallee biomass yield data were derived and adjusted from Spencer et al. (2019) and summarized in Tables S4 and S5. First, fresh biomass data were used for the purpose of economic analysis, to be consistent with the on-farm gate price for unprocessed fresh woody biomass. Second, the 2 m wide crop exclusion zone (Figure 2) on both sides of the belt was added to account for the displaced cropping/pasture area. Thirdly, the biomass data are expressed as actual fresh harvest yield (Mg/ha) for each treatment rather than annualized increments ($\text{Mg ha}^{-1}\ \text{year}^{-1}$).

Above-ground dry biomass was calculated for the unharvested treatments for carbon sequestration estimations as detailed in Spencer et al. (2019). Total biomass was calculated as the biomass produced over the 6 year length of the study.

## 2.3 | Mallee carbon estimation

After harvest, mallee shed their fine roots but maintain the lignotuber and structural woody root architecture (Wildy & Pate, 2002). Below-ground biomass was estimated for each coppice treatment using the general mallee eucalypt allometric model from Paul et al. (2014). This model estimates

**TABLE 1** Site characteristics and planting designs for mallee trial sites including mallee species, year of planting, number of rows in each belt, the alley width between belts and the harvest frequency at each site. Mean annual rainfall (MAR) data from 1970 to 2011 were obtained through SILO data sets (Jeffrey et al., 2001)

| Site number | Species | Year planted | Number of rows | Alley width (m) | Harvest frequency (years) | MAR (mm) |
|---|---|---|---|---|---|---|
| 1 | $E_{pol}$ | 1996 | 2 | 70 | 3 and 4 | 432 |
| 3 | $E_{lox}$ | 2000 | 3 | 50 | 3 and 4 | 353 |
| 8 | $E_{pol}$ | 1998 | 4 | 180 | 3 and 4 | 368 |
| 13 | $E_{lox}$ | 1997 | 2 | 48 | 3 and 4 | 326 |
| 18 | $E_{pol}$ | 2001 | 6 | 95 | 3 and 4 | 539 |
| 19 | $E_{pol}$ | 2001 | 6 | 130 | 3 and 4 | 539 |
| 16 | $E_{koc}$ | 1994 | 2 | 95 | 3 | 321 |
| 5 | $E_{lox}$ | 1998 | 4 | 250 | 4 | 327 |
| 12 | $E_{lox}$ | 2000 | 6 | 55 | 6 | 370 |
| 20 | $E_{lox}$ | 2001 | 6 | 180 | 6 | 457 |
| 15 | $E_{koc}$ | 1998 | 2 | 95 | 6 | 321 |

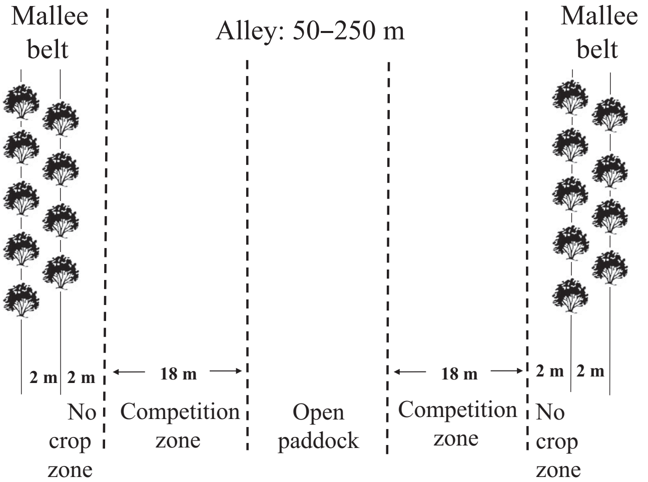

**FIGURE 2** Schematic cross section of a two-row mallee belt with 2 m exclusion zones and an alley between the belts comprising of competition zones and open paddock

below-ground biomass based on the height of the coppice; however, the accuracy of the model in estimating mallee below-ground biomass under frequent harvest management has not been exhaustively evaluated. Thus, to assess impact of possible under- or overestimation of mallee root biomass on LC estimates, a sensitivity analysis was carried out using three below-ground biomass estimates; the minimum, maximum and average over the 6 years of trial.

For the unharvested mallee plots, the carbon sequestered in the above- and below-ground biomass over the 6 years of trial period was estimated by assuming dry biomass to be 50% carbon. Below-ground biomass was calculated as a proportion of the above-ground biomass using the data from Brooksbank and Goodwin (in press).

## 2.4 | Crop and pasture yield

The methodology for measurement of crop and pasture yield adjacent to the belt is described in Sudmeyer et al. (2012) and summarized in the Supplementary Materials.

Open paddock yield was determined as the average crop/pasture yield ≥20 m from mallee belt for all treatments given the greatest lateral extent of mallee competition was 18.7 m from the belt (Sudmeyer et al., 2012). To standardize yield across all sites and treatments, the yield in the competition zone (Figure 2; <20 m from the belts) was expressed as the percentage of the open paddock yield (relative yield; Sudmeyer et al., 2012).

The open paddock crop yields and relative yields used in this study were mostly derived from Sudmeyer et al. (2012) and are detailed in Table S1. However, at site 8 in 2008 and site 13 in 2010, crop data were not available. For such cases, average regional yield data from that growing season were used (Planfarm-Bankwest, 2007, 2008, 2009, 2010, 2011, 2012). When data were not collected for a particular treatment, the data were patched using the average yield proportion of the treatment relative to open paddock yield across all other measured years.

## 2.5 | Economic analysis

The economic analysis was done over 6 years using reported estimates of returns and costs for mallee production (autumn and spring 2006–2012) and regional averages for crop and sheep enterprises (growing seasons 2006–2011). To standardize sites with different paddock dimensions and belt design, it was assumed that all sites were 100 ha in area assuming nil loss of crop area due to fences, tracks or other obstructions.

Alley and belt widths from each site were maintained and all belts were assumed to be straight and parallel.

At each site, the 100 ha paddock was divided into three components: (a) the mallee belt plus 2 m uncropped (exclusion zone) on either side of the belt; (b) the competition zone, being the area of mallee crop/pasture interaction 2–20 m from both sides of each belt; and (c) the area of open paddock outside the competition zone (Figure 2). The area of each component at each site is detailed in Table 2. Total mallee biomass was calculated by multiplying the yield per hectare for each treatment by the belt area at each site. The total crop/pasture yield was calculated by multiplying the total area of crop/pasture in both the competition zone and the open paddock (ha) by the respective yield (Mg/ha) and adding the quantities.

## 2.6 | Production costs and prices for mallee biomass or carbon sequestration

The costs of production for mallee belts were estimated for establishment, maintenance and harvest. Establishment cost used in this study was AUD1,334 ha$^{-1}$ (Cooper et al., 2006) which was amortized over a period of 30 years per year using equivalent annual annuity:

$$C=\frac{r\times \mathrm{NPV}}{1-(1+r)^{n}}, \tag{1}$$

where $C$ is equivalent annuity cash flow, $r$ is the discount rate per period and is assumed to be 13%, NPV is the net present value of the establishment costs and $n$ is the project life in years. NPV is used to account for the time value of funds invested in the paddock, including mallee and crops, over several seasons (Peirson et al., 2002). Maintenance cost was assumed to be AUD15 ha$^{-1}$ year$^{-1}$ or AUD55 ha$^{-1}$ following harvest (Cooper et al., 2006). Harvest cost was assumed to be AUD22 per chipped fresh harvested Mg which is the low end of the range as measured by Spinelli et al. (2014) using conventional forestry equipment and in the range estimated by Abadi et al. (2012). Storage and transport costs are assumed to be zero as biomass is assumed to be sold as fresh chips at the farm gate (El Kasmioui & Ceulemans, 2013). No harvest cost was applied to unharvested treatments which were assumed to be used for carbon sequestration.

**TABLE 2** Breakdown of the 100 ha paddock into three components (in hectares) across all sites. Three components are area of mallee belt (including 2 m exclusion zone on either side of mallee belt), area where crop/pasture was subject to competition and the area of open paddock where crop/pasture was not subject to competition

| Site number | Area mallee belt (ha) | Area competition zone (ha) | Area of open paddock (ha) |
|---|---|---|---|
| 1 | 8.4 | 50.4 | 41.2 |
| 3 | 15.2 | 68.4 | 16.4 |
| 5 | 4.0 | 14.4 | 81.6 |
| 8 | 5.0 | 18.0 | 77.0 |
| 12 | 21.0 | 54.0 | 25.0 |
| 13 | 12.0 | 72.0 | 16.0 |
| 20 | 7.0 | 18.0 | 75.0 |
| 15 | 6.0 | 36.0 | 58.0 |
| 16 | 6.0 | 36.0 | 58.0 |
| 18 | 14.0 | 36.0 | 50.0 |
| 19 | 9.8 | 25.2 | 65.0 |

## 2.7 | Production costs and prices for grain and sheep production

The operational costs associated with crop and sheep production were estimated using regional data for each experimental year (Planfarm-Bankwest, 2007, 2008, 2009, 2010, 2011, 2012). These costs are summarized in Table S2.

Crop prices for the WA regional export terminal (Kwinana) were obtained for 1 January (or as close as possible) for each year following the growing season (ABARE, 2015; Grain & Graze3, 2020). Sheep income was calculated as the sum of the wool and sheep returns per hectare for each region using industry benchmarks (Planfarm-Bankwest, 2007, 2008, 2009, 2010, 2011, 2012). Crop and sheep prices are detailed in Table S3.

The economic analysis used the actual crop yields (open paddock and competition zone) achieved at each site. Pasture yield was measured annually by Sudmeyer et al. (2012) and regional returns from sheep enterprises were used and discounted by relative pasture yield in the competition zone.

## 2.8 | Economic model

Four scenarios were modelled: (a) base-case—exclusively agricultural with no mallee in the system, (b) agroforestry utilizing above-ground mallee biomass, (c) agroforestry utilizing above-ground mallee biomass plus below-ground biomass sequestered, (d) sequestration using unharvested mallee above- and below-ground biomass. The economic analysis presents estimates of the financial viability of each scenario by comparing the base-case model with agroforestry at each site and harvest treatment applied to a 100 ha area. LC analysis was used to determine the price of mallee biomass and sequestered carbon required for mallee agroforestry to break-even with agriculture. LC standardizes the unit price needed over time to break-even with variable capital and operating expenses over several seasons (Peirson et al., 2002). LC, being a modified version of the net present value calculation, accounts for time value of money using a discount rate.

The model calculates gross margins (GM) in each year for both agroforestry and agricultural paddocks. The GM of the agricultural system and the crop/pasture component of the agroforestry system were calculated as crop and pasture income less production costs. Mallee production costs were calculated for each year. The annual break-even income required from mallee production was calculated by subtracting the mallee production cost and GM of the crop/pasture in the agroforestry system from the GM of the agriculture system.

To compare the agroforestry with the agricultural system over the 6 years of study, an LC analysis was performed by calculating the net present value of the annual break-even income and comparing this to the discounted mallee biomass production, using the following equation:

$$\mathrm{LC} = \frac{\sum_{t=0}^{n} (1+r)^{-t} . A_t}{\sum_{t=0}^{n} (1+r)^{-t} . Y_t}, \tag{2}$$

where LC is the levelized cost, $t$ is time (in years), $A_t$ is the break-even income of the agroforestry in year $t$, and $Y_t$ is mallee biomass yield in year $t$.

A discount rate of 10% was utilized in the calculation of the LC for all scenarios. Sensitivity analysis was performed on the *scenarios b* and *d* at low (7%) and high (13%) discount rates.

Sensitivity analysis was also conducted on $CO_2$e price. There was no price on carbon in Australia in 2006 when this experiment commenced so the minimum price used per Mg $CO_2$e was AUD15 based on the current Australian average price (Clean Energy Regulator, 2020), but AUD30 $Mg^{-1}$ was also evaluated to reflect higher carbon prices elsewhere (Ramstein et al., 2019).

## 3 | RESULTS

The economic analysis presented here shows that the cost of mallee production integrated into an annual farming system in the wheatbelt of WA is driven by seven parameters: (a) site and its productivity, (b) frequency of mallee harvest, (c) season of mallee harvest, (d) the crop/pasture rotation used by the farmer, (e) discount rate, (f) $CO_2$e price, and (g) the method of estimation of below-ground biomass in a coppice system.

### 3.1 | *Scenario a*—Agricultural paddock (base case)

Over the 6 years of this study, the crop/pasture rotations of farmers and the site productivity/seasons, GM from the 100 ha agricultural paddock ranged from a loss of AUD12,922 to a profit of AUD390,226 with an average of AUD114,598 (Table 3). The returns from cropping were consistently greater than from sheep enterprises due to the very low prices for wool and sheep meat over the study period (compare Tables S2 and S3). For instance, at sites 3 and 12, losses were incurred for the 5 years in pasture, yet were profitable for the year in crop (data not shown). Over this study, all other sites were profitable due to returns from 2 or more years of cropping.

### 3.2 | *Scenario b*—Agroforestry utilizing above-ground mallee biomass

Over 6 years, the break-even income required to offset mallee costs ranged from under AUD25,000 at site 5 to nearly AUD90,000 at site 19 (excluding site 16 with a truncated data set) (Table 3) with total fresh biomass production ranging from over 1,500 Mg at sites 1 and 18 to 150 Mg at site 15 (Table 3). There was a large range in productivity across all sites ranging from over 30 Mg $ha^{-1}$ $year^{-1}$ at site 1 to below 5 Mg $ha^{-1}$ $year^{-1}$ at site 15 (Table 3). The LC of mallee biomass production among the 11 sites also varied widely (>6-fold) ranging from AUD40 $Mg^{-1}$ at site 1 to AUD261 $Mg^{-1}$ at site 20 (Table 3). There were also considerable differences in LC of mallee biomass within sites across treatments; however, six of the 11 sites had under 20% difference between treatments.

Table 4 groups and compares sites by harvest treatments: those with a full set of harvest treatments (spring and autumn harvests at 3 and 4 years); those with either 3 or 4 years of harvest across different seasons; and low productivity sites with only one harvest in year 6. The LC were generally higher for spring harvests, an effect that was most pronounced at the low productivity sites (6 years of harvests) with a difference of AUD55 $Mg^{-1}$. Regardless of season of harvest, on average, the LCs of the low productivity sites were double the LCs of the intermediate and high productivity sites. There were also higher LC for the longer harvest frequencies, especially between the 3 years (at AUD68–76 $Mg^{-1}$) and the 6 years of harvest frequencies (at AUD139–194 $Mg^{-1}$).

The cost of mallee in the agroforestry system was split between the direct costs of mallee establishment and maintenance, and harvesting and the indirect opportunity costs from foregone agricultural production on land occupied by the belts and the loss of yield due to mallee crop competition. Averaged across all sites and harvest treatments, competition costs accounted for approximately 38% of total costs, followed by harvest costs (32%), opportunity cost (16%) and establishment and maintenance costs (14%; Table 5 or Table S5 for individual harvest treatment data).

**TABLE 3** Gross margin (AUD) of the solely agricultural paddock and the break-even mallee income (AUD) required to offset the costs incurred by mallee from the agroforestry paddock compared to agricultural paddock over the 6 years of trial. The cost of mallee includes establishment, maintenance, harvesting, opportunity and competition costs. The production of fresh mallee biomass produced and mallee productivity over the trial for all treatments and the levelized cost for above-ground fresh biomass (AUD $Mg^{-1}$) using a 10% discount rate. Treatments varied between sites with either 3 and 4 years of harvest or 6 years of harvest regime. Site 16 had only one 3 years of harvest cycle

| Site | Gross Margin ($) | Season of harvest | Frequency of harvest (years) | Costs of mallee ($) | Productivity (Mg $ha^{-1}$ $year^{-1}$) | Mallee biomass (Mg) | Levelized cost ($ $Mg^{-1}$) |
|---|---|---|---|---|---|---|---|
| 1 | 94,288 | Autumn | 3 | 67,653 | 33.1 | 1,670 | 42.7 |
| | | Spring | 3 | 73,097 | 30.6 | 1,540 | 52.2 |
| | | Autumn | 4 | 61,187 | 32.3 | 1,629 | 40.1 |
| | | Spring | 4 | 58,700 | 22.2 | 1,118 | 58.8 |
| 3 | −12,922 | Autumn | 3 | 37,203 | 9.8 | 891 | 50.1 |
| | | Spring | 3 | 38,656 | 9.6 | 874 | 54.2 |
| | | Autumn | 4 | 35,248 | 8.1 | 743 | 57.9 |
| | | Spring | 4 | 41,843 | 10.4 | 944 | 53.3 |
| 8 | 156,839 | Autumn | 3 | 34,239 | 14.5 | 436 | 82.4 |
| | | Spring | 3 | 38,075 | 14.1 | 423 | 97.3 |
| | | Autumn | 4 | 34,501 | 15.1 | 454 | 81.4 |
| | | Spring | 4 | 40,571 | 15.8 | 475 | 93.1 |
| 13 | 27,752 | Autumn | 3 | 53,783 | 8.8 | 632 | 88.4 |
| | | Spring | 3 | 56,644 | 7.6 | 550 | 110.3 |
| | | Autumn | 4 | 50,708 | 7.0 | 507 | 106.1 |
| | | Spring | 4 | 60,503 | 7.5 | 541 | 116.3 |
| 18 | 65,867 | Autumn | 3 | 74,939 | 18.7 | 1,573 | 48.5 |
| | | Spring | 3 | 77,074 | 19.3 | 1,625 | 48.6 |
| | | Autumn | 4 | 81,789 | 20.4 | 1,717 | 49.0 |
| | | Spring | 4 | 83,290 | 22.4 | 1,884 | 45.6 |
| 19 | 390,226 | Autumn | 3 | 86,257 | 14.8 | 869 | 109.4 |
| | | Spring | 3 | 82,043 | 16.3 | 957 | 94.1 |
| | | Autumn | 4 | 85,522 | 15.4 | 904 | 107.9 |
| | | Spring | 4 | 89,605 | 16.8 | 985 | 103.0 |
| 16 | 17,877 | Autumn | 3 | 16,231 | 17.2 | 309 | 54.1 |
| | | Spring | 3 | 12,107 | 11.2 | 202 | 62.9 |
| 5 | 131,045 | Autumn | 4 | 29,761 | 16.2 | 388 | 82.7 |
| | | Spring | 4 | 23,850 | 9.5 | 227 | 117.9 |
| 12 | −12,922 | Autumn | 6 | 40,371 | 6.2 | 775 | 70.7 |
| | | Spring | 6 | 45,348 | 7.4 | 936 | 64.7 |
| 15 | 146,155 | Autumn | 6 | 40,439 | 8.6 | 308 | 156.3 |
| | | Spring | 6 | 32,246 | 4.2 | 150 | 256.9 |
| 20 | 256,371 | Autumn | 6 | 51,757 | 7.8 | 329 | 190.1 |
| | | Spring | 6 | 45,671 | 5.1 | 216 | 261.0 |
| Average | 114,598 | | | 52,380 | 14.2 | 817 | 91.4 |
| *SD* | 117,291 | | | 21,445 | 7.5 | 510 | 54.3 |
| CV (%) | 100 | | | 40.9 | 52.7 | 62.4 | 59.4 |

These costs, however, are not consistent between sites. Proportion of harvest costs was greatest at sites with high mallee production (sites 1, 3, 12 and 18). The opportunity cost was highest at site 19 which had a very high base-case scenario GM, while it was negative where a focus on sheep production incurred a net loss (sites 3 and 12; Tables S1 and S4). The remaining sites (5, 8, 13, 15, 16 and 20) incurred higher competition costs and were predominately cropped over the study period.

Sensitivity analysis was performed using low (7%), medium (10%) and high (13%) discount rates. This revealed only small differences (1%–4%) in LC among treatments at each site (Table S4). Across sites, the average difference in LC between high discount rate and low discount rate ranged from 1% at site 18 to 20% at site 12 and averaged 9.5% across all sites.

Across all sites and harvest treatments, there was a negative exponential relationship between total mallee biomass production and LC of mallee biomass with a coefficient of determination of 0.50 (Figure 3). This shows that the LC of biomass production is substantially greater at sites with lower productivity due to the diminishing marginal costs of production. There is a floor of LC of AUD58.6.

**TABLE 4** The averaged levelized cost of mallee biomass across sites for each harvest treatment. Sites are separated into groups with the full set of four treatments (frequency and season of harvest), those with either 3 or 4 years of harvests, and the low productivity sites with only 6 year harvest cycles. A discount rate of 10% was applied in the net present value calculation

| Sites | Frequency of harvest (years) | Season of harvest | Levelized cost (AUD $Mg^{-1}$) |
|---|---|---|---|
| 1, 3, 8, 13, 18 and 19 | 3 | Autumn | 70.2 |
| | 3 | Spring | 76.1 |
| | 4 | Autumn | 73.8 |
| | 4 | Spring | 78.3 |
| 5 and 16 | 3 or 4 | Autumn | 68.4 |
| | | Spring | 90.4 |
| 12, 15 and 20 | 6 | Autumn | 139.0 |
| | | Spring | 194.2 |

## 3.3 | *Scenario c*—Agroforestry with above-ground production and below-ground carbon sequestration

When above-ground mallee biomass production plus carbon sequestration in below-ground biomass is considered, the LC of mallee biomass production is reduced (Table 6 or Table S6 for individual harvest treatment data). Compared to *scenario b*, a carbon price of AUD15 $Mg^{-1}$ $CO_2e$ at the average below-ground biomass estimate reduces the LC of biomass production by between 3% and 27% and averaged 12% across all sites and harvest treatments (Table S6). If the $CO_2e$ price is

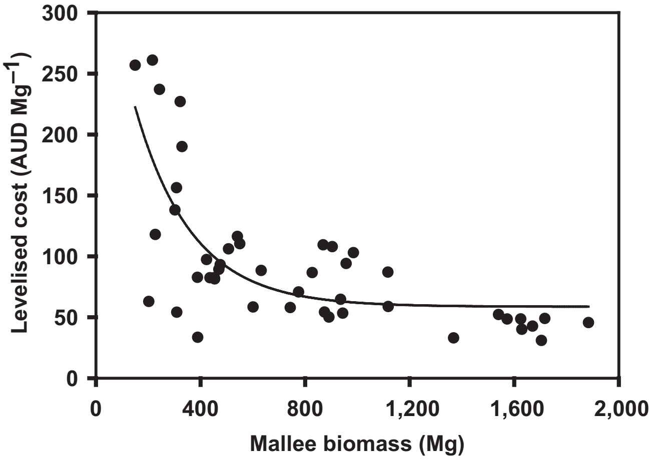

**FIGURE 3** The levelized cost (LC) of fresh above-ground mallee biomass production across all 11 mallee sites and treatments including the unharvested treatments. Line of best fit is a power function, $LC = 58.6 + 326*\exp - 0.005\ Mg$ with a coefficient of determination of 0.50

**TABLE 5** The proportion of levelized cost, averaged across all harvest treatments, that is attributable to direct and indirect costs incurred when introducing mallee into the farming system. The direct cost of mallee includes establishment and maintenance and harvest. The indirect costs consist of the opportunity cost, being the land no longer available for crop, and the competition cost, being income lost from lower crop yields in the competition zone

| | Direct costs (%) | | Indirect costs (%) | |
|---|---|---|---|---|
| Site | Establishment and maintenance cost | Harvest costs | Opportunity cost | Competition cost |
| 1 | 5 | 51 | 12 | 32 |
| 3 | 25 | 50 | −5 | 30 |
| 8 | 10 | 27 | 21 | 42 |
| 13 | 9 | 22 | 6 | 63 |
| 18 | 16 | 47 | 12 | 25 |
| 19 | 10 | 24 | 45 | 21 |
| 16 | 9 | 40 | 8 | 43 |
| 5 | 11 | 25 | 20 | 44 |
| 12 | 42 | 44 | −6 | 20 |
| 15 | 7 | 14 | 24 | 55 |
| 20 | 12 | 12 | 37 | 39 |
| Average | 14 | 32 | 16 | 38 |
| *SD* | 11 | 14 | 16 | 14 |
| CV (%) | 75 | 44 | 100 | 36 |

**TABLE 6** The range of levelized costs (AUD $\text{Mg}^{-1}$) at each site across harvest treatments at a discount rate of 10%. Levelized cost from *Scenario b* is presented for comparison. Sensitivities were performed at AUD15 and AUD30 $\text{Mg}^{-1}$ $\text{CO}_2$e. Sensitivities were also performed on the below-ground carbon biomass estimates with three categories: minimum below-ground biomass (Min BGB), average (Avg BGB) and maximum (Max BGB) over the 6 years of experimental data

| | $0 $\text{CO}_2$e | $15 $\text{Mg}^{-1}$ $\text{CO}_2$e | | | $30 $\text{Mg}^{-1}$ $\text{CO}_2$e | | |
|---|---|---|---|---|---|---|---|
| Site | *Scenario b* | Min BGB ($ $\text{Mg}^{-1}$) | Avg BGB ($ $\text{Mg}^{-1}$) | Max BGB ($ $\text{Mg}^{-1}$) | Min BGB ($ $\text{Mg}^{-1}$) | Avg BGB ($ $\text{Mg}^{-1}$) | Max BGB ($ $\text{Mg}^{-1}$) |
| 1 | 40–59 | 38–58 | 37–55 | 36–54 | 36–56 | 34–52 | 31–49 |
| 3 | 50–58 | 44–52 | 42–48 | 40–45 | 38–45 | 35–39 | 31–33 |
| 8 | 81–97 | 77–92 | 74–91 | 72–89 | 72–88 | 67–84 | 63–81 |
| 13 | 88–116 | 85–111 | 83–109 | 81–107 | 81–106 | 77–101 | 74–97 |
| 18 | 46–49 | 41–44 | 39–43 | 38–41 | 37–40 | 33–37 | 30–33 |
| 19 | 94–109 | 89–104 | 88–102 | 86–100 | 84–98 | 82–94 | 79–91 |
| 16 | 54–63 | 48–55 | 45–51 | 43–47 | 41–47 | 36–39 | 31–32 |
| 5 | 83–118 | 81–116 | 79–112 | 76–109 | 79–114 | 75–105 | 70–99 |
| 12 | 65–71 | 56–57 | 48–50 | 43–44 | 43–46 | 29–32 | 16–21 |
| 15 | 156–257 | 153–253 | 151–250 | 149–245 | 150–250 | 146–243 | 141–234 |
| 20 | 190–261 | 166–233 | 154–211 | 146–195 | 141–206 | 117–162 | 102–128 |

**TABLE 7** Productivity of unharvested mallee belts and the levelized cost for above-ground fresh biomass. Total Mg $\text{CO}_2$e generated and $\text{CO}_2$e productivity of above- and below-ground biomass of over 6 years at each site and the levelized cost with a 10% discount rate. Bold figures indicate sites that would be profitable with the current price of $\text{CO}_2$e (AUD15 $\text{Mg}^{-1}$). Site 16 only included 3 years of data

| | *Scenario b* | | | | *Scenario d* | | |
|---|---|---|---|---|---|---|---|
| Site | Productivity (Mg $\text{ha}^{-1}$ $\text{year}^{-1}$) | Total cost of mallee ($) | Mallee biomass (Mg) | Levelized cost (AUD $\text{Mg}^{-1}$) | Total $\text{CO}_2$e (Mg) | $\text{CO}_2$e productivity (Mg $\text{ha}^{-1}$ $\text{yr}^{-1}$) | Levelized cost (AUD $\text{Mg}^{-1}$ $\text{CO}_2$e) |
| 1 | 27.1 | 34,345 | 1,368 | 33.0 | 2,526 | 50.1 | **11.1** |
| 3 | 6.6 | 21,604 | 601 | 58.4 | 1,138 | 12.5 | 19.2 |
| 8 | 10.1 | 32,707 | 302 | 138 | 506 | 16.9 | 51.1 |
| 13 | 11.5 | 57,008 | 827 | 86.6 | 1,666 | 23.1 | 26.7 |
| 18 | 20.3 | 45,256 | 1,704 | 30.9 | 2,804 | 33.4 | **11.7** |
| 19 | 19.0 | 75,349 | 1,117 | 87.0 | 1,825 | 31.0 | 33.1 |
| 16 | 10.8 | 12,486 | 389 | 33.5 | 889 | 49.4 | **12.1** |
| 5 | 18.9 | 29,759 | 453 | 82.4 | 922 | 38.4 | 25.2 |
| 12 | 3.7 | 26,313 | 470 | 89.3 | 877 | 7.0 | 29.7 |
| 15 | 8.9 | 58,451 | 322 | 227.0 | 889 | 24.7 | 63.8 |
| 20 | 5.8 | 46,138 | 243 | 237.0 | 911 | 21.7 | 39.3 |
| Average | 13.0 | 39,947 | 709 | 100.0 | 1,359 | 28.0 | 29.4 |
| *SD* | 7.3 | 18,486 | 487 | 72.5 | 749 | 14.1 | 16.9 |
| CV (%) | 56.4 | 46 | 69 | 72.3 | 55 | 50.2 | 57.0 |

increased to AUD30 $\text{Mg}^{-1}$, this decreases the LC by between 6% and 54% with an average of 23%.

The difference in LC between the minimum and the maximum root biomass estimates averaged 8% or 17% $\text{Mg}^{-1}$ $\text{CO}_2$e price of AUD15 or AUD30 respectively (Table S6). This ranged between 3% and 23% across all sites and treatments.

### 3.4 | *Scenario d*—Agroforestry with unharvested mallee sequestering carbon in AGB and BGB

The total above-ground mallee biomass produced over the 6 years of the trial ranged from 1,704 Mg at site 18 to 243 Mg at site 20 and averaged 709 Mg across all sites (Table 7), approximately 110 Mg less than the average harvested treatments from *scenario b*. For the unharvested belts, the undiscounted break-even mallee income ranged from AUD21,604 at site 3 (excluding site 16 with truncated data) to AUD75,349 at site 19, with an average of AUD39,947 across all sites (Table 3). This was about AUD12,000 less than the harvested treatments from *scenario b* mainly driven by the absence of harvest costs.

The LC of the unharvested belts under *scenario b* methodology ranged from AUD33 $Mg^{-1}$ at site 1 to nearly AUD240 at site 20 (Table 7). Compared to the harvested belts, the LC of the unharvested mallee were cheaper at 6 of the 11 sites (cf. Tables 3 and 7). If mallee is grown solely to generate above- and below-ground carbon credits, then the LC ranged between AUD11 and AUD64 and averaged AUD29 $Mg^{-1}$ $CO_2e$, a reduction across all sites ranging from 62% at both sites 18 and 19 and up to 83% at site 20. Lower LC were realized at sites with higher $CO_2e$ productivity.

Across all sites, greater differences were observed between discount rates for the unharvested mallee agroforestry compared to the harvested mallee agroforestry (generally > 15%; Table S4). The proportion of costs of unharvested mallee belts was considerably different to the harvested mallee with higher average costs (66%), attributable to competition (Table S5).

## 4 | DISCUSSION

Understanding the economic consequences of integrating mallee belts into annual crop/pasture farming systems is essential for mallee agroforestry development. The data presented here show large site and regional differences in the LC of mallee biomass production or carbon credit production, but less variation arising from the management choices of season or frequency of harvest.

Mallee agroforestry systems can generate direct income by selling biomass, $CO_2e$ or both. Under the Australian Carbon Farming Initiative, sequestration projects can generate carbon credits over 25 years of period, although the net abatement of $CO_2e$ is reduced by 20% if the planting is removed before 100 years (Department of the Environment, 2015) and this applies to above- or below-ground biomass components. Over the trial, the above- and below-ground carbon sequestration by unharvested mallee would be profitable given current Australian $CO_2e$ prices at three of the 11 trial sites. At AUD30 $Mg^{-1}$ $CO_2e$, mallee agroforestry would have been profitable at seven sites.

In WA, crop and sheep enterprises generally generate annual positive cash flows while a coppice harvest regime for mallee generates periodic positive cash flows after harvest. This may well affect the willingness of landholders to grow the mallee or provide land to third parties to plant and harvest the mallee under a lease agreement. Given the 2006–2011 agricultural GM, four of the 11 study sites had a LC of mallee biomass production in the range AUD40–60 $Mg^{-1}$. These sites were generally characterized by high biomass production or moderate biomass production with low agricultural GM. This price range may be economically attractive to farmers to sell into biomass processing markets to take advantage of the on-farm benefits of mallee crops. The remaining seven sites had levelized biomass costs ranging from AUD70 $Mg^{-1}$, with two sites exceeding AUD200 $Mg^{-1}$, and were less commercially attractive. There was a reduction in LC when below-ground biomass was used to generate carbon credits especially at AUD30 $Mg^{-1}$ $CO_2e$, which although nearly double the current Australian price, is comparable to the price in some large carbon credit markets around the world (Ramstein et al., 2019).

Some caution needs to be exercised with these numbers as the opportunity cost and consequent LC of mallee biomass production was heavily influenced by crop/pasture rotation decisions of the landholders, with lower opportunity and competition costs associated with sheep grazing due to low wool and sheep prices over the study period (cf. Tables S2 and S3). This resulted in some sites with low biomass production with a low LC because the sites were in pasture for 5 of the trial 6 years. Conversely, two sites were moderately productive but had high LC due to high proportion of years where growers chose to grow grain crops. In the intervening years, there has been a substantial increase in returns for wool and sheep meat producers.

The sites with the lower LC were consistent with previous work on mallee economics. Abadi et al. (2012) estimated a range of AUD44–55 $Mg^{-1}$ for biomass at the farm gate, or AUD53–70 $Mg^{-1}$ including off-farm transport and supply chain costs. McGrath et al. (2016) showed that, excluding harvesting and delivery costs, mallee agroforestry would be marginally economic from AUD24 $Mg^{-1}$, but AUD34 $Mg^{-1}$ was required for large-scale adoption.

There are on-farm and natural resource management benefits of mallee integration including: dewatering the soil profile below and adjacent to belts (Robinson et al., 2006; Sudmeyer & Goodreid, 2007; Wildy et al., 2004) with potential to enhance salinity mitigation (Clarke et al., 2002; George, 1990); erosion control and provision of shade and shelter for stock which is especially useful during lambing (Abadi et al., 2012; Baker et al., 2018) and provision of shelter for crops (Baker et al., 2018; Bennell & Verbyla, 2008; Sudmeyer et al., 2002). Abadi et al. (2012) estimated the value of these benefits was between AUD2 and AUD13 per fresh Mg of mallee biomass produced, excluding payment for carbon sequestration. About 75% of the upper estimate was associated with mitigation of waterlogging which is only frequent on particular soil types and in higher rainfall growing season (May–October in WA) and is becoming less common as average rainfall in the south-west of WA is diminishing (Asseng & Pannell, 2013). In estimating the required price per Mg of $CO_2$e to make agroforestry viable for carbon farming, Flugge and Abadi (2006) modelled the value of salinity mitigation at AUD5 $Mg^{-1}$ $CO_2$e.

The quantity of biomass produced per unit area has a large effect on the LC. The biomass productivity achieved at each site is a combination of several quantifiable factors, including season and frequency of harvest (Spencer et al., 2019) and planting configuration (number of rows, between row spacing and alley widths; Spencer et al., 2020). There are some less quantifiable factors, including reconfiguration of paddock shape, size and infrastructure to better integrate mallee belts. For instance, gains in mallee productivity could be realized by including small (40–50 cm) water retention bunds to capture any surface water flow. Experimental data show that after 3 years, belts with bunds produced 35% more biomass (Bennett et al., 2015). Spencer et al. (2019) found edaphic factors (EC, pH and nutrition) were strong predictors of productivity across the sites in this study. This decadal research project reveals declining mallee productivity with proximity to shallow saline water tables, and alkaline and nutrient-poor soils profiles. To reduce opportunity costs, mallee species have often been allocated suboptimal landscape positions, generally into saline valley floors. This economic analysis shows that, assuming a market for biomass, this paradigm should be questioned, with mallee capable of delivering greater financial reward to the landholder when planted in productive sites. Prospective mallee species have a range of site preferences indicating that matching species to site will be important in maximizing production and economic viability (Eastham et al., 1993; Wildy et al., 2000).

Mallee species productivity can be influenced by the season of harvest, with Spencer et al. (2019) showing $E_{koc}$ more productive following autumn harvest, $E_{lox}$ following spring harvest and no significant seasonal response for $E_{pol}$. This study showed that spring harvest resulted in higher LC of production (AUD83.4 $Mg^{-1}$) compared to autumn harvest (AUD99.4 $Mg^{-1}$). It supports Sudmeyer et al. (2012) who found that adjacent crop competition by mallee was reduced when harvest was undertaken before the growing season (i.e. in autumn) for both initial and second harvest. The mallee belts used in this study were between 5 and 12 years old before the initial harvest and therefore had well-advanced root systems with considerable lateral reach and depth (Robinson et al., 2006; Sudmeyer & Goodreid, 2007). Depletion of stored soil water by mallee prior to spring harvest would have increased competition between mallee and the adjacent annual crop, as well as exposing the mallee belt to harsher coppice regeneration conditions going into the dry summer, thereby increasing the spring LC. However, any large-scale mallee industry is likely to only be viable if it can deliver a continuous supply of biomass (Enecon, 2001) and growers may be limited in their choice of harvest season. To reduce competition costs, with increasing mallee size, the grower could increase the width of the exclusion zone; only cropping where returns are greater than input costs (Sudmeyer et al., 2012).

This study also demonstrates that longer harvest intervals increase LC. There was only a slight increase in LC when comparing the 3 years of harvests to the 4 years of harvests, but there was a much greater difference when comparing the 3 or 4 years of harvests to the 6 years of harvests. This is consistent with the finding that competition is positively correlated with tree height (Sudmeyer et al., 2002, 2012). The longer harvest frequencies will result in delayed returns from mallee production and a lower net present value.

Harvest costs account for almost a third of the total cost (32%) of mallee biomass production. These estimates were based on mallee harvesting using conventional forestry equipment. This study assumed a fixed harvesting cost, which would underestimate the cost of harvest at the sites with less standing biomass because harvest costs have been found to be dependent on the standing biomass per km of belt (Spinelli et al., 2014). A prototype single-row chipper–harvester has been developed to reduce harvest cost using technology capable of processing the high wood density and multiple stems of mallee (Abadi et al., 2012; Goss et al., 2014). Harvesting single rows would be more cost-effective for single or double row belts.

Reducing belt width (i.e. number of rows) can reduce LC by increasing mallee productivity and reducing opportunity costs. Wider belts (more rows) take up more paddock area and internal rows are suppressed by the larger trees in the external rows which have greater access to additional resources from the alley (Huxtable et al., 2012; Prasad et al., 2010; Spencer et al., 2020). Consequently, the internal rows have reduced the productivity per hectare of the belt. Fewer rows, or wide between-row spacing, may allow for shorter harvest frequency intervals and generate earlier positive cash flows for investors with larger discount rate. Increased harvesting frequencies will also improve cash flow. Fewer rows will also reduce establishment and maintenance costs, and if using a single-row chipper–harvester, could further reduce harvest costs.

Results from this study rely on the accuracy of BGB estimates from allometric equations. The 'best' current model for estimating below-ground biomass of mallee is not species specific and uses mallee height which alone explains less than 50% of actual biomass (Paul et al., 2014). Large species differences have been found in unharvested root/shot ratios of the mallee species used in this study (Brooksbank & Goodwin, in press) which are likely to persist post-harvest. Furthermore, the allometric models are likely to underestimate below-ground biomass because the models do not take into consideration the likely increase of biomass with subsequent harvests. A below-ground mallee root biomass conceptual model was proposed by Bartle and Abadi (2010) who suggest that below-ground biomass accumulates over time. This arises from the loss of fine root biomass with harvest (Wildy & Pate, 2002) and the considerable depth to which mallee roots can penetrate (Nulsen et al., 1986), and over regular harvests, additional woody root biomass sequestered between harvests would likely persist. Currently, no mallee allometry exists over multiple harvest cycles and further research is required to provide greater confidence in the below-ground biomass estimates.

Future research is required for multi-criteria mapping of the WA wheatbelt to locate land which could most benefit from mallee integration. For instance, such criteria include targeting areas that are most in need of salinity mitigation, with high suitability for mallee productivity, and where farmers could benefit from having shelter for sheep breeding. Such assessments have been undertaken for the agricultural sector in WA (DAFWA, 2013; Schoknecht, 2015) and could be adapted for mallee. For instance, in comparison to agricultural crops, mallee can tolerate and respond better to acidic soils (Spencer et al., 2019; Symonds et al., 2001). This assessment would also help investors who, for example, are looking for carbon offset projects, to have more confidence with where to grow mallee and the level of compensation required for landholders.

There are distinct advantages for both the coppice and unharvested system. Mallee are capable of stable biomass production with regular harvests (Davis, 2002; Spencer et al., 2019) but without harvest, the growth rates will slow reducing the rate of carbon sequestration while increasing competition to agriculture. Cash flows from the coppice system will occur with harvests, likely every 3–4 years, but in large operations, harvesting could be structured to provide annual income, although this will add annual costs for mobilizing harvesting equipment. Under current legislation, payments from sequestration occur at agreed reporting periods between 6 months and 5 years (Department of the Environment, 2014). The markets for biomass and carbon credits will ultimately determine whether the mallee will be harvested or left without harvest for 25 or 100 years, with our modelling suggests could be profitable based on carbon price.

## 5 | CONCLUSION

Mallee, integrated into a farming system, imposes additional costs on farmers, especially through competition and harvest costs, and, to a lesser extent, opportunity and establishment costs. For widespread adoption, farmers will require markets for biomass or carbon credits that equal or exceed the profitability of traditional agriculture.

Our estimates show that mallee can cost farmers from AUD40 to over AUD250 Mg$^{-1}$ of fresh biomass to produce. Lower LC are realized at sites with high mallee growth rates. The second most important determinant of LC was the relative returns from agricultural activities.

The LC could be reduced by 11% on average, if below-ground biomass was sold at the present $CO_2e$ price in Australia. More accurate allometric models are required to estimate below-ground biomass, especially over multiple harvests. If Australia's $CO_2e$ price were aligned with other developed nations at AUD30 Mg$^{-1}$, the LC would be halved. Given the current carbon prices, the price generated by carbon from unharvested mallee at high productivity sites is already comparable with agricultural returns.

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## CONFLICT OF INTEREST

The authors declare no conflicts of interests in the subject matter discussed in this manuscript.

## AUTHORS' CONTRIBUTIONS

J.B. and R.S. designed the experiment responsible for the experimental data used in this study. B.S. and A.A. conceived the conceptual design of the study. S.V.G. and B.S. built the economic model. B.S. drafted the manuscript. R.S., S.V.G, J.B., A.Z., A.A. and M.G. contributed to the final version of the manuscript.

## DATA AVAILABILITY STATEMENT

Most data are available in article supplementary material or in these cited articles: Spencer et al. (2019) or Sudmeyer et al. (2012). Where the data are not available, data will be made available on reasonable request from the authors.

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## SUPPORTING INFORMATION

Additional supporting information may be found online in the Supporting Information section.

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# Chapter 7

## Conclusion

This thesis investigated many aspects of mallee agronomy and breeding with the view to inform industry of the potential for mallee to be an economically viable crop in the Western Australian (WA) wheatbelt. After nearly 30 years of research into mallee, both interest and funding from the government sector ceased in the middle of last decade. Recently however, there has been renewed enthusiasm for climate change mitigation arising from the Paris Agreement and with many industrialised countries declaring zero-net-emissions targets by 2050. The agricultural sector has been highlighted by the Australian government as having a large capacity for carbon sequestration and biofuel production (Climate Change Authority, 2019) based on the conversion of cellulosic matter to hydrocarbons, a demonstrated replacement for conventional fuels (Azad *et al.,* 2015).

This thesis addresses key knowledge gaps in four broad areas that may otherwise discourage or delay industry engagement:

(i) advances in mallee breeding, primarily focused on biomass gains (chapters 4 and 5), 1,8-cineole production (chapter 4) and the genetic parameters of flowering phenology (chapter 4);

(ii) accurate and standardised mallee yield estimates (chapter 2);

(iii) silvicultural and agronomic factors to maximise mallee productivity including response to harvest management (chapter 2 and 6), site characteristics (chapter 2), and planting configuration (chapter 3); and

(iv) economic consequences for landholders from mallee agroforestry (chapter 6).

*Mallee breeding – biomass, 1,8-cineole and reproductive phenology*

Many mallee species have been screened and several have been selected for suitability as a tree crop over large areas of the Western Australian wheatbelt (Olsen *et al.*, 2004; Hobbs *et al.*, 2009). The selected species also have potential to be suitable crops in other dryland agricultural regions of Australia although this awaits rigorous evaluation. Based on two traits, foliar 1,8-cineole concentrations and superior growth rates, mallee research has been narrowed to three taxa: *E. loxophleba (*subsp. *lissophloia* and *gratiae)*; *E. polybractea*; and *E. kochii* (subsp. *plenissima* and *borealis*) (Bartle, 2009). These taxa, have shown potential to dewater soil profiles (Wildy *et al.*, 2004; Robinson *et al*., 2006) and can be used as a tool to help mitigate dryland salinity and other landscape-scale sustainability issues. However, estimates of the proportion of catchment required to be revegetated to control salinity are up to 70-80% (George *et al.*, 1999). Thus, for an industry to emerge, mallee will need to be profitable to the landholder as a bioenergy or carbon crop.

The commercialisation of mallee for biomass is primarily going to be driven by the productivity of mallee plantings. This can be achieved in two ways: firstly, selecting suitable sites and regions for mallee cropping and secondly, having improved genetic stock that is capable of high productivity.

This thesis aims to address important aspects for mallee breeding by estimating the genetic parameters of key mallee traits: i.e. sapling and coppice biomass, and foliar 1,8-cineole concentration. Chapter 4 assesses the key reproductive traits and the genetic correlations with biomass and 1,8-cineole concentration at an *E. loxophleba* seed orchard; and Chapter 5 estimates the productivity of sapling and coppice biomass at three *E. polybractea* progeny trials.

Synchronicity of flowering is important in seed orchards as it facilitates transfer of genes to the next generation. Historically the flowering of semi-arid eucalypt species has not attracted much research interest. A *E. loxophleba* seed orchard containing nine provenances from both subsp. *lissophloia* and *gratiae*, was subject to two annual assessments. Flowering peaked in spring but a high proportion of trees from the eastern provenances with the two lowest rainfall regions flowered twice in an assessment year (28 to 34% respectively). Water availability is the likely cause of the additional reproductive event, given rainfall has been observed to induce anthesis in species from arid environments (Davies, 1976; Friedel *et al.*, 1993). Out-of-phase flowering will reduce outcrossing potential. Since the provenances displaying out-of-phase flowering also tended to be poor biomass producers, it would be desirable to remove them from the breeding population. About 20% of the southern *gratiae* subspecies flowered twice in one assessment year and this, coupled with the risk of genetic pollution from farmland plantings in the vicinity of native *E. lissophloia* (Sampson & Byrne, 2008), suggests that this subspecies should be split into a separate breeding populations to service the southern WA wheatbelt. The remaining four *E. loxophleba* subsp. *lissophloia* provenances from the central wheatbelt should be represented in future breeding populations to service the central and northern WA wheatbelt.

In the *E. loxophleba* seed orchard, heritability of biomass productivity was initially weak (0.10 ± 0.03) prior to thinning but increased to 0.29 ± 0.08 post thinning. This may have arisen from bias introduced as a result of the thinning, and the post-thinning estimate and should be treated with caution. However, the pre-thinned estimate revealed that the timing of flowering, based on the genetic correlations, can influence gains in biomass: i.e. earlier commencement and termination of flowering is correlated with larger biomass and higher 1,8-cineole concentration. Biomass and foliar 1,8-cineole concentrations were weakly positively correlated and thus selection for one trait should result in corresponding gains in the second trait via index selection. These findings differ to those from three progeny trials for both *E. loxophleba* subsp. *lissophloia* and *E. loxophleba* subsp. *gratiae* where weak negative genetic correlations were found between biomass and foliar 1,8-cineole concentration (Mazanec *et al.*, 2017; Mazanec *et al.*, 2020). This has important implications for a mallee industry where extraction of 1,8-cineole can provide a useful additional revenue stream.

The heritability and genetic parameters of biomass were estimated at three *E. polybractea* progeny trials located at multiple locations in WA, New South Wales (NSW) and Victoria. After harvest, very high survival rates were observed at all three sites of (>98%) which is higher than other harvested trials of this species (Milthorpe *et al.*, 1994; Milthorpe *et al.*, 1998; Wildy *et al.*, 2000). Surviving mallee exhibited a range of responses to harvest; some families were either elite or poor performers across sites and biomass assessments, whereas others performed better as either saplings or coppice. Across the three states, the strength of heritabilities varied across the sapling and coppice assessments. However, high genetic correlation between the sites revealed that, although there were large site differences, the trials could be treated as one breeding population. This indicates that seed from each of these progeny trials is appropriate for use across a wide range of environments, including the semi-arid zones in WA, NSW, Victoria and probably South Australia (SA).  This will reduce cost of mallee seed production with no requirement to maintain different breeding populations for different regions.

Selection simulations to convert progeny trials to seed orchards revealed that gains could be made when selecting for biomass yield for either sapling (17% to 24%) or coppice (8% – 15%). However, negative or lower gains (-7% to 13%) were obtained when sapling selections were applied to coppice biomass estimates. This demonstrates that to optimise biomass yield of *E. polybractea*, selections for coppice production should be based on coppice biomass data. This has implications for the *E. polybractea* breeding program previously established by the Department of Conservation of Land Management of Western Australia where all biomass breeding decisions were made using sapling biomass data. These results indicate that any future breeding program incorporating seed from wild parent trees will need take into consideration the time required for establishment, harvest at 5 to 6 years of age followed by 3 to 4 years of coppice growth before initial selection and thinning, and an additional 3 to 4 years for seed production to be realised. As an interim measure, prior to establishment of new breeding populations, the three progeny trials in this study could immediately be converted into seed orchards with the execution of a thinning program.

For other species of plantation eucalypts, there has been a range of genetic correlations between sapling and coppice biomass, for instance, *E. globulus* has a shown weak correlation (Whittock *et al.*, 2003) whereas strong correlations were found for *E. dunnii* (Li et al., 2012), *E. grandis*, *E. urograndis, E. saligna* and *E. urophylla* (Amâncio *et al.*, 2020). The weak correlation for *E. globulus* has been attributed to the variation of lignotuber size and development between sub-races (Whittock *et al.*, 2003). The other mallee species under investigation in this thesis, *E. loxophleba* and *E. kochii*, are both known to coppice strongly after harvest (Eastham *et al.*, 1993; Wildy *et al.*, 2000) and it is anticipated that selections at coppice age will improve biomass productivity for these species.

### *Mallee yield*

Mallee yield varies across species, climatic zones and site conditions. Thirty years of trial planting have shown that the two *E. loxophleba* subspecies grow well across most of the wheatbelt but exhibit slow growth rates around the southern coast. *Eucalyptus polybractea* suffers progressively more serious drought stress and mortality with the decreasing rainfall and increasing evaporation in transition to the north half of the WA wheatbelt at around 31° south latitude. The northern wheatbelt is the preferred region for the two *E. kochii* subspecies. However, research has shown large variation in growth rates both between and within regions and across sites, making accurate estimates of mallee biomass yield difficult to predict. To compound this, early assessment of biomass productivity did not use standardised methodologies.

The imprecise estimates of mallee biomass have the potential to hamper industry development. To address this, Chapter 2 of this thesis reports on the biomass yield potential of the three main mallee taxa subject to four harvest regimes with uncut controls and uniform methodologies at 19 sites over 10-years. *Eucalyptus polybractea* was the fastest growing species across the trial sites, with biomass production ranging from 7 to 33 dry $Mg^{-1}$ $ha^{-1}$ $year^{-1}$, which is in agreement with findings from the review carried out by Huxtable *et al.* (2012). Most of these sites were in the higher rainfall areas with deep soil profiles.

There was substantial variation between the *E. loxophleba* sites with three of twelve sites deemed to have failed due to high mortality and low growth rates following the initial harvest. At the failed sites, the harvested biomass yield ranged from 0.1 to 6.1 dry $Mg^{-1}$ $ha^{-1}$ $year^{-1}$. However, in the unharvested treatments at the same sites, growth rates of up to 18 dry $Mg^{-1}$ $ha^{-1}$ $year^{-1}$ were achieved. The failed sites were all characterised by shallow soil profiles which at two sites were confined by a saline groundwater and at the third, a silcrete hardpan. This demonstrates that, although moderately tolerant to both saline ground water and drought, *E. loxophleba* has hard limits to its tolerance of saline soils of $EC_e$ below 15 dS $m^{-1}$ when subjected to even a single harvest. However, unharvested *E. loxophleba* has been known to use saline groundwater up to 16.1 dS $m^{-1}$ (Brooksbank, 2011) and up to 30 dS $m^{-1}$ (Pepper & Craig, 1986). If landholders decide to plant *E. loxophleba* in these saline areas, substantial biomass may be produced, but if left for a considerable time (>15 years), the large size of trees could reduce harvestability with an increased risk of high post-harvest mortality. Saline perennial grazing systems might prove a better option at such locations (Bennett *et al.*, 2009; Revell *et al.*, 2013).

At the remaining nine *E. loxophleba* sites, observed yield ranged from 4 to 12 dry $Mg^{-1}$ $ha^{-1}$ $year^{-1}$. The sites that exhibited the lowest growth rates were north of Esperance (-33.52°, 122.15°). Those results support other observations that *E. loxophleba* performs poorly on sites along the WA southern coast (Wildy *et al.*, 2000). Due to the salt-tolerance of *E. loxophleba*, landholders have historically used this species around saline valley floors, including at most of the sites within this study, suggesting that the growth production potential of this species may not be reflected in this chapter. This was demonstrated at a site near Buntine (-30.97°, 116.48°) which traversed three separate landscape positions. When examining the data from each individual treatment and replicate, productivity ranged from 1 to 20 dry $Mg^{-1}$ $ha^{-1}$ $year^{-1}$ over a distances of 540 m. The higher biomass estimate of 20 dry $Mg^{-1}$ $ha^{-1}$ $year^{-1}$ might be a better reflection of the production potential of *E. loxophleba*. The substantial variation at this site suggests the need for careful site selection to locate future plantings for a commercially viable mallee planting.

*Eucalyptus kochii* demonstrated high variability of growth rates, which ranged from 4 to 16 dry $Mg^{-1}$ $ha^{-1}$ $year^{-1}$ similar to that found elsewhere (Huxtable *et al.*, 2012). These sites were all in the northern wheatbelt and ranged from saline valley floor to upland sites. An upland site displayed the highest growth rates where hydrological drilling located perched freshwater aquifers (Bennett *et al.*, 2005). At these sites, the mallee were subjected to three 3-year harvest cycles and productivity dropped following each harvest cycle from 18 to 15 to 11 dry $Mg^{-1}$ $ha^{-1}$ $year^{-1}$ potentially due to diminishing availability of the fresh sub-soil water. Productivity was lowest at the site with silcrete hardpans and saline ground water which revealed slow growth rates ranging from 3 to 5 dry $Mg^{-1}$ $ha^{-1}$ $year^{-1}$.

This work reveals the large variation of site potential across the landscape and supports earlier work that *E. polybractea* has high growth rates (Wildy *et al.*, 2000; Brooksbank, 2011). However, the *E. polybractea* sites were generally less saline and located in higher rainfall areas than the majority of the *E. loxophleba* or *E. kochii* sites (Peck *et al.*, 2012). *Eucalyptus loxophleba* is capable of fast growth rates but at many sites, growth rates have been compromised due to shallow saline groundwater, which when combined with harvest, can result in high mortality. This study consisted of many sub-optimal *E. loxophleba* sites and increase in production is likely to occur if planted in more favourable conditions. *Eucalyptus kochii*, when planted in areas with constrained soil profiles has slow growth rates but can be productive in upland slopes, especially with access to fresh groundwater. This species is well suited to the lower rainfall areas within the wheatbelt. All three species are capable of harvest cycles of 3-7 years, with eight of the nine sites subject to three 3-year harvest showing no progressive decline in biomass production. This demonstrates that on favourable sites, stable production is likely over at least three harvest cycles with little risk of failure.

The three mallee species studied in this thesis have been found to be well suited to various landscape positions and rainfall zones for large-scale revegetation across the WA wheatbelt. However, these species could also be used across much of southern Australia for biomass production or carbon sequestration. *Eucalyptus polybractea*, naturally occurs in northern Victoria and southern central NSW and has been used for cineole production in the eastern states for over 100 years (Davis, 2002). Trials of *E. kochii* have successfully occurred in NSW (Milthorpe *et al.*, 1998). Recently, progeny trials of *E. loxophleba* subsp. *gratiae* and *lissophloia* have been successfully established in Condobolin (NSW) and Monarto (SA) and are suitable candidates for adoption across southern Australia (Mazanec *et al.*, 2017; Mazanec *et al.*, 2020).

### *Site characteristics*

The WA wheatbelt has dry hot summers with declining winter rainfall (Frederiksen & Grainger, 2015). For tree crops in this semi-arid area, sunlight is in abundance, precipitation is irregular but there are opportunities for site selection to capture run-off (Bennett *et al.*, 2015), deeper sub-soil water flows and storage (Wildy *et al.*, 2004; Robinson *et al.*, 2006; Brooksbank *et al.*, 2011). Nutrients are widely limiting in the ancient soils of the WA wheatbelt, but usually corrected by annual crop agriculture (Schoknecht, 2015).

Across the 19 trial sites from Chapter 2, the primary factor determining productivity was the unique set of conditions of each site. More than half (56%) of the variation in productivity of unharvested treatments across sites was related to three soil properties; $EC_e$, pH, and percent organic carbon. More productive sites were characterised by non-alkaline soil (pH <8), low soil salinity ($EC_e$ <15 dS $m^{-1}$) and high organic soil carbon. For coppice biomass, 72% of the variation was explained by the three variables above, with the addition of soil nitrogen ($NH_4^+$ plus $NO_3^-$) and maximum daily temperature. These results support earlier findings: the mallee species selected for commercialisation are hardy, but can be sensitive to unfavourable conditions (Wildy *et al.*, 2000; Brooksbank, 2011; Brooksbank *et al.*, 2011). This work identified soil pH, $EC_e$ and fertility as being important determinants of mallee productivity. Thus, in future mallee plantings, consideration of these factors and locating plantings at sites with soil pH <8, ECe < 15, high percentage of organic carbon and total nitrogen, and saline water table at > 4 m will increase productivity.

For long-term mallee harvest systems, nutrient management will likely be necessary to prevent declining yields. Mallee production did not decline at eight of the nine experimental sites that underwent 3-year harvests treatments from Chapter 2, but soil nitrogen ($NO_{3-}$ plus $NH_{4+}$) was significant in predicting productivity from the harvest treatments while being non-significant in the unharvested treatments. The productivity impacts may be due to soil nitrogen concentration decreasing as a result of harvest. At a 5-year old E. polybractea trial yielding 25 Mg $ha^{-1}$,Grove *et al.* (2007) found that the harvested biomass contained approximately 189 kg $ha^{-1}$ of nitrogen and 12kg ha- of phosphorus. These nutrients are removed from the system following harvest and if not returned through application of ash, biochar (after biomass processing) or fertiliser, productivity may decline. The addition of fertilisers, especially nitrogen, have been found to contribute 59 to 72% of the total energy input, and 70 to 81% of greenhouse gas emissions from a mallee life cycle analysis (Yu *et al.*, 2015). Using ash or biochar will reduce the input and emissions, but any inputs must be considered when regarding the carbon neutrality of any mallee biomass system.

### *Harvest effects*

There was a species-dependent response to season of harvest with *E. loxophleba* responding better to spring harvests whereas *E. kochii* was more productive following autumn harvests. This is likely due to the faster re-shooting from *E. loxophleba* which is able to increase the leaf area and utilise the available soil moisture. *Eucalyptus kochii* takes longer to achieve the same amount of leaf area and autumn harvests allow more growth before the peak growing period in spring and early summer. Further, *E. kochii* responds to drought by reducing leaf area, where in contrast, *E. polybractea* will increase root growth (Brooksbank, 2011). *Eucalyptus polybractea* was unaffected by season of harvest.

Mallee are sensitive to short harvest intervals (6 months to 2 years) especially after autumn and summer cuts (Wildy *et al.*, 2000; Wildy & Pate, 2002; Noble & Diggle, 2014). However, in this chapter, frequency of harvest had less impact than season of harvest which was probably due to the longer harvest intervals imposed at the less productive sites. For *E. loxophleba*, there was a season and frequency of harvest interaction with higher productivity from autumn harvest under short harvest interval whereas the spring harvest benefited from long harvest intervals. *Eucalyptus kochii* were slower to coppice following harvest with more biomass produced in the uncut treatments compared to the harvested treatments. The opposite trend was observed for the *E. polybractea* sites with faster growth rates for the harvest treatments. There was no consistent response at the *E. loxophleba* sites, with five of the eight sites producing higher productivity under harvest.

The seasonal variation in harvest response indicates the need for careful timing of harvest. *Eucalyptus polybractea* is quite tolerant of both spring and autumn harvest, whereas *E. loxophleba* has better yields when subject to spring harvest and *E. kochii* to autumn. Autumn harvests will reduce the competition with adjacent crops and pastures (Sudmeyer *et al.*, 2012) which suggests that *E. polybractea* and *E. kochii* can be harvested to concurrently optimise yield and reduce competition with agriculture.

Biomass harvest of 3.5 year-old coppice from three *E. polybractea* progeny trials were assessed and significant family effects were observed. This demonstrated a family response to harvest indicating that not all families are well adapted to coppice production. Future planting of *E. polybractea* should utilise seed sourced from a breeding program that has selected for biomass on coppice biomass at harvest age.

### *Planting configuration*

Mallee belts have been planted in a wide range of row number and within row spacing but have mainly been 2-4 row belts with 2 m between rows and 1.5 m within-row spacing (URS, 2008). Wider belts (>2 rows) have resulted in suppressed growth rates (Huxtable *et al.*, 2012) yet an assessment of the effect of spacing has not been conducted. Here we sought to determine the planting configuration (number of rows and within-row spacing) that optimise productivities. A *E. polybractea* and a *E. loxophleba* spacing experiment comprising 20 different planting configurations yielded productivity ranging from 2.7 to 21.2 dry Mg$^{-1}$ ha$^{-1}$ year$^{-1}$. Productivity was highest in narrow belts with a biomass yield penalty arising from the addition of inner rows. In the wider belts, the internal rows underwent growth suppression which has been observed at mallee plantings elsewhere (Bartle *et al.*, 2012; Huxtable *et al.*, 2012). Growth of the external rows is enhanced by the additional resources (water and nutrients) available from the annual crop or pasture outside the belt. There were also gains in productivity from shorter within-row spacing. This has been found in other eucalypt spacing experiments where higher productivity is found at plantings with greater density before canopy closure (Pinkard & Neilsen, 2003; West & Smith, 2019).

At both sites, there were significant effects of both row number and within-row spacing on tree size. Larger trees were found in narrower belts and with wider within-row spacing, due to the reduced competition from the lower planting density. Large variation in tree size between spacing treatments will have an effect on harvestability.

### *Mallee economics*

Over the period 1994 to 2014 some 1,000 WA landholders planted mallee on their properties in anticipation of landcare benefits and to attract commercial processing interests (URS, 2008; Bartle & Abadi, 2010). No large-scale processor interest has yet emerged but potential for commercial operations remains (Abadi *et al.*, 2012; McGrath *et al.*, 2016). Barriers to commercialisation include the unknown price of biomass and reliable estimates of the production potential of mallee. To address this, experimental mallee and agricultural production were economically modelled at 11 sites over 6-years using various harvest regimes. Costs of mallee were separated into direct and indirect costs. Cash outlays for establishment and maintenance accounted for 14% of the total cost to farmers and harvest accounted for 32%. Indirect non-cash costs, imposed a further 38% on total costs due to competition imposed on adjacent agricultural annual crop or pasture and 16% from the opportunity costs associated with land use. Levelised costs were determined for each site and harvest treatment. A wide range of levelised costs were estimated (AUD40 to AUD257) in the base-case harvest scenario. A lower levelised cost was realised if below-ground biomass generated carbon credits on the Australian market. If left unharvested, three of the sites were profitable with income generated from the current Australian rules for carbon offsets.

This research found that the main driver for reduced cost of mallee biomass is the productivity of the mallee planting, i.e. productive sites had lower levelised costs. The two decades of extensive mallee planting by WA farmers was strongly motivated by the opportunity for a low-cost treatment for the salinity problem, and planting was often biased to lower rainfall regions and on land adjacent to areas of shallow saline groundwater discharge. This compromises profitability. The case for up-slope belt formation plantings: where mallee yields will be greater, and economic viability enhanced, while still intercepting rainfall recharge and diminishing groundwater flow to discharge areas, should be favoured. High productivity upslope mallee sites will exhibit greater water use having a greater impact on hydrological imbalances. Furthermore, there are other economically attractive perennial plant options such as saltbush well suited to the saline discharge areas (Bennett *et al.,* 2009; Bennett *et al.,* 2014).

### Conclusion

The mallee species the subject of this thesis, grown in alley farming configuration, show potential as a short-rotation coppice crop with multiple benefits for the WA wheatbelt. In particular they may be developed into a profitable crop that could also help manage chronic problems with salinity and conservation. As set out in the thesis objectives in Chapter 1, gains in breeding for biomass can be achieved by implementing reproductive phenological assessments and also, selecting for elite biomass producers at coppice harvest age (gains of up to 15%). Gains may be realised concurrently in both 1,8-cineole and biomass yields with breeding programs. This should increase mallee productivity and commercial viability in future plantings.

Mallee biomass yields exhibit high variability across sites and to optimise production will require landscape scale design and site selection to maximise water use, and local scale management of soil attributes including $EC_e$, pH and nutrition (which can generate up to a 10-fold difference in productivity). Landscape scale design indicates planting in narrow belts (1 or 2 rows) and with short within-row spacing (1 to 1.5m) (up to 6-fold difference in productivity). The increases in productivity due to site factors and planting configuration will increase the economic viability of mallee plantings. Mallee plantings used to generate carbon credits at productive sites are likely to be economically viable at current Australian carbon prices.

### Future research

Chapter 4 outlines a methodology for assessing the outcrossing potential between families and provenances at an *E. loxophleba* seed orchard. Assessment of reproductive phenology has not been published for the other two taxa of most commercial interest, *E. polybractea* and *E. kochii*. This research would reveal any genetic correlations between key flowering traits with biomass productivity and foliar 1,8-cineole yield. This would inform breeding decisions for any future mallee breeding program.

To quantify the effect of out-of-phase flowering, it would be of interest to establish new trials using seedlots from early, peak and late flowering individuals. This would be especially pertinent for the *E. loxophleba* seed orchards where trees that flower twice in a year could be identified. Although 16% of the trees flowered twice, this represents less than 4% of the total reproductive output from the orchard. The first flowering event from the trees that flowered twice was not aligned with the orchard's peak flowering and present a high probability of producing inbred or self-fertilised seed. Comparing the growth rates from offspring of trees that flowered out-of-phase to those flowering at the peak of flowering would further inform breeding decisions on how aggressively to select against trees that flower out-of-phase or twice per year.

Chapter 6 demonstrated that larger gains of coppice biomass were obtained when breeding selections were applied to coppice. However, there was not a consistent site response after harvest. There was a reduction of additive variance at Condobolin, a rise at Drummartin, yet Newdegate was stable. A further harvest rotation and biomass assessment of coppice at harvest age at the three progeny trials may clarify this variation. The site response to harvest appears most likely to be caused by the genetic control of lignotubers or an interaction between coppice performance and the conditions at the three specific sites. Further, the heritability of coppice should be estimated for both *E. loxophleba* subsp. *gratiae* and *lissophloia.* There are three progeny trials established for each of these and which were harvested at the same time at the *E. polybractea* progeny trials. This work was planned to be included in this thesis, but due to COVID-19 travel restrictions, assessment of the coppice was not possible. The results from these experiments would provide more conclusive evidence regarding the genetic control and growth rates of coppiced mallee eucalypts.

For the mallee biomass estimates in Chapter 2, nitrogen was a predictor of mallee growth rates under harvest regimes. Research has found high nitrogen removal with the export of biomass at harvest. It is likely that nutrient replacement to mallee after harvest will be required to sustain economically viable growth rates. The amount of nutrient required is yet to be defined, but particular examination of the impact of the extensive lateral extension of the mallee root systems in belts configuration will be required. A significant increase in growth rates may be important for mallee commercialisation and increasing income to growers. Further, if nitrogen was constraining productivity at low productive sites, without other constraining soil factors, additions of nitrogen might increase the area that mallee could be economically viable. This may reduce the competition for land use between tree crops and conventional annual agriculture.

Chapter 3 demonstrated that there was a significant productivity response to mallee planting configurations with the internal rows from wider belts undergoing significant growth suppression due to competition from the external rows. This work was completed using saplings and the response to harvest is unknown. Harvesting may exacerbate or alleviate suppression of internal rows. These spacing trials should be harvested and biomass should be reassessed at coppice harvest age.

The mallee economic chapter used below-ground biomass estimates to quantify the carbon that was sequestered. Currently, legislation in Australia under the farm forestry methodology states that under a coppice system, below ground biomass cannot exceed the estimation at first harvest. Currently there is no research estimating below ground biomass of coppicing eucalypts over multiple harvest cycles. Two aspects need to be clarified: 1) allometric modelling of below-ground biomass over multiple harvests cycles; and 2) the quantity of total soil organic carbon of both coppice and unharvested system, e.g. how long is the additional carbon stored in the soil from senesced mallee roots after harvest. This research could be used to update the below-ground carbon estimations from the farm forestry methodology and guide government legislation relating to carbon credits.

Lastly, for a prospective mallee industry, multi-criteria mapping needs to be undertaken to determine the geographical extent of high mallee productivity, especially for extensive belt configurations. There is substantial knowledge of where mallee taxa are suited climatically, but lateral surface and subsurface water flows, and edaphic factors will be also be strong determinants of productivity. Such regional scale investigation should also identify locations at high risk of salinisation and suitable infrastructure for large scale mallee biomass processing industry.

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# Appendices

# Appendix A:

## Supplementary material- Chapter 2

# Supplementary Material for *A decadal multi-site study of the effects of frequency and season of harvest on biomass production from mallee eucalypts*

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**Table S1**- Spatial definition of experiment site hillslope sections: from local catchment divide to stream line or valley floor (m). Landscape elevation data was obtained using Google Maps (Google Earth, 2018). Landform classifications have been adapted from Commander (2001).

| **Site** | **Geological province and landform** | **Site and soil description** | **Elevation (m above sea level)** | | |
|---|---|---|---|---|---|
| | | | **Valley floor** | **Experiment site** | **Δ exp site and valley floor** |
| Site 1 | Yilgarn craton, zone of rejuvenated drainage, mid-slope. | Mid-slope between laterite upland to non-saline annual stream, sandy clay gravel soils | 347 | 359 - 364 | 12 - 17 |
| Site 2 | Yilgarn craton, zone of ancient drainage, lower-slope. | Lower slope to broad flat with pale loamy sand over clay. No surface expression of salt. | 301 | 308 | 7 |
| Site 3 | Yilgarn craton, zone of ancient drainage, mid-slope. | Mid slope, pale loamy sand over clay, no surface expression of salt. | 319 | 324 | 5 |
| Site 5 | Yilgarn craton, zone of ancient drainage, sandplain | Foot of sandy soil slope above a salt seepage area that forms the head of tributary saline valley. | 320 | 321 - 327 | 1 - 7 |
| Site 6 | Yilgarn craton, zone of ancient drainage, valley floor | Broad valley floor, red loam soils south of Lake Moore, hollows on site show incipient salt expression | 312 | 314 - 316 | 2 - 4 |
| Site 7 | Yilgarn craton, zone of ancient drainage, valley floor | Narrow valley floor with shallow saline groundwater discharge | 318 | 318 | 0 |
| Site 8 | Yilgarn craton, zone of ancient drainage, lower-slope. | Lower slope to broad flat with pale loamy sand over clay. No stream line or surface expression of salt. | 305 | 325 | 20 |
| Site 9 | Yilgarn craton, zone of rejuvenated drainage, valley floor. | Adjacent to valley floor, little fall from exp site to saline discharge, sandy loam over clay soil. | 238 | 240 - 243 | 2 - 5 |
| Site 10 | Yilgarn craton, zone of ancient drainage, valley floor. | Broad valley floor with shallow saline groundwater, pale sandy clay soils | 298 | 300 | 2 |
| Site 11 | Yilgarn craton, zone of ancient drainage, elevated valley | Sandplain grading to red loam soil, no surface expression of salt | 298 | 333 | 35 |
| Site 12 | Yilgarn craton, zone of ancient drainage, valley floor. | Adjacent to narrow saline valley with 1:5000 gradient, pale sandy clay soils. | 264 | 267 | 3 |
| Site 13 | Yilgarn craton, zone of ancient drainage, valley floor. | Broad flat valley floor 1:2000 gradient, incipient surface salt expression adjacent to site red loam clay soils | 270 | 279 | 9 |
| Site 14 | Yilgarn craton, zone of ancient drainage, valley floor. | Adjacent to valley floor, 3m fall from plot to discharge zone, sandy surface soil with hard pan | 288 | 291 | 3 |
| Site 15 | Yilgarn craton, zone of ancient drainage, valley floor | Downslope from Site 16 on broad valley floor red loam soils west of Lake Moore | 311 | 317 - 320 | 6 - 9 |
| Site 16 | Yilgarn craton, zone of ancient drainage, sandplain | Low on a long sandy soil slope adjacent to saline sandplain seep abutting red loam slopes | 311 | 328 - 334 | 17 - 23 |
| Site 17 | Yilgarn craton, zone of ancient drainage, narrow valley floor | Valley floor at head of drainage line, plot at foot of hillslope, no surface salt expression, red loam soils | 315 | 324 | 9 |
| Site 18 | Esperance sandplain, undulating Tertiary sediments overlying igneous basement, open to external drainage. | Elevated deep grey/yellow sandy duplex soil | 132 | 139 | 7 |
| Site 19 | | Elevated deep grey/yellow sandy duplex soil | 128 | 140 | 12 |
| Site 20 | Esperance sandplain, Tertiary sediments overlying igneous basement, no external drainage. | Elevated deep grey/yellow sandy duplex soil | 140 | 161 | 21 |

**Table S2** - Site soils physico-chemical characteristics for the 19 mallee Eucalypt sites in the Western Australian Wheatbelt. This data was originally collected by R. Sudmeyer for Peck *et al.* (2012) and later used in Sudmeyer *et al.* (2012).

| Site | Rep | Spp | Max_pH | Min_pH | Max_EC | P_Colw | Organic_C | NH4_NO3 | K_Colw | Sulphur | m_clay | SoilDth |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 1 | 1 | Epoly | 6.3 | 5.0 | 6.83 | 36.8 | 2686.4 | 44.9 | 1286.8 | 1261.2 | 0.6 | 11.5 |
|  | 2 | Epoly | 6.4 | 4.9 | 9.37 | 28.2 | 1993.4 | 35.3 | 627.5 | 641.1 | 0.4 | 7.5 |
|  | 3 | Epoly | 5.8 | 5.1 | 1.21 | 22.1 | 1667.8 | 27.0 | 310.5 | 440.5 | 0.5 | 5.5 |
| 2 | 1 | Elox | 9.1 | 5.4 | 4.50 | 17.8 | 884.5 | 26.1 | 477.6 | 95.4 | 0.3 | 3.5 |
|  | 2 | Elox | 8.2 | 8.2 | 2.31 | 18.3 | 888.9 | 26.0 | 474.8 | 93.7 | 0.35 | 3.5 |
|  | 3 | Elox | 7.9 | 4.9 | 2.92 | 18.8 | 893.3 | 25.8 | 471.9 | 92.0 | 0.4 | 3.5 |
| 3 | 1 | Elox | 7.6 | 5.1 | 10.79 | 18.7 | 1454.4 | 21.6 | 1073.4 | 342.8 | 0.1 | 7.5 |
|  | 2 | Elox | 7.7 | 5.5 | 8.76 | 21.1 | 1600.2 | 24.1 | 1212.0 | 483.7 | 0.06 | 8 |
|  | 3 | Elox | 7.4 | 5.9 | 27.60 | 19.4 | 1498.1 | 22.4 | 1115.0 | 385.1 | 0.1 | 8 |
| 5 | 1 | Elox | 6.7 | 4.8 | 0.72 | 15.2 | 644.2 | 22.8 | 159.6 | 106.3 | 1.75 | 2.45 |
|  | 2 | Elox | 6.3 | 5.0 | 5.27 | 17.8 | 771.3 | 28.3 | 335.8 | 120.4 | 1.9 | 3.5 |
|  | 3 | Elox | 6.6 | 4.6 | 15.12 | 15.6 | 659.8 | 24.2 | 158.1 | 116.8 | 1.95 | 2.4 |
| 6 | 1 | Elox | 7.0 | 3.8 | 13.12 | 26.8 | 1100.5 | 17.6 | 3372.8 | 410.3 | 0.2 | 3.2 |
|  | 2 | Elox | 6.2 | 3.8 | 14.26 | 25.9 | 1019.3 | 16.3 | 3101.0 | 316.6 | 0.2 | 2.9 |
|  | 3 | Elox | 6.1 | 3.7 | 15.17 | 26.2 | 1046.4 | 16.7 | 3191.6 | 347.8 | 0.2 | 3 |
| 7 | 1 | Elox | 6.5 | 4.0 | 19.20 | 8.5 | 381.0 | 6.4 | 211.6 | 79.5 | 0.12 | 2.1 |
|  | 2 | Elox | 6.6 | 3.9 | 19.96 | 11.2 | 592.0 | 9.0 | 488.6 | 266.9 | 0.12 | 2.35 |
|  | 3 | Elox | 6.4 | 3.9 | 21.10 | 13.8 | 807.7 | 12.3 | 733.4 | 454.7 | 0.12 | 2.35 |
| 8 | 1 | Epoly | 7.9 | 7.3 | 3.51 | 35.9 | 1771.0 | 43.9 | 1359.9 | 981.7 | 0.2 | 5 |
|  | 2 | Epoly | 7.4 | 5.6 | 18.93 | 33.4 | 1644.2 | 42.9 | 1203.9 | 833.4 | 0.2 | 11.5 |
|  | 3 | Epoly | 8.5 | 6.3 | 11.58 | 35.9 | 1771.0 | 39.5 | 1359.9 | 981.7 | 0.2 | 11.5 |
| 9 | 1 | Elox | 7.7 | 4.5 | 10.15 | 11.6 | 581.1 | 17.8 | 324.9 | 154.4 | 0.3 | 2.6 |
|  | 2 | Elox | 7.3 | 4.5 | 10.85 | 10.8 | 837.8 | 15.4 | 204.0 | 108.1 | 0.5 | 2.75 |
|  | 3 | Elox | 8.0 | 4.5 | 10.18 | 8.7 | 295.4 | 13.5 | 274.8 | 56.4 | 0.35 | 5.2 |
| 10 | 1 | Elox | 7.6 | 4.1 | 6.01 | 18.5 | 1017.6 | 18.2 | 563.2 | 590.1 | 0.3 | 3.5 |
|  | 2 | Elox | 7.3 | 4.5 | 8.34 | 15.3 | 841.7 | 15.9 | 544.2 | 545.3 | 0.1 | 3.2 |
|  | 3 | Elox | 7.3 | 4.3 | 8.12 | 17.0 | 933.8 | 16.7 | 528.5 | 538.0 | 0.25 | 3.2 |
| 11 | 1 | Ekoch | 6.8 | 4.2 | 36.60 | 18.7 | 1095.9 | 21.6 | 1014.6 | 260.9 | 0.15 | 3.8 |
|  | 2 | Ekoch | 8.2 | 4.3 | 2.99 | 18.4 | 1077.1 | 21.1 | 991.8 | 249.0 | 0.3 | 3.7 |
|  | 3 | Ekoch | 7.9 | 4.0 | 36.60 | 19.0 | 1114.7 | 22.1 | 1037.4 | 272.7 | 0.5 | 3.9 |
| 12 | 1 | Elox | 7.9 | 4.0 | 18.90 | 12.8 | 865.1 | 20.4 | 663.4 | 436.7 | 0.1 | 3 |
|  | 2 | Elox | 8.2 | 4.0 | 17.17 | 12.3 | 685.7 | 9.6 | 488.8 | 165.3 | 0.1 | 1.95 |
|  | 3 | Elox | 8.1 | 4.0 | 15.90 | 13.5 | 641.1 | 12.9 | 482.5 | 259.3 | 0.1 | 2.55 |

| Site | Rep | Spp | Max_pH | Min_pH | Max_EC | P_Colw | Organic_C | NH4_NO3 | K_Colw | Sulphur | m_clay | SoilDth |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 13 | 1 | Elox | 8.1 | 3.8 | 8.36 | 28.1 | 1721.3 | 56.3 | 3847.6 | 1007.0 | 0.25 | 5.5 |
|  | 2 | Elox | 8.2 | 3.7 | 8.41 | 26.5 | 1614.0 | 53.8 | 3815.4 | 947.8 | 0.2 | 5 |
|  | 3 | Elox | 8.0 | 3.6 | 12.87 | 26.8 | 1665.2 | 54.5 | 3789.7 | 948.1 | 0.25 | 5 |
| 14 | 1 | Elox | 7.4 | 5.4 | 2.03 | 10.9 | 498.6 | 9.0 | 678.9 | 55.2 | 0.1 | 2.6 |
|  | 2 | Elox | 7.1 | 5.2 | 3.53 | 18.5 | 620.2 | 11.6 | 982.9 | 66.7 | 0.15 | 3.7 |
|  | 3 | Elox | 6.5 | 5.0 | 3.04 | 15.1 | 534.6 | 10.2 | 934.2 | 61.4 | 0.1 | 3.4 |
| 15 | 1 | Ekoch | 8.1 | 5.0 | 13.03 | 18.5 | 794.3 | 91.9 | 4272.4 | 758.3 | 0.1 | 3.1 |
|  | 2 | Ekoch | 7.4 | 4.5 | 13.89 | 18.8 | 815.5 | 93.5 | 4354.5 | 789.5 | 0.1 | 3.2 |
|  | 3 | Ekoch | 7.9 | 4.2 | 12.84 | 19.6 | 885.3 | 96.5 | 4544.6 | 870.2 | 0.1 | 3.5 |
| 16 | 1 | Ekoch | 5.8 | 4.3 | 0.31 | 11.7 | 698.9 | 26.3 | 273.0 | 240.5 | 10 | 4.7 |
|  | 2 | Ekoch | 6.3 | 4.4 | 1.03 | 12.4 | 736.5 | 27.7 | 324.7 | 243.6 | 10 | 5 |
|  | 3 | Ekoch | 6.1 | 4.1 | 0.28 | 11.3 | 673.8 | 25.4 | 238.6 | 238.4 | 10 | 4.5 |
| 17 | 1 | Elox | 6.1 | 4.1 | 8.51 | 9.7 | 718.2 | 9.7 | 593.7 | 120.6 | 0.1 | 3.2 |
|  | 2 | Elox | 6.2 | 3.9 | 17.81 | 17.3 | 1119.0 | 19.0 | 959.2 | 279.9 | 0.18 | 3.3 |
|  | 3 | Elox | 7.0 | 3.9 | 18.27 | 18.0 | 1092.8 | 18.9 | 819.0 | 218.9 | 0.25 | 3.3 |
| 18 | 1 | Epoly | 6.6 | 4.4 | 2.25 | 22.2 | 859.8 | 29.7 | 1008.8 | 502.1 | 0.3 | 8 |
|  | 2 | Epoly | 6.6 | 3.9 | 3.26 | 27.6 | 1225.3 | 24.5 | 1769.1 | 632.0 | 0.4 | 7.2 |
|  | 3 | Epoly | 6.2 | 4.3 | 1.92 | 17.1 | 736.8 | 36.0 | 996.8 | 253.7 | 0.45 | 7.7 |
| 19 | 1 | Epoly | 8.2 | 4.4 | 8.15 | 17.0 | 1237.7 | 24.0 | 697.1 | 65.7 | 0.1 | 3.8 |
|  | 2 | Epoly | 6.4 | 4.6 | 1.60 | 16.9 | 994.8 | 19.5 | 653.1 | 291.1 | 0.3 | 4 |
|  | 3 | Epoly | 7.6 | 4.5 | 7.60 | 16.7 | 819.6 | 26.3 | 592.7 | 347.4 | 0.2 | 5 |
| 20 | 1 | Elox | 8.4 | 3.8 | 10.58 | 28.2 | 1561.3 | 34.9 | 2843.8 | 846.0 | 0.1 | 10 |
|  | 2 | Elox | 8.3 | 4.3 | 10.08 | 22.5 | 1288.7 | 25.1 | 2891.5 | 882.3 | 0.35 | 5.8 |
|  | 3 | Elox | 9.0 | 4.0 | 10.43 | 26.5 | 1239.6 | 30.0 | 2376.7 | 1907.7 | 0.08 | 6.1 |

**Table S3** – The measured physico-chemical and landscape characterises used in multiple stepwise regressions.

| **Site characteristics** | **Units** |
|---|---|
| Mean Annual Precipitation | (mm) |
| mean annual pan evaporation | (mm) |
| Minimum Temperature | $^{o}$C |
| Maximum Temperature | $^{o}$C |
| Maximum pH | $CaCl_2$ |
| Minimum pH | $CaCl_2$ |
| Maximum ECe | ds/m |
| Cowell Phosphorus | mg/kg |
| NO3- plus NH4 | mg/kg |
| Cowell Potassium | mg/kg |
| Sulphur | mg/kg |
| Organic Carbon | % weight |
| Depth to clay layer | m |
| Soil Depth to hardpan or watertable | m |
| Relief to valley floor -elevation difference to nearest area of local saline discharge | m |

**Table S4** - Parameterisation of allometric model for site-specific prediction of above ground fresh biomass (AGFB) of tree (not coppiced) mallees from ‘equivalent diameter near root collar’ (EDRC). Sample number (n), coefficient of determination (R2), Mean Squared Error (MSE), and values in parentheses are standard errors of parameter estimates.

| Site | Spp | n | Lower domain (kg) | Upper domain (kg) | Intercept | Slope | $R^2$ | MSE |
|---|---|---|---|---|---|---|---|---|
| 1 | Epol | 45 | 11.4 | 771.5 | -1.400 (0.335) | 2.273 (0.114) | 0.902 | 0.045 |
| 2 | Elox | 55 | 1.7 | 98.8 | -1.433 (0.275) | 2.189 (0.115) | 0.873 | 0.030 |
| 3 | Elox | 50 | 0.8 | 111.5 | -1.094 (0.249) | 2.154 (0.114) | 0.881 | 0.037 |
| 6 | Elox | 52 | 2.6 | 90.0 | -2.368 (0.252) | 2.737 (0.116) | 0.918 | 0.048 |
| 7 | Elox | 20 | 2.4 | 53.5 | -1.259 (0.252) | 2.190 (0.122) | 0.947 | 0.009 |
| 8 | Epol | 27 | 24.7 | 110.2 | -1.204 (0.499) | 2.149 (0.202) | 0.820 | 0.040 |
| 9 | Elox | 36 | 8.2 | 164.5 | -1.671 (0.274) | 2.332 (0.114) | 0.924 | 0.040 |
| 10 | Elox | 90 | 2.0 | 157.5 | -1.679 (0.129) | 2.327 (0.059) | 0.946 | 0.031 |
| 11* | Ekoc | 45 | 1.8 | 96.2 | 1.044 (0.087) | 0.924 (0.039) | 0.928 | 0.038 |
| 12 | Elox | 101 | 1.0 | 113.1 | -2.364 (0.162) | 2.612 (0.083) | 0.909 | 0.066 |
| 13 | Elox | 80 | 4.4 | 213.5 | -1.973 (0.311) | 2.541 (0.131) | 0.828 | 0.093 |
| 14 | Elox | 47 | 1.1 | 60.6 | -2.492 (0.189) | 2.740 (0.097) | 0.947 | 0.043 |
| 15 | Ekoc | 50 | 6.4 | 143.5 | -1.658 (0.321) | 2.181 (0.121) | 0.872 | 0.052 |
| 16 | Ekoc | 122 | 0.8 | 331.5 | -2.697 (0.153) | 2.639 (0.06) | 0.942 | 0.055 |
| 17 | Elox | 70 | 0.8 | 176.0 | -2.106 (0.103) | 2.446 (0.045) | 0.977 | 0.032 |
| 18&19 | Epol | 39 | 4.5 | 184.5 | -1.967 (0.257) | 2.415 (0.103) | 0.937 | 0.058 |
| 20 | Elox | 75 | 0.5 | 96.7 | -1.815 (0.165) | 2.315 (0.088) | 0.904 | 0.079 |

* Equation used CVI not EDRC

**Table S5** - Parameterisation of allometric model for site-specific prediction of above ground fresh biomass (AGFB) of coppiced mallees from ‘equivalent diameter near root collar’ (EDRC). Sample number (n), coefficient of determination (R2), Mean Squared Error (MSE), and values in parentheses are standard errors of parameter estimates.

| Site | Spp | n | Lower domain (kg) | Upper domain (kg) | Intercept | Slope | $R^2$ | MSE |
|---|---|---|---|---|---|---|---|---|
| 1 | $E_{pol}$ | 142 | 2.1 | 316.0 | 0.285 (0.133) | 1.029 (0.035) | 0.859 | 0.075 |
| 2 | $E_{lox}$ | 45 | 3.6 | 74.5 | 0.173 (0.173) | 0.984 (0.048) | 0.903 | 0.023 |
| 3 | $E_{lox}$ | 142 | 0.4 | 53.0 | 0.016 (0.093) | 1.033 (0.033) | 0.878 | 0.058 |
| 6 | $E_{lox}$ | 60 | 0.9 | 43.5 | -0.589 (0.136) | 1.215 (0.05) | 0.912 | 0.055 |
| 8 | $E_{pol}$ | 120 | 3.0 | 104.0 | 0.226 (0.105) | 1.068 (0.036) | 0.882 | 0.045 |
| 9 | $E_{lox}$ | 113 | 0.3 | 69.0 | -0.145 (0.129) | 1.085 (0.043) | 0.849 | 0.044 |
| 10 | $E_{lox}$ | 81 | 0.1 | 35.9 | 0.075 (0.163) | 1.015 (0.059) | 0.789 | 0.072 |
| 12 | $E_{lox}$ | 50 | 1.7 | 63.5 | -0.223 (0.159) | 1.107 (0.054) | 0.897 | 0.057 |
| 13 | $E_{lox}$ | 78 | 5.4 | 63.5 | -0.107 (0.133) | 1.159 (0.047) | 0.887 | 0.028 |
| 15 | $E_{koc}$ | 60 | 8.2 | 95.3 | 0.651 (0.145) | 0.995 (0.045) | 0.893 | 0.029 |
| 16 | $E_{koc}$ | 94 | 1.0 | 107.3 | 0.860 (0.108) | 0.934 (0.035) | 0.887 | 0.031 |
| 17 | $E_{lox}$ | 97 | 1.5 | 53.0 | -0.273 (0.119) | 1.118 (0.04) | 0.890 | 0.058 |
| 18 | $E_{pol}$ | 124 | 1.0 | 197.0 | -0.064 (0.102) | 1.14 (0.033) | 0.909 | 0.065 |
| 19 | $E_{pol}$ | 118 | 0.1 | 75.5 | 0.231 (0.103) | 1.061 (0.036) | 0.882 | 0.052 |

## Figures S1-S20: Pairwise T-tests for symmetry of outer rows in a mallee eucalypt belt

The paper, *A decadal multi-site study of the effects of frequency and season of harvest on biomass production from mallee eucalypts*, assessed three species of mallee eucalypts, hereafter referred to as ‘mallee’, planted in belts of varying numbers of rows. Of the 19 sites considered by this study there were 10 sites with 2-row belts, one site with a 3-row belt, four sites with 4-row belts and four sites with 6-row belts. At each site, the diameters of an edge row and the adjacent row were measured. Therefore, not all the stem diameters were measured at sites with more than two rows.

This paper considers the total biomass of the mallee belt (measured and unmeasured trees) in which the unmeasured trees were estimated using principles of symmetry. For instance, in a 3-row belt, the biomass of the measured edge row was doubled to compensate for the unmeasured edge row. The outer row was selected rather than the adjacent (internal) row to account for the biomass suppression that occurs in internal rows (Peck *et al.*, 2012). Likewise, for a 4-row belt, the biomass from the two measured rows (rows one and two) were doubled to account for the biomass of the unmeasured rows (rows three and four).

The hypothesis tested below is that there will be no difference in biomass between the outer rows from each side of a mallee belt. The data presented here is from 20 sites across the Western Australian wheatbelt which included mallee of different species, ages and number of rows. Some of the sites were also presented in the decadal study. Some sites had been harvested while others were uncut. At each site, the stem diameter or volume were measured for a minimum of eight trees from each row and from at least three replicates. To estimate biomass allometric equations were applied from this study. The biomass of each of the groups compared in the t-tests using the sum of total green biomass. Natural logarithmic transformation was applied to these data. Where there was a three-row planting, row 1 was tested against row 3. When there were four or more rows, the sum of row one and two were tested against the alternative outer two rows.

At 18 of the 20 datasets compared, there was no significant difference between the biomass of the groups suggesting that symmetry exists within a mallee belt. At one of the sites with a significant difference between rows 1 and 2 vs rows 3 and 4 (Fig. S9), there was drain installed above the tree belt approximately 1 metre deep with a bund installed. This may have contributed to the uneven biomass distribution due to unequal water allocation.

**Figure S1** – Comparison of biomass from rows 1 and 3 in a 3-row belt at Site 3 in this publication. This is an uncut *Eucalyptus loxophleba* ssp *lissophloia* site measured at approximately 17 years old. Below is the output from the pair-wise t-test. The solid lines represent the natural log–transformed biomass of each rep at row 1 and 3. The broken line represents the average biomass.

| N | Mean | Std Dev | Std Err | Minimum | Maximum |
|---|---|---|---|---|---|
| 3 | -0.0116 | 0.2148 | 0.1240 | -0.2557 | 0.1488 |

| Mean | 95% CL Mean | | Std Dev | 95% CL Std Dev | |
|---|---|---|---|---|---|
| -0.0116 | -0.5453 | 0.5220 | 0.2148 | 0.1118 | 1.3501 |

| DF | t Value | Pr > \|t\| |
|---|---|---|
| 2 | -0.09 | 0.9337 |

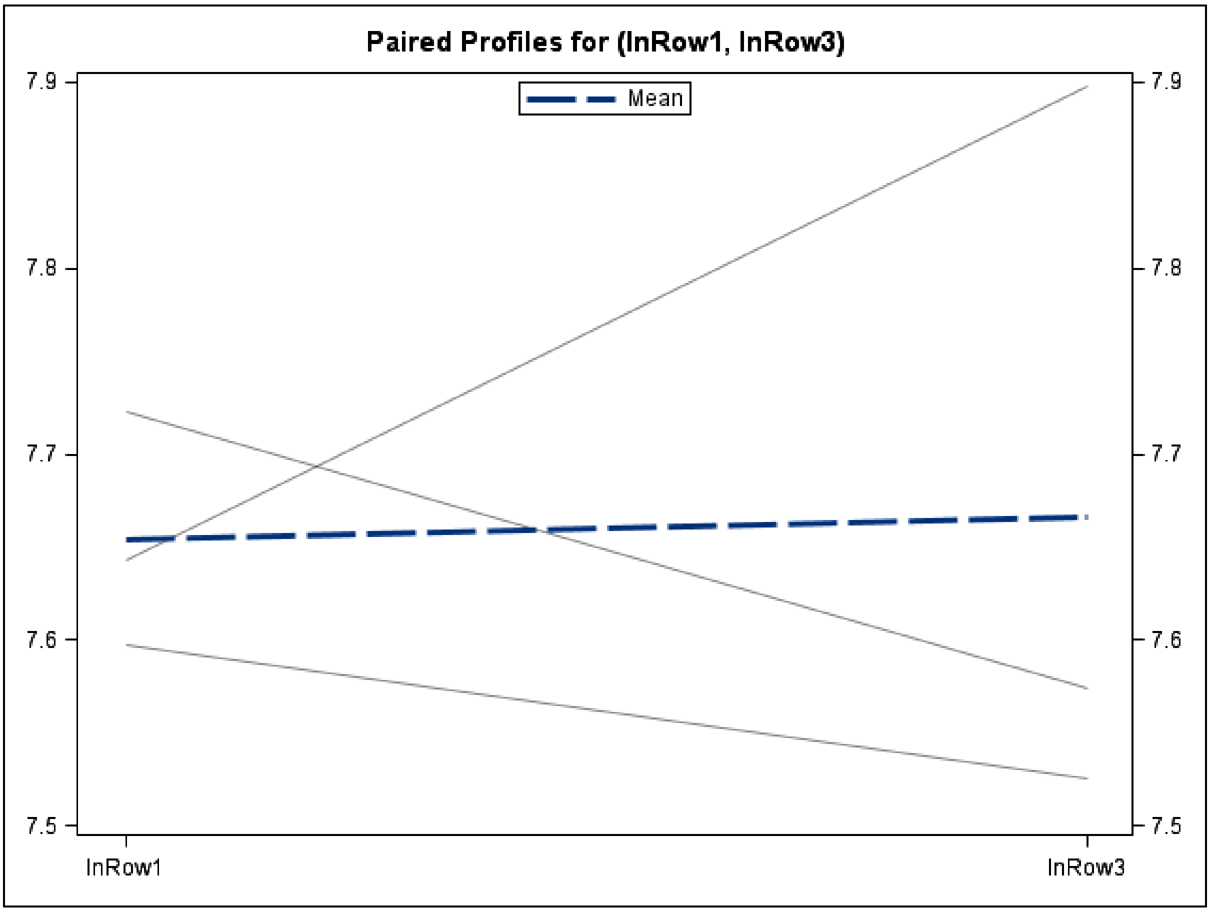

**Figure S2** – Comparison of the combined biomass of rows 1 and 2 versus rows 3 and 4 in a 4-row belt near Gibson. This is an uncut *E. polybractea* site measured at approximately 19 years old. Below is the output from the pair-wise t-test. The solid lines are the natural log–transformed biomass at row 1 and 2 compared to rows 3 and 4. The broken line represents the average biomass.

| N | Mean | Std Dev | Std Err | Minimum | Maximum |
|---|---|---|---|---|---|
| 12 | 0.1529 | 0.3105 | 0.0896 | -0.2457 | 0.7305 |

| Mean | 95% CL Mean | | Std Dev | 95% CL Std Dev | |
|---|---|---|---|---|---|
| 0.1529 | -0.0444 | 0.3501 | 0.3105 | 0.2199 | 0.5272 |

| DF | t Value | Pr > \|t\| |
|---|---|---|
| 11 | 1.71 | 0.1161 |

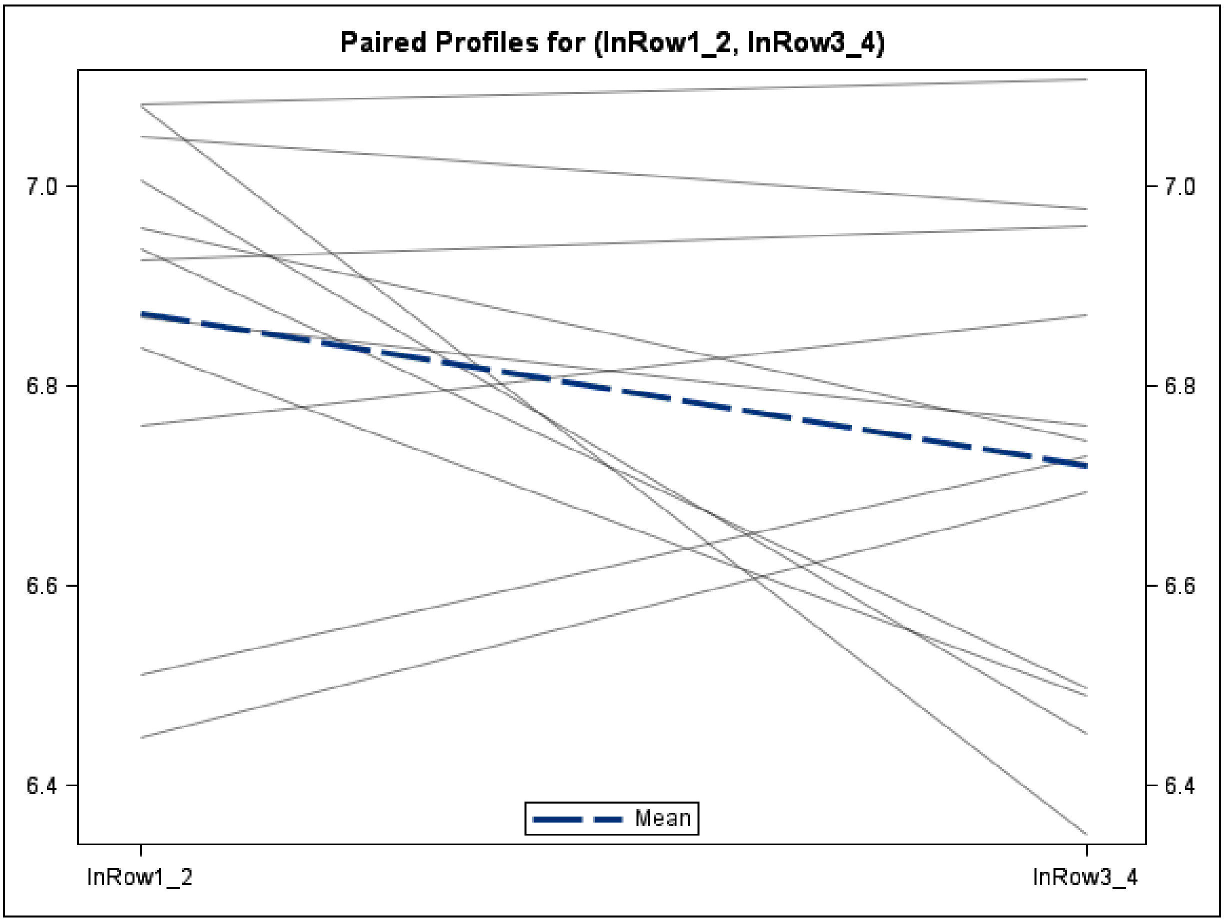

**Figure S3** – Comparison of the combined biomass of rows 1 and 2 versus rows 3 and 4 in a 4-row belt in a spacing trial located near Cuballing. This is an uncut *E. polybractea* site measured at approximately 13 years old. Below is the output from the pair-wise t-test. The solid lines are the natural log–transformed biomass at row 1 and 2 compared to rows 3 and 4. The broken line represents the average biomass.

| N | Mean | Std Dev | Std Err | Minimum | Maximum |
|---|---|---|---|---|---|
| 4 | -0.1485 | 0.4129 | 0.2065 | -0.5736 | 0.3711 |

| Mean | 95% CL Mean | | Std Dev | 95% CL Std Dev | |
|---|---|---|---|---|---|
| -0.1485 | -0.8055 | 0.5086 | 0.4129 | 0.2339 | 1.5396 |

| DF | t Value | Pr > \|t\| |
|---|---|---|
| 3 | -0.72 | 0.5240 |

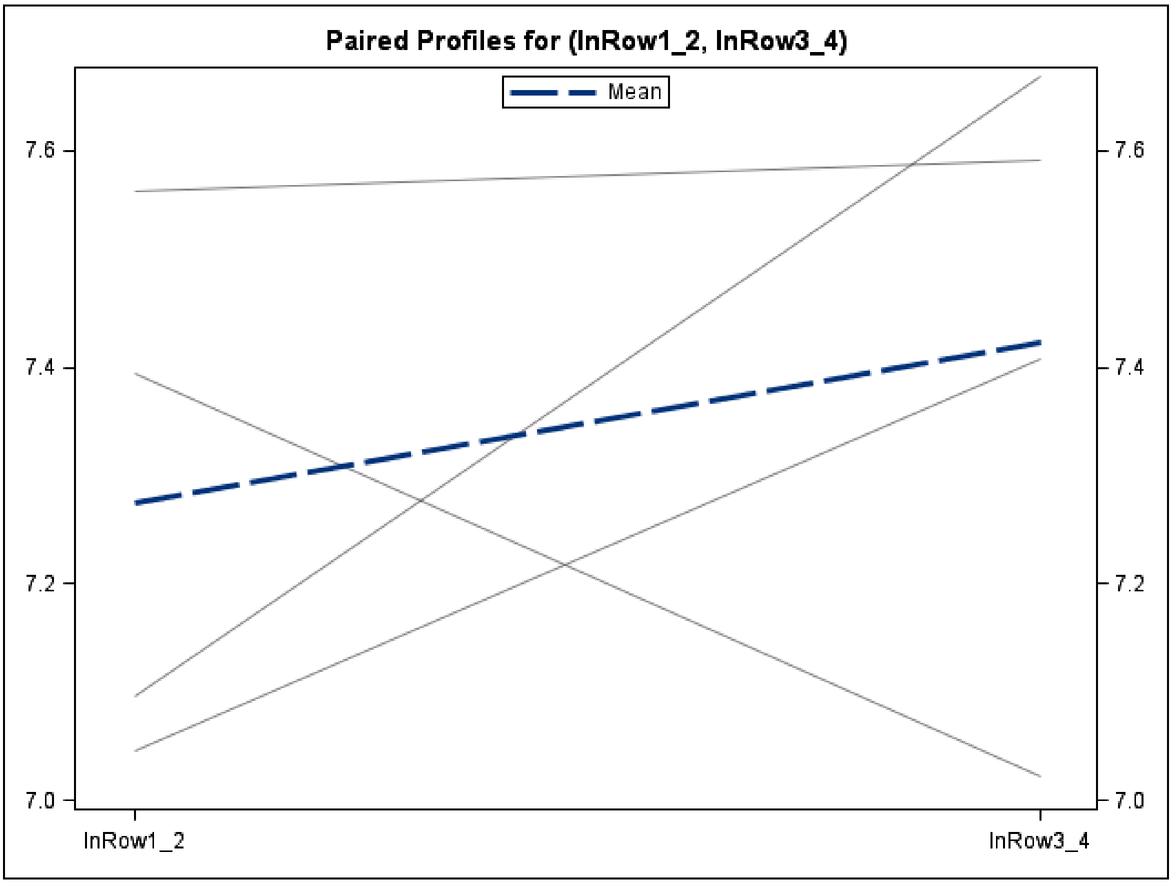

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**Figure S4** – Comparison of the combined biomass of rows 1 and 2 versus rows 3 and 4 in a 4-row belt located near Pingelly. This is an *E. loxophleba* spp *lissophloia* coppice site measured 4-years after harvest and the trees were established approximately 16 years prior to measurement. Below is the output from the pair-wise t-test. The solid lines are the natural log–transformed biomass at row 1 and 2 compared to rows 3 and 4. The broken line represents the average biomass.

| N | Mean | Std Dev | Std Err | Minimum | Maximum |
|---|---|---|---|---|---|
| 3 | 0.0760 | 0.1427 | 0.0824 | -0.0585 | 0.2256 |

| Mean | 95% CL Mean | | Std Dev | 95% CL Std Dev | |
|---|---|---|---|---|---|
| 0.0760 | -0.2785 | 0.4304 | 0.1427 | 0.0743 | 0.8967 |

| DF | t Value | Pr > \|t\| |
|---|---|---|
| 2 | 0.92 | 0.4538 |

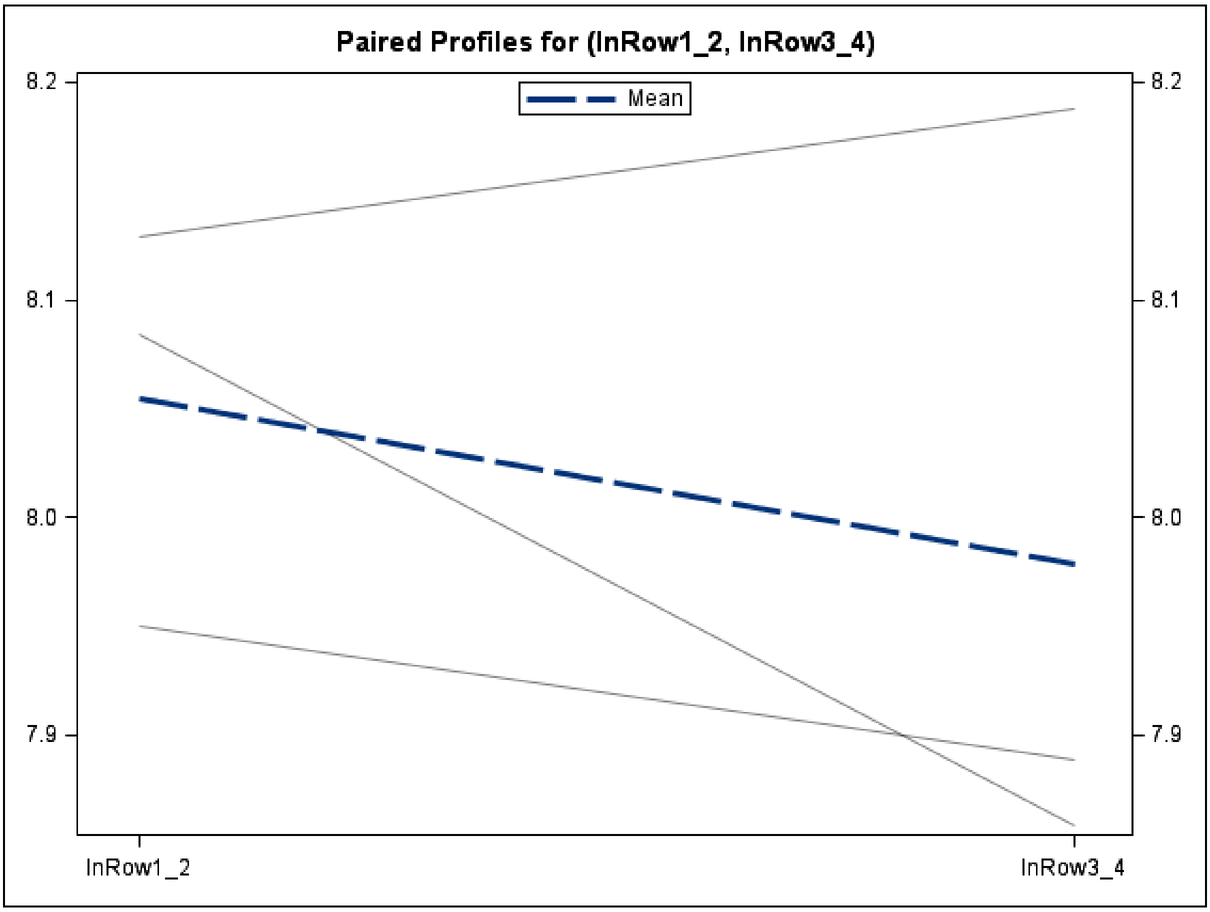
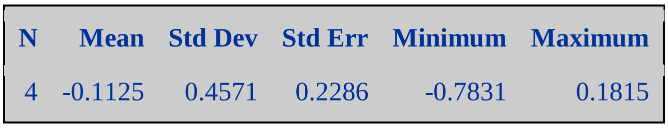
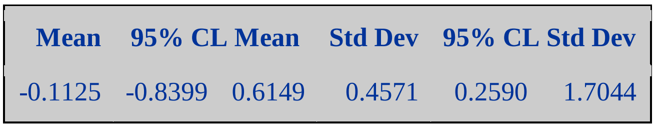


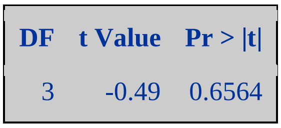
**Figure S5** – Comparison of the combined biomass of rows 1 and 2 versus rows 3 and 4 in a 4-row belt Spacing Trial located near Pingelly. This is an *E. loxophleba* spp *lissophloia* site measured when the trees were approximately 13 years old. Below is the output from the pair-wise t-test. The solid lines are the natural log–transformed biomass at row 1 and 2 compared to rows 3 and 4. The broken line represents the average biomass.



| N | Mean | Std Dev | Std Err | Minimum | Maximum |
|---|---|---|---|---|---|
| 4 | -0.1125 | 0.4571 | 0.2286 | -0.7831 | 0.1815 |



| Mean | 95% CL Mean | | Std Dev | 95% CL Std Dev | |
|---|---|---|---|---|---|
| -0.1125 | -0.8399 | 0.6149 | 0.4571 | 0.2590 | 1.7044 |



| DF | t Value | Pr > \|t\| |
|---|---|---|
| 3 | -0.49 | 0.6564 |

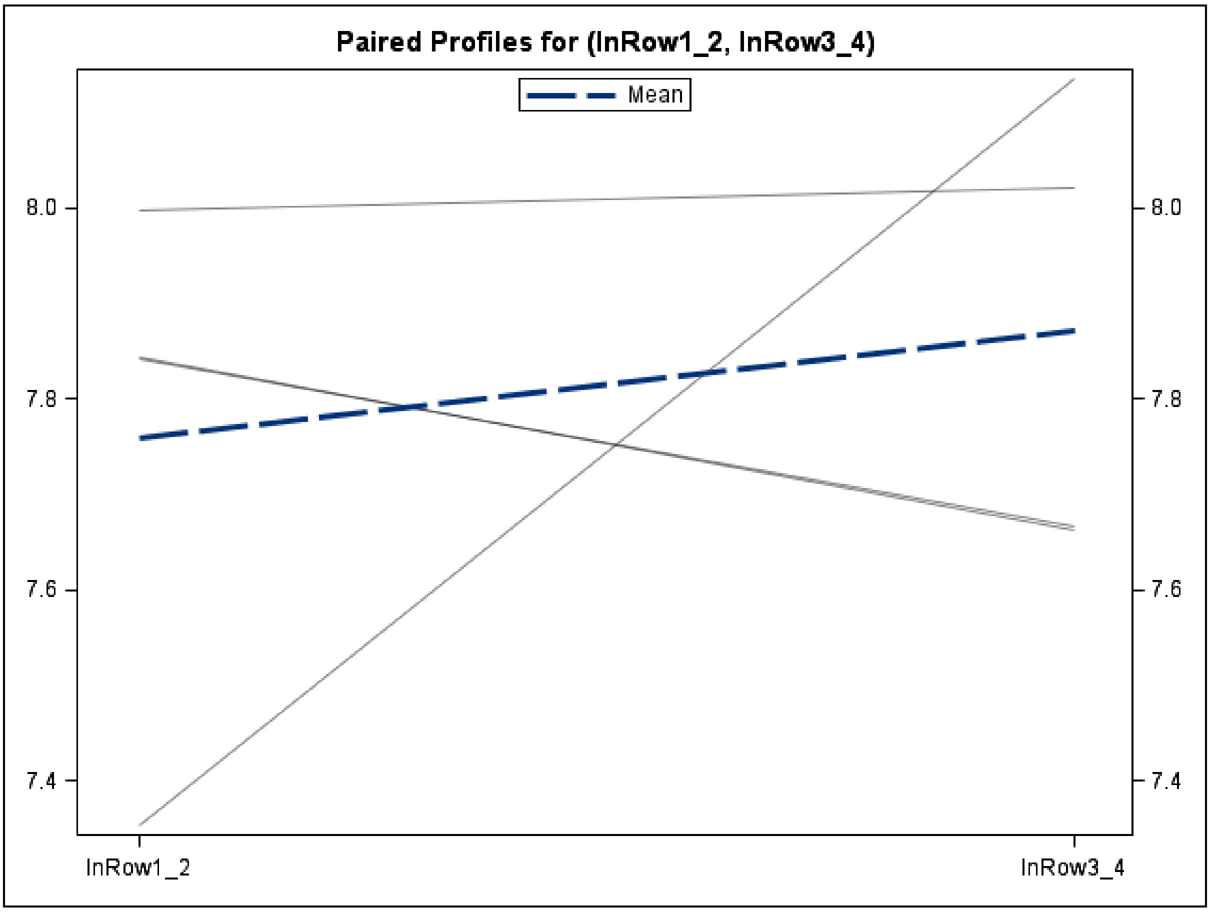


**Figure S6** – Comparison of the combined biomass of rows 1 and 2 versus rows 3 and 4 in a 4-row belt located near Pingelly. This is an *E. loxophleba* spp *lissophloia* site measured when the trees were approximately 16 years old. Below is the output from the pair-wise t-test. The solid lines are the natural log–transformed biomass at row 1 and 2 compared to rows 3 and 4. The broken line represents the average biomass.

| N | Mean | Std Dev | Std Err | Minimum | Maximum |
|---|---|---|---|---|---|
| 3 | 0.2746 | 0.2029 | 0.1171 | 0.0439 | 0.4254 |

| Mean | 95% CL Mean | | Std Dev | 95% CL Std Dev | |
|---|---|---|---|---|---|
| 0.2746 | -0.2294 | 0.7786 | 0.2029 | 0.1056 | 1.2752 |

| DF | t Value | Pr > \|t\| |
|---|---|---|
| 2 | 2.34 | 0.1438 |

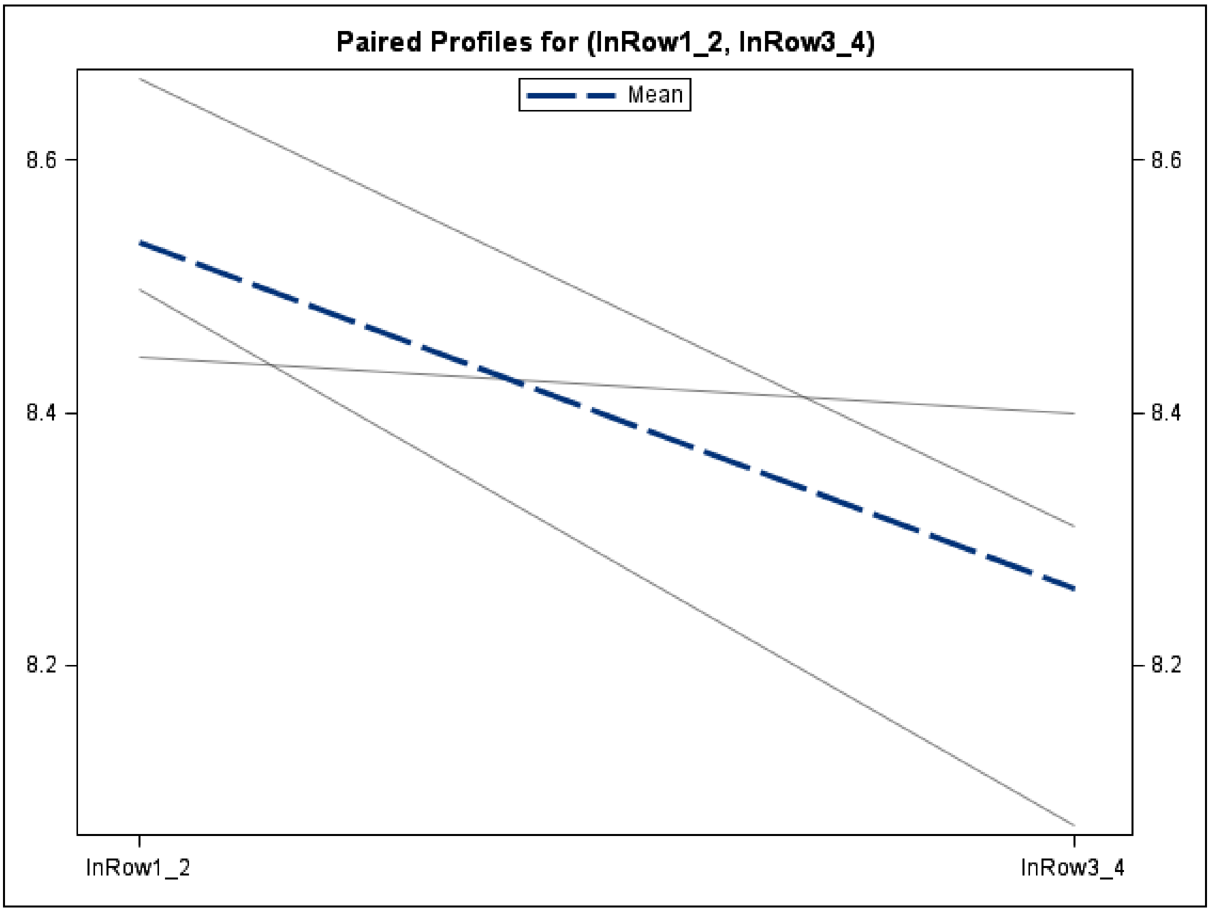


**Figure S7** – Comparison of the combined biomass of rows 1 and 2 versus rows 3 and 4 in a 4-row belt located near Pingelly. This is an *E. loxophleba* spp *lissophloia* site measured when the trees were approximately 13 years old. Below is the output from the pair-wise t-test. The solid lines are the natural log–transformed biomass at row 1 and 2 compared to rows 3 and 4. The broken line represents the average biomass.

| **N** | **Mean** | **Std Dev** | **Std Err** | **Minimum** | **Maximum** |
|---|---|---|---|---|---|
| 16 | 0.3455 | 0.2551 | 0.0638 | -0.0581 | 0.8889 |

| **Mean** | **95% CL Mean** | | **Std Dev** | **95% CL Std Dev** | |
|---|---|---|---|---|---|
| 0.3455 | 0.2096 | 0.4814 | 0.2551 | 0.1885 | 0.3948 |

| **DF** | **t Value** | **Pr > \|t\|** |
|---|---|---|
| 15 | 5.42 | <.0001 |

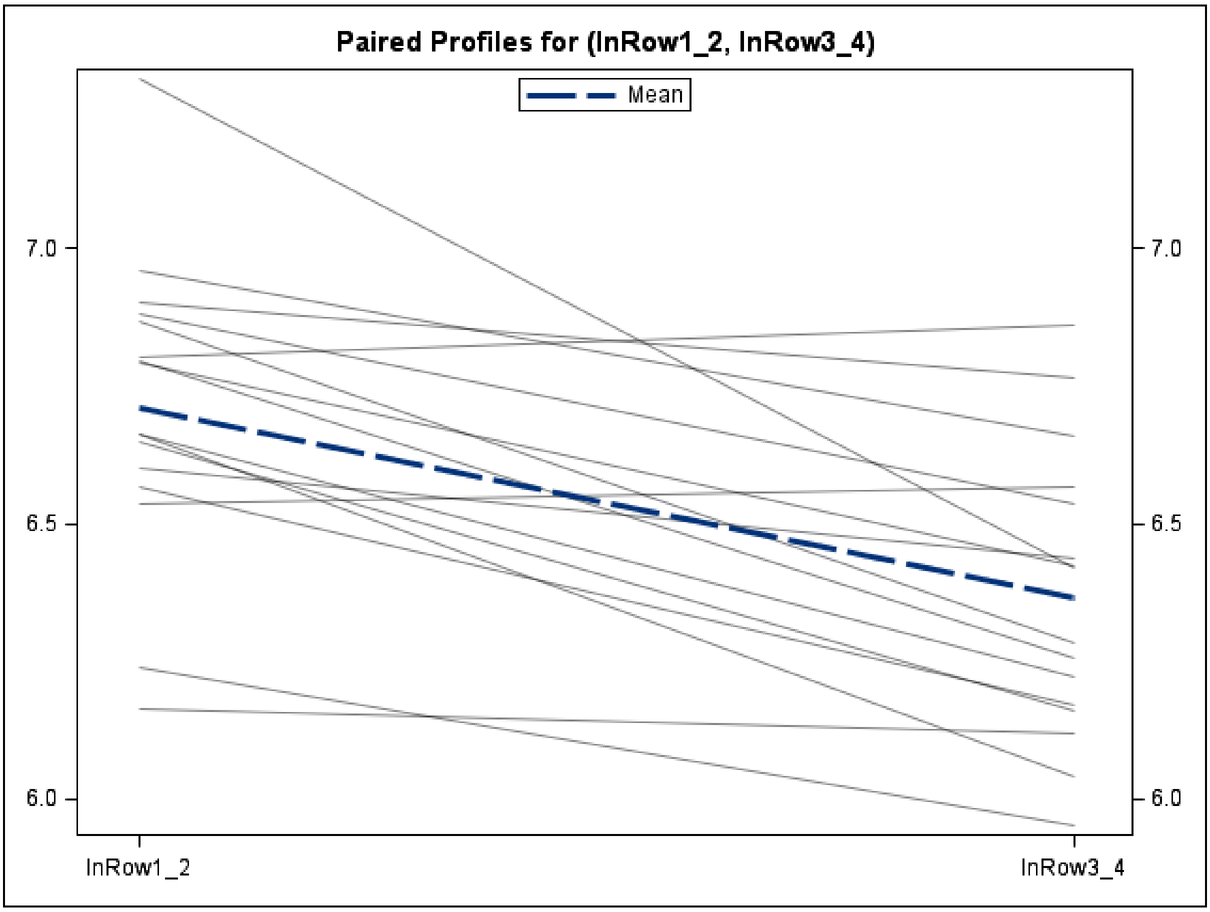


**Figure S8** – Comparison of the combined biomass of rows 1 and 2 versus rows 3 and 4 in a 4-row belt at Site 8 in this publication. This is an *E. polybractea* coppice site measured 4 years after harvest and the trees were established about 19 years prior to measurement. Below is the output from the pair-wise t-test. The solid lines are the natural log–transformed biomass at row 1 and 2 compared to rows 3 and 4. The broken line represents the average biomass.

| N | Mean | Std Dev | Std Err | Minimum | Maximum |
|---|---|---|---|---|---|
| 3 | 0.2262 | 0.1772 | 0.1023 | 0.0487 | 0.4030 |

| Mean | 95% CL Mean | | Std Dev | 95% CL Std Dev | |
|---|---|---|---|---|---|
| 0.2262 | -0.2139 | 0.6663 | 0.1772 | 0.0922 | 1.1134 |

| DF | t Value | Pr > \|t\| |
|---|---|---|
| 2 | 2.21 | 0.1575 |

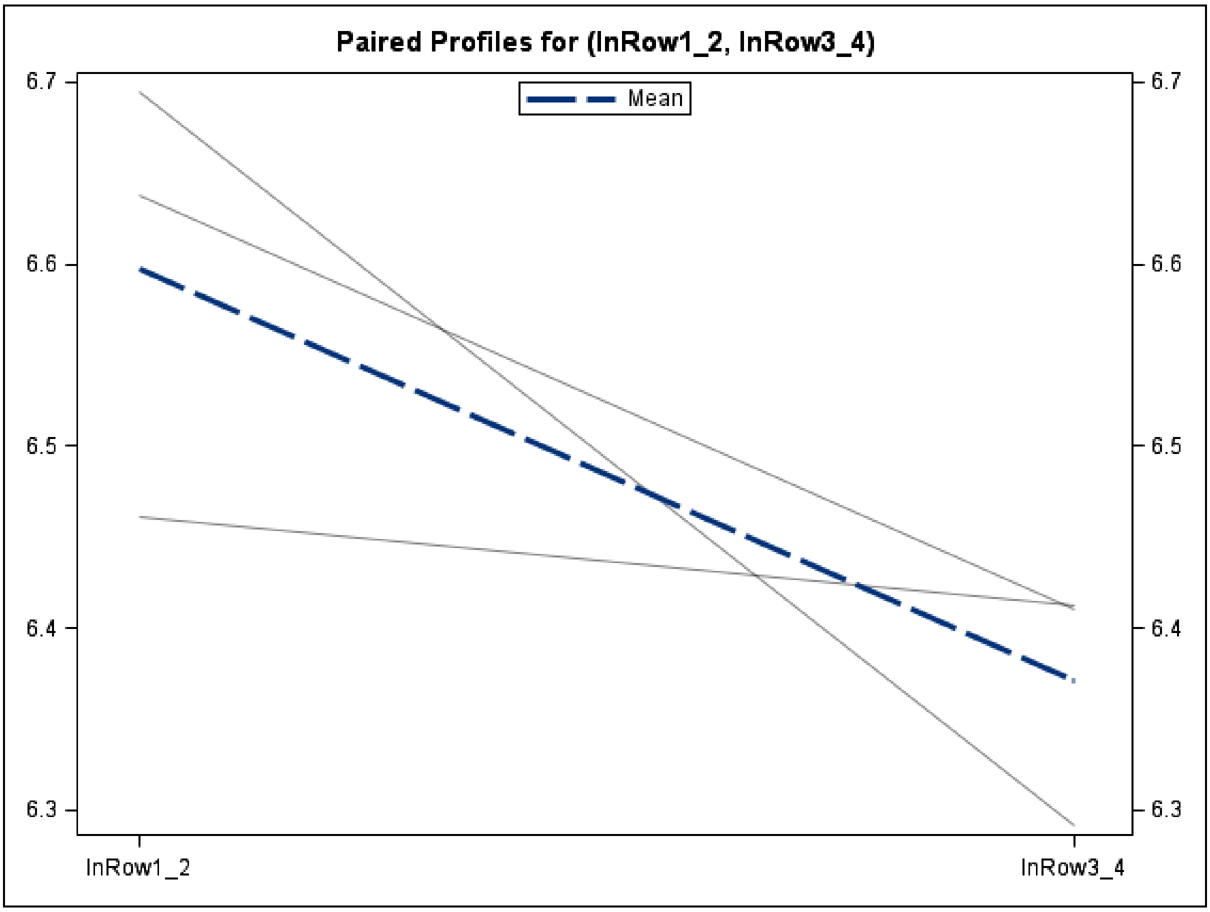


**Figure S9** – Comparison of the combined biomass of rows 1 and 2 versus rows 3 and 4 in a 4-row belt at Site 8 in this publication. This is an *E. polybractea* coppice site measured 7 years after harvest and the trees were established about 19 years prior to measurement. Below is the output from the pair-wise t-test. The solid lines are the natural log–transformed biomass at row 1 and 2 compared to rows 3 and 4. The broken line represents the average biomass.

| N | Mean | Std Dev | Std Err | Minimum | Maximum |
|---|---|---|---|---|---|
| 3 | 0.4548 | 0.0115 | 0.00663 | 0.4441 | 0.4669 |

| Mean | 95% CL Mean | | Std Dev | 95% CL Std Dev | |
|---|---|---|---|---|---|
| 0.4548 | 0.4262 | 0.4833 | 0.0115 | 0.00598 | 0.0722 |

| DF | t Value | Pr > \|t\| |
|---|---|---|
| 2 | 68.56 | 0.0002 |

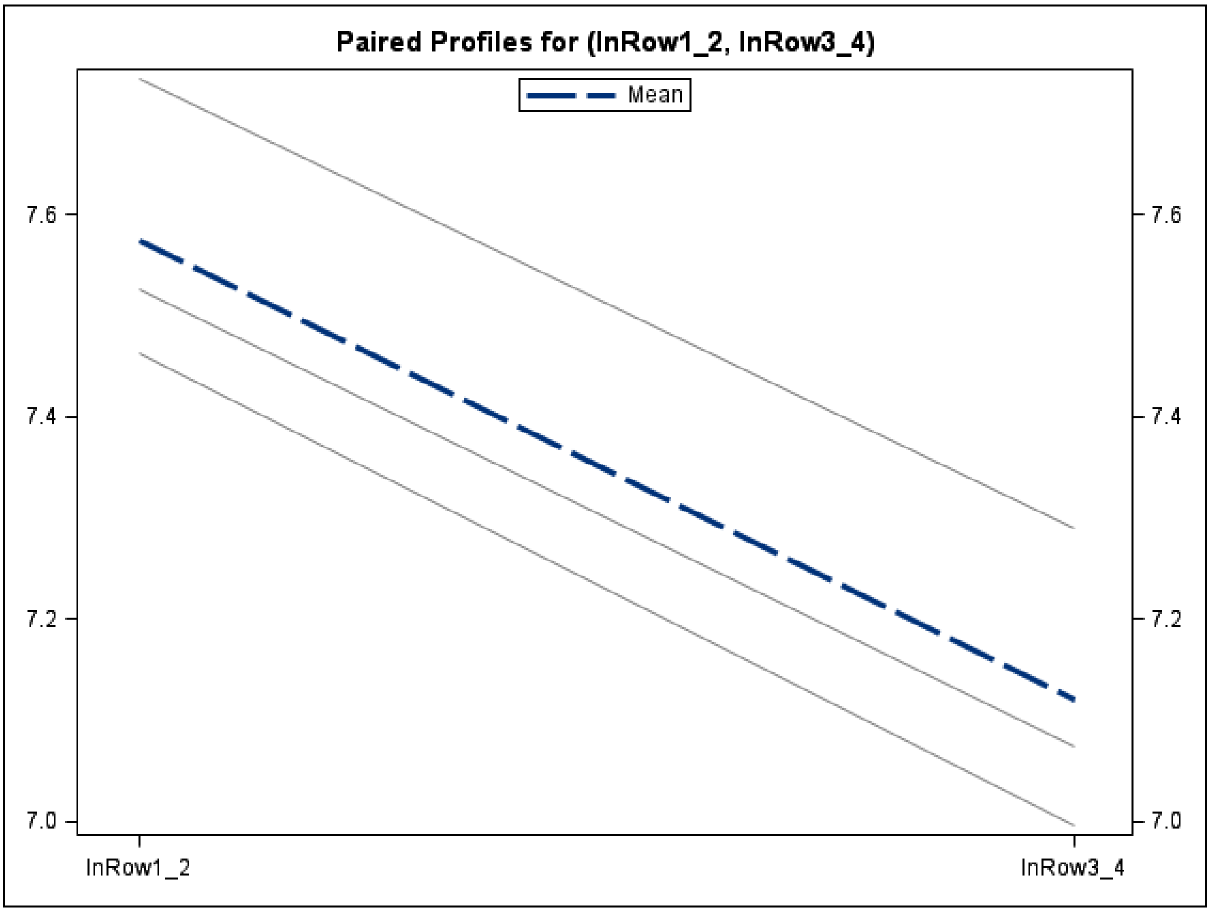


**Figure S10** – Comparison of the combined biomass of rows 1 and 2 versus rows 3 and 4 in a 4-row belt at Site 8 in this publication. This is an *E. polybractea* site measured when the trees were approximately 19 years old. Below is the output from the pair-wise t-test. The solid lines are the natural log–transformed biomass at row 1 and 2 compared to rows 3 and 4. The broken line represents the average biomass.

| N | Mean | Std Dev | Std Err | Minimum | Maximum |
|---|---|---|---|---|---|
| 3 | 0.4548 | 0.0115 | 0.00663 | 0.4441 | 0.4669 |

| Mean | 95% CL Mean | | Std Dev | 95% CL Std Dev | |
|---|---|---|---|---|---|
| 0.4548 | 0.4262 | 0.4833 | 0.0115 | 0.00598 | 0.0722 |

| DF | t Value | Pr > \|t\| |
|---|---|---|
| 2 | 68.56 | 0.0002 |

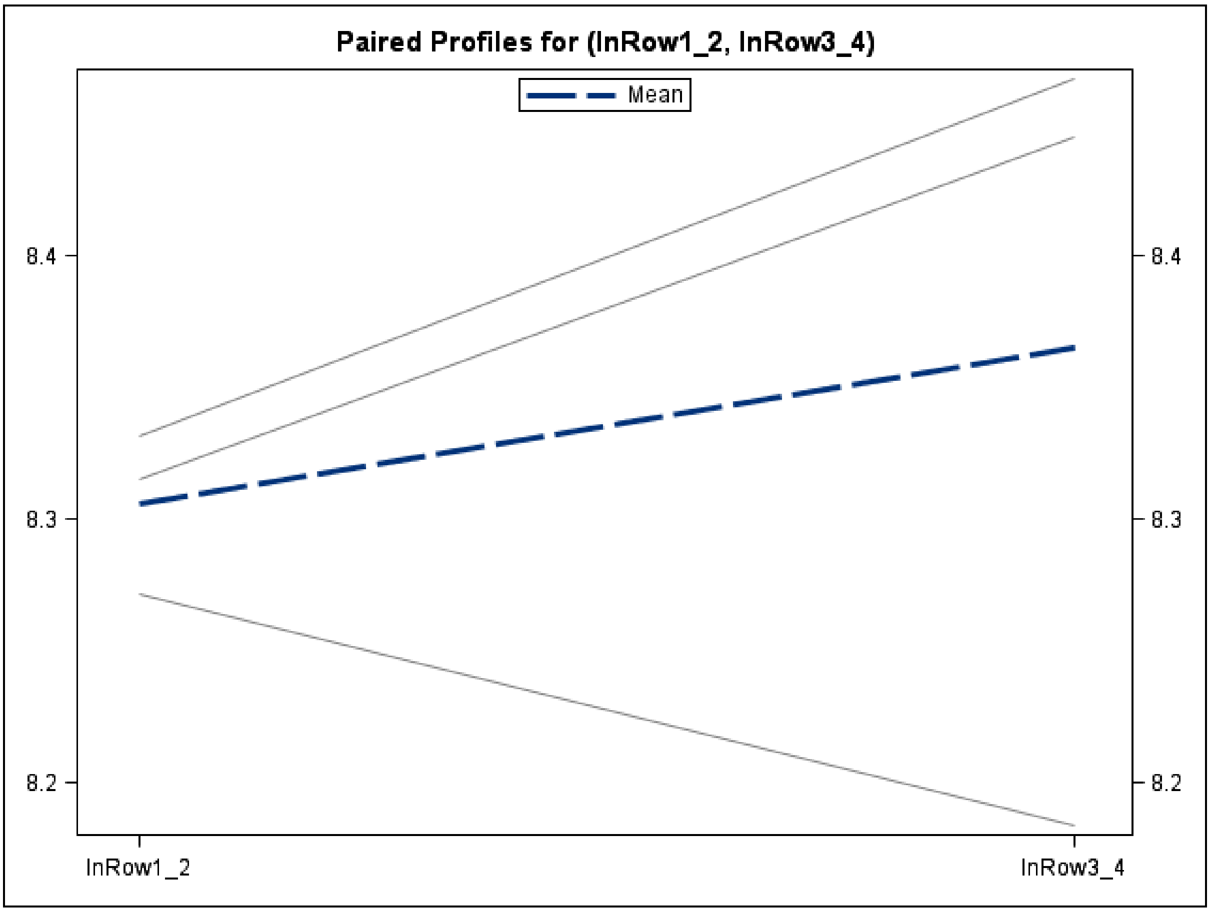


**Figure S11** – Comparison of the combined biomass of rows 1 and 2 versus rows 3 and 4 in a 4-row belt located near Harrowsmith. This is an *E. loxophleba* spp *lissophloia* site measured when the trees were approximately 13 years old. Below is the output from the pair-wise t-test. The solid lines are the natural log–transformed biomass at row 1 and 2 compared to rows 3 and 4. The broken line represents the average biomass.

| N | Mean | Std Dev | Std Err | Minimum | Maximum |
|---|---|---|---|---|---|
| 3 | 0.1242 | 0.2568 | 0.1483 | -0.1721 | 0.2843 |

| Mean | 95% CL Mean | | Std Dev | 95% CL Std Dev | |
|---|---|---|---|---|---|
| 0.1242 | -0.5139 | 0.7622 | 0.2568 | 0.1337 | 1.6141 |

| DF | t Value | Pr > \|t\| |
|---|---|---|
| 2 | 0.84 | 0.4905 |

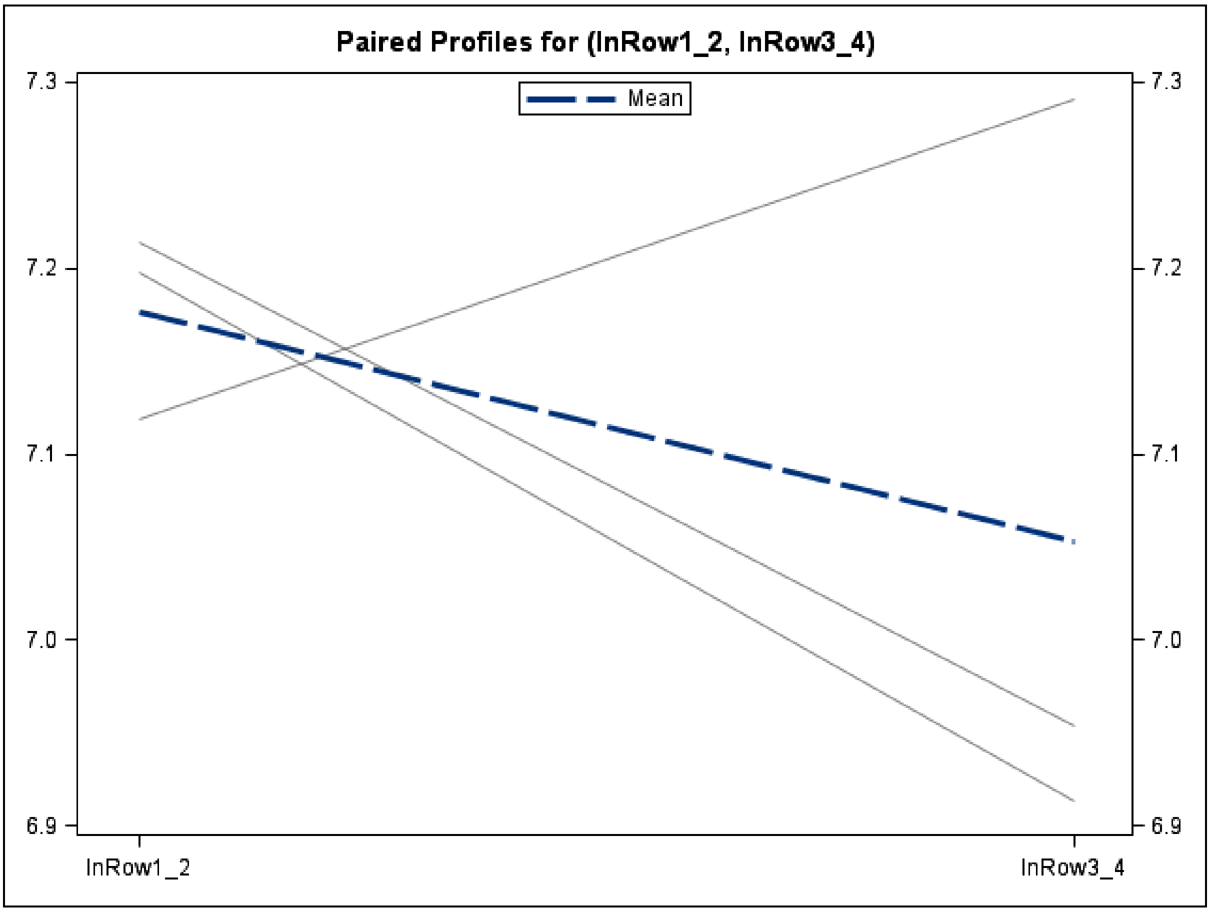


**Figure S12** – Comparison of the combined biomass of rows 1 and 2 versus rows 3 and 4 in a 4-row belt in a Spacing Trial near Narrogin.  This is an *E. polybractea* site measured when the trees were approximately 12 years old. Below is the output from the pair-wise t-test. The solid lines are the natural log–transformed biomass at row 1 and 2 compared to rows 3 and 4. The broken line represents the average biomass.

| N | Mean | Std Dev | Std Err | Minimum | Maximum |
|---|---|---|---|---|---|
| 4 | -0.0317 | 0.1904 | 0.0952 | -0.3170 | 0.0753 |

| Mean | 95% CL Mean | | Std Dev | 95% CL Std Dev | |
|---|---|---|---|---|---|
| -0.0317 | -0.3347 | 0.2712 | 0.1904 | 0.1079 | 0.7099 |

| DF | t Value | Pr > \|t\| |
|---|---|---|
| 3 | -0.33 | 0.7607 |

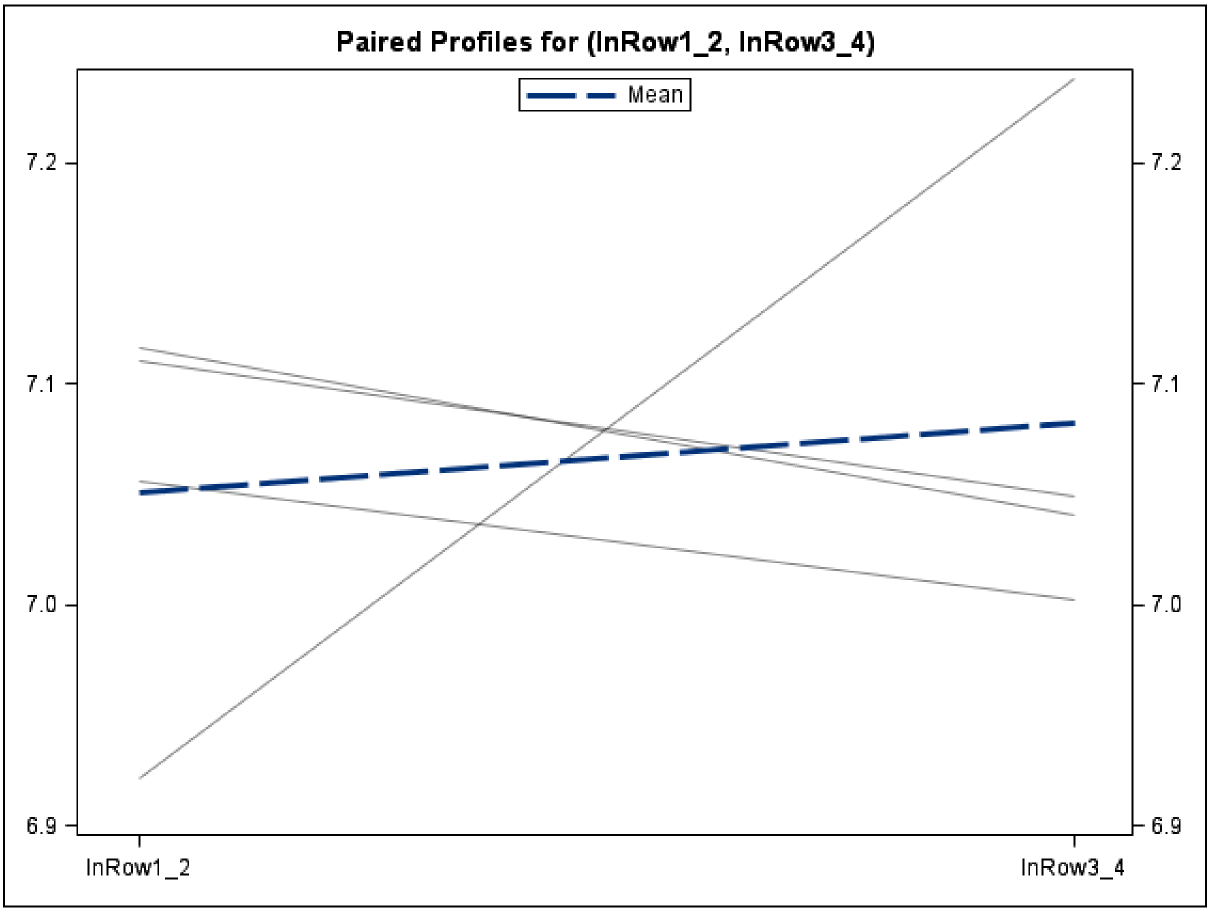


**Figure S13** – Comparison of the combined biomass of rows 1 and 2 versus rows 5 and 6 in a 6-row belt in a Spacing Trial near Cuballing. This is an *E. polybractea* site measured when the trees were approximately 13 years old. Below is the output from the pair-wise t-test. The solid lines are the natural log–transformed biomass at row 1 and 2 compared to rows 5 and 6. The broken line represents the average biomass.

| N | Mean | Std Dev | Std Err | Minimum | Maximum |
|---|---|---|---|---|---|
| 3 | -0.3370 | 0.3112 | 0.1797 | -0.6251 | -0.00697 |

| Mean | 95% CL Mean | | Std Dev | 95% CL Std Dev | |
|---|---|---|---|---|---|
| -0.3370 | -1.1101 | 0.4360 | 0.3112 | 0.1620 | 1.9557 |

| DF | t Value | Pr > \|t\| |
|---|---|---|
| 2 | -1.88 | 0.2015 |

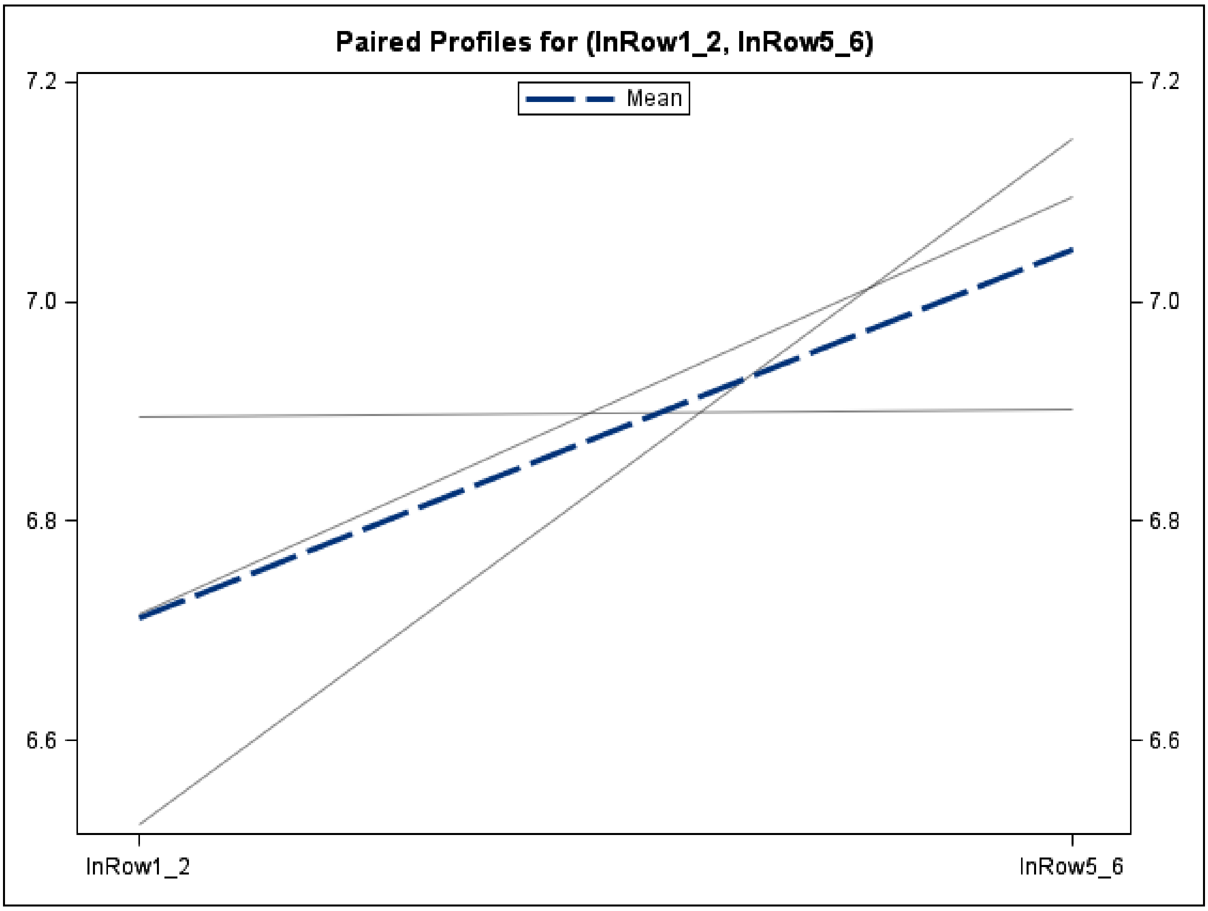
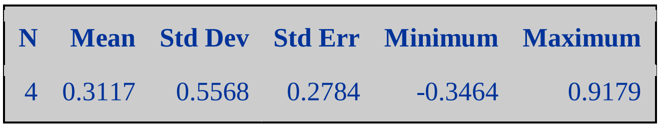
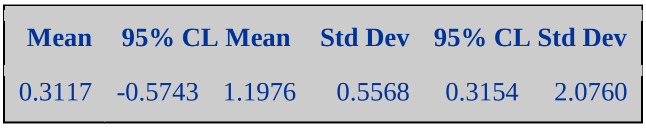


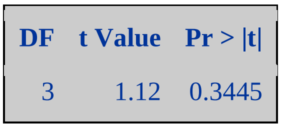
**Figure S14** – Comparison of the combined biomass of rows 1 and 2 versus rows 5 and 6 in a 6-row belt in a Spacing Trial near Pingelly. This is an *E. loxophleba* spp *lissophloia*_site measured when the trees were approximately 13 years old. Below is the output from the pair-wise t-test. The solid lines are the natural log–transformed biomass at row 1 and 2 compared to rows 5 and 6. The broken line represents the average biomass.



| N | Mean | Std Dev | Std Err | Minimum | Maximum |
|---|---|---|---|---|---|
| 4 | 0.3117 | 0.5568 | 0.2784 | -0.3464 | 0.9179 |



| Mean | 95% CL Mean | | Std Dev | 95% CL Std Dev | |
|---|---|---|---|---|---|
| 0.3117 | -0.5743 | 1.1976 | 0.5568 | 0.3154 | 2.0760 |



| DF | t Value | Pr > \|t\| |
|---|---|---|
| 3 | 1.12 | 0.3445 |

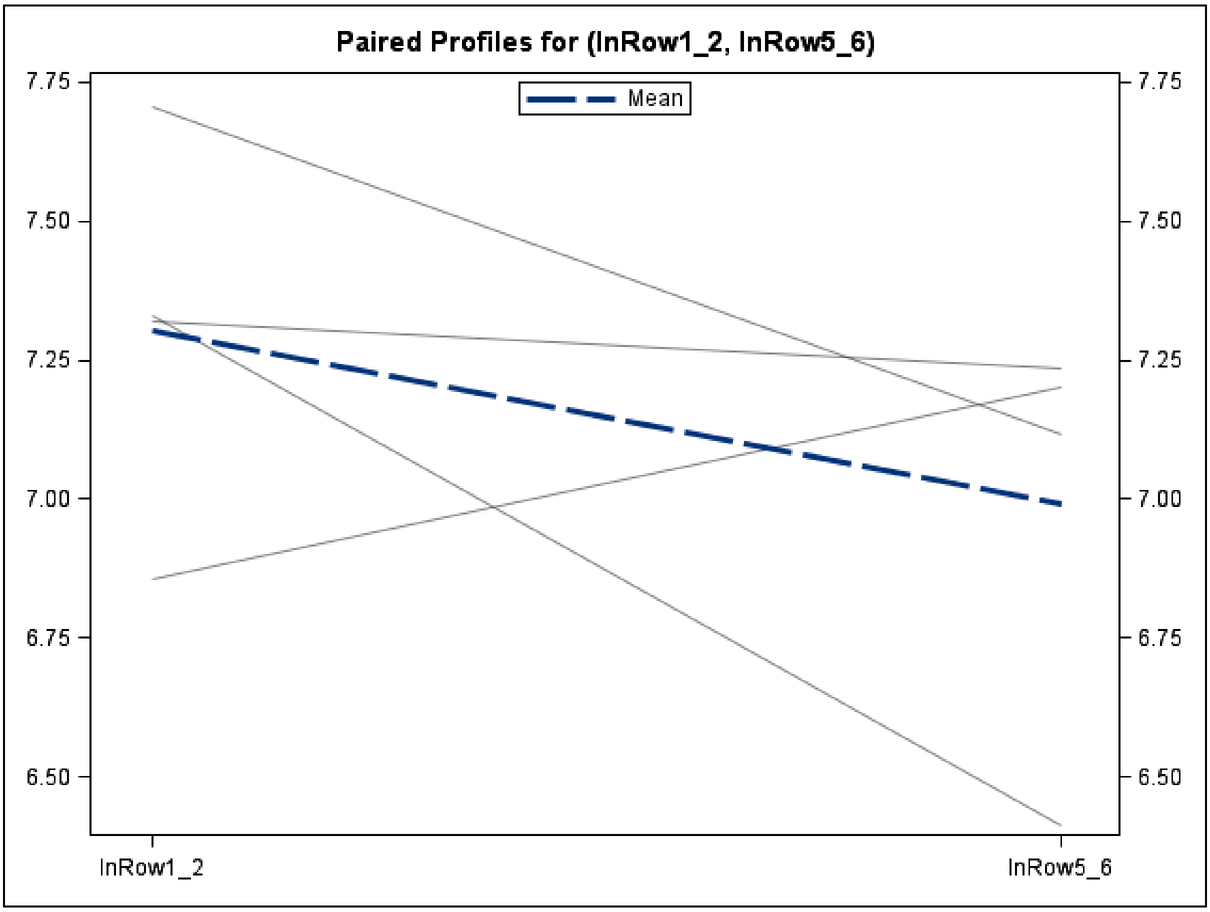


**Figure S15** – Comparison of the combined biomass of rows 1 and 2 versus rows 5 and 6 in a 6-row belt at Site 12 in this publication. This is an *E. loxophleba* spp *lissophloia* coppice site measured 5 years after harvest and the trees were established 17 prior to measurement. Below is the output from the pair-wise t-test. The solid lines are the natural log–transformed biomass at row 1 and 2 compared to rows 5 and 6. The broken line represents the average biomass.

| N | Mean | Std Dev | Std Err | Minimum | Maximum |
|---|---|---|---|---|---|
| 3 | 0.1231 | 0.0616 | 0.0356 | 0.0823 | 0.1940 |

| Mean | 95% CL Mean | | Std Dev | 95% CL Std Dev | |
|---|---|---|---|---|---|
| 0.1231 | -0.0300 | 0.2762 | 0.0616 | 0.0321 | 0.3873 |

| DF | t Value | Pr > \|t\| |
|---|---|---|
| 2 | 3.46 | 0.0744 |

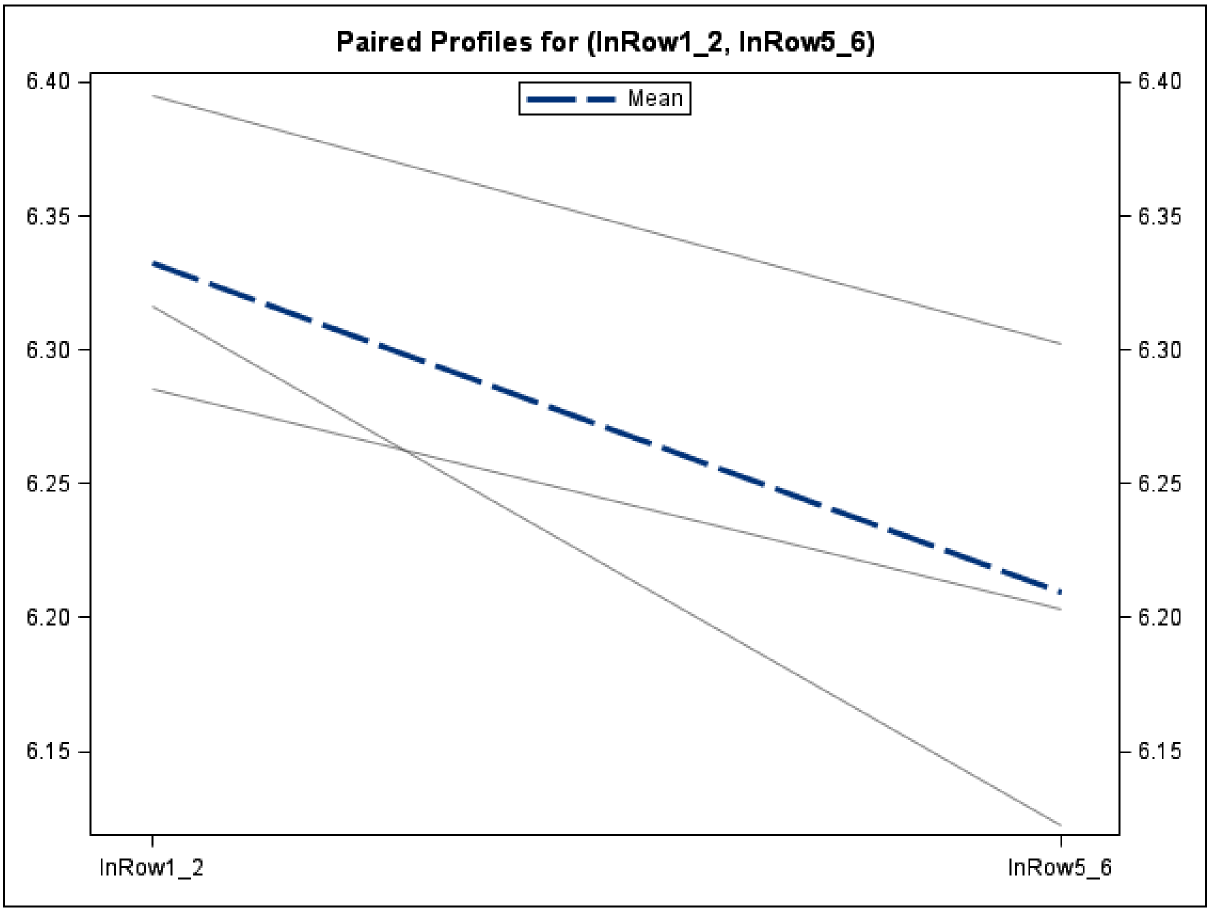


**Figure S16** – Comparison of the combined biomass of rows 1 and 2 versus rows 5 and 6 in a 6-row belt at Site 12 in this publication. This is an *E. loxophleba* spp *lissophloia* site measured when the trees were approximately 17 years old. Below is the output from the pair-wise t-test. The solid lines are the natural log–transformed biomass at row 1 and 2 compared to rows 5 and 6. The broken line represents the average biomass.

| N | Mean | Std Dev | Std Err | Minimum | Maximum |
|---|---|---|---|---|---|
| 3 | -0.0422 | 0.2141 | 0.1236 | -0.1966 | 0.2021 |

| Mean | 95% CL Mean | | Std Dev | 95% CL Std Dev | |
|---|---|---|---|---|---|
| -0.0422 | -0.5740 | 0.4895 | 0.2141 | 0.1115 | 1.3453 |

| DF | t Value | Pr > \|t\| |
|---|---|---|
| 2 | -0.34 | 0.7651 |

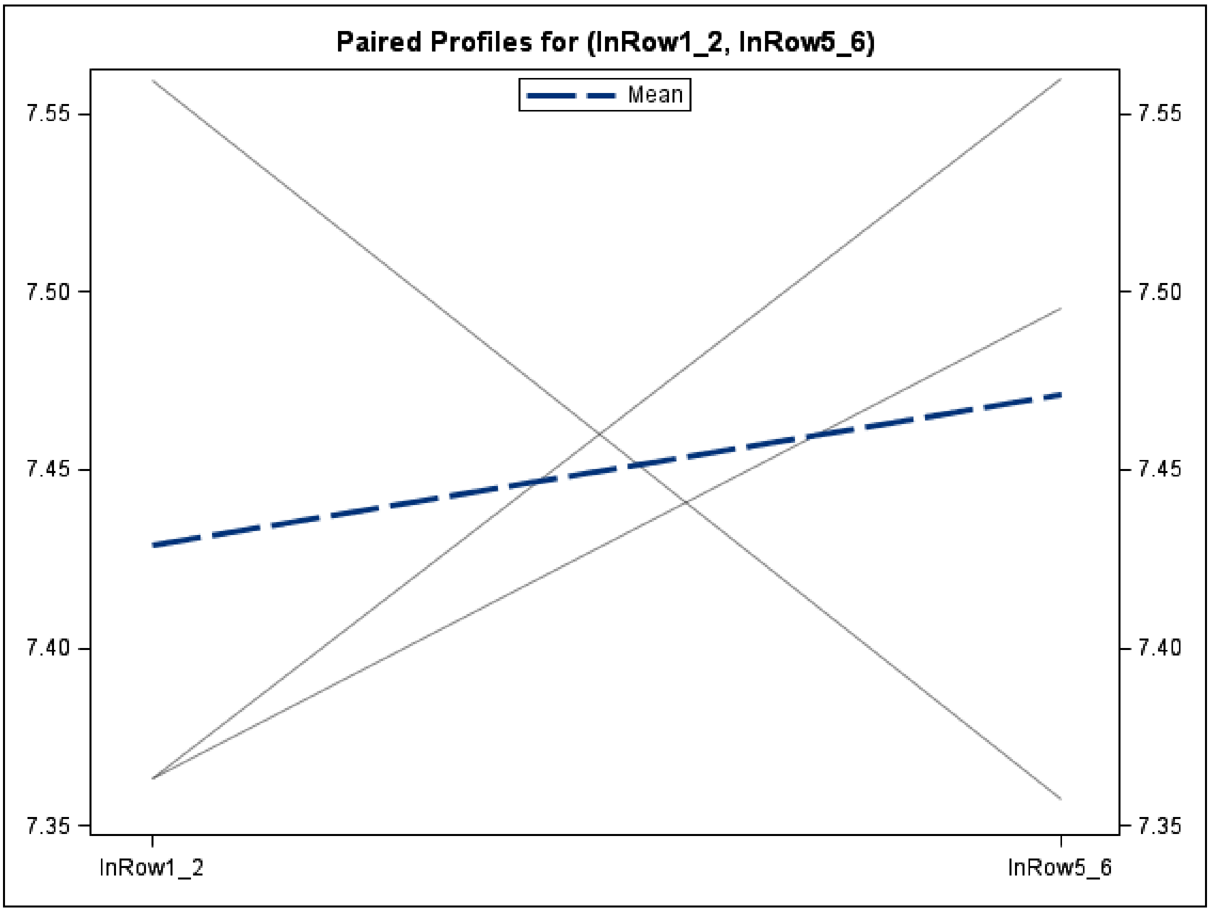


**Figure S17** – Comparison of the combined biomass of rows 1 and 2 versus rows 5 and 6 in a 6-row belt located at Harrowsmith. This is an *E. loxophleba* spp *lissophloia* site measured when the trees were approximately 13 years old. Below is the output from the pair-wise t-test. The solid lines are the natural log–transformed biomass at row 1 and 2 compared to rows 5 and 6. The broken line represents the average biomass.

| N | Mean | Std Dev | Std Err | Minimum | Maximum |
|---|---|---|---|---|---|
| 3 | -0.0963 | 0.2575 | 0.1487 | -0.3625 | 0.1516 |

| Mean | 95% CL Mean | | Std Dev | 95% CL Std Dev | |
|---|---|---|---|---|---|
| -0.0963 | -0.7361 | 0.5435 | 0.2575 | 0.1341 | 1.6186 |

| DF | t Value | Pr > \|t\| |
|---|---|---|
| 2 | -0.65 | 0.5835 |

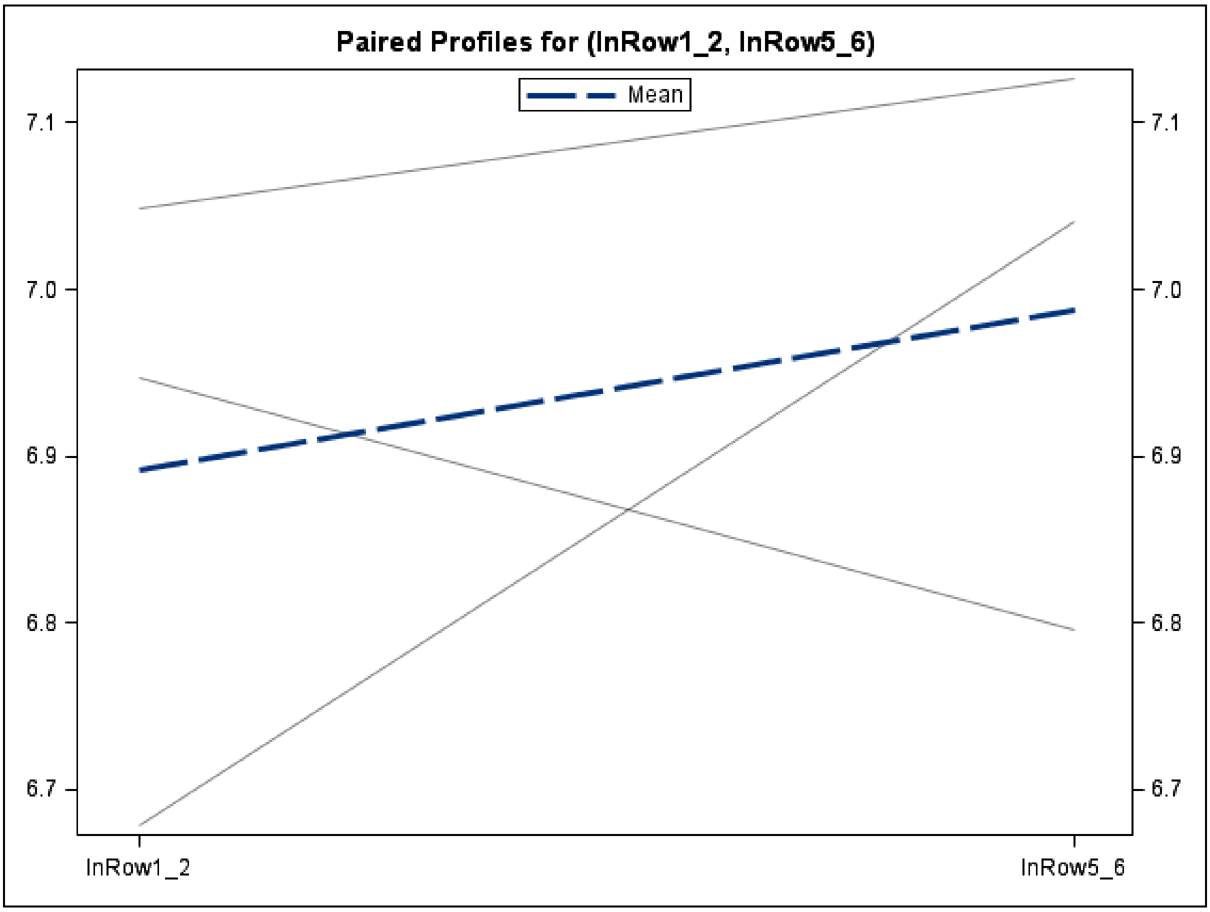
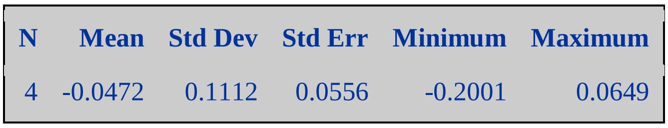
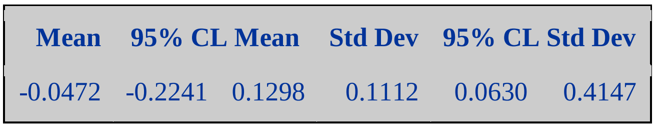


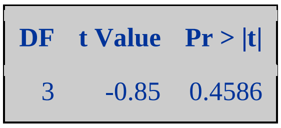
**Figure S18** – Comparison of the combined biomass of rows 1 and 2 versus rows 5 and 6 in a 6-row belt in a Spacing Trial near Narrogin. This is an *E. polybractea* site measured when the trees were approximately 12 years old. Below is the output from the pair-wise t-test. The solid lines are the natural log–transformed biomass at row 1 and 2 compared to rows 5 and 6. The broken line represents the average biomass.



| N | Mean | Std Dev | Std Err | Minimum | Maximum |
|---|---|---|---|---|---|
| 4 | -0.0472 | 0.1112 | 0.0556 | -0.2001 | 0.0649 |



| Mean | 95% CL Mean | | Std Dev | 95% CL Std Dev | |
|---|---|---|---|---|---|
| -0.0472 | -0.2241 | 0.1298 | 0.1112 | 0.0630 | 0.4147 |



| DF | t Value | Pr > \|t\| |
|---|---|---|
| 3 | -0.85 | 0.4586 |

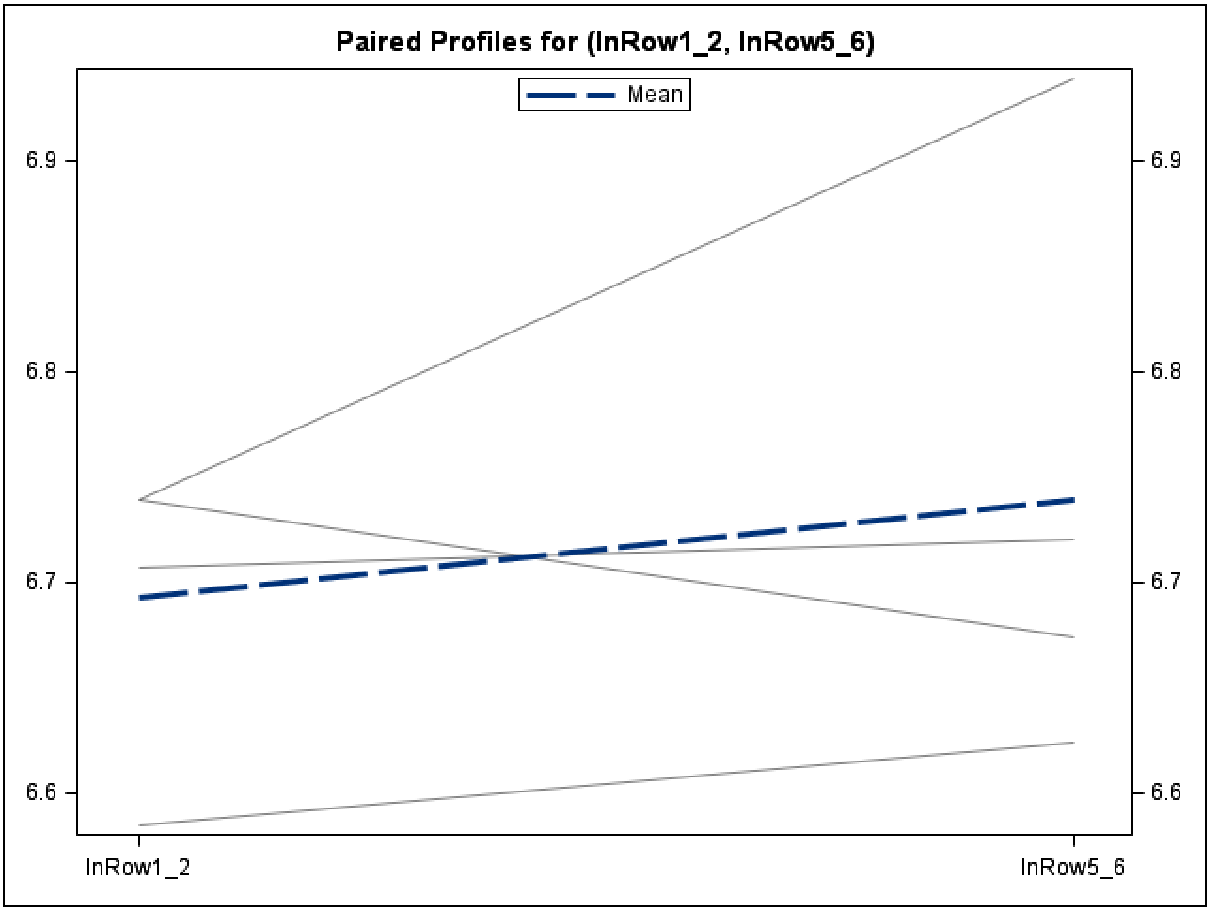
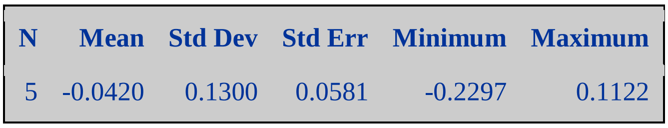
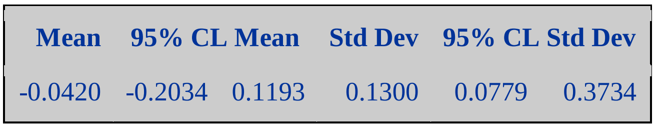


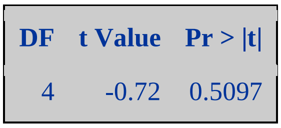
**Figure S19** – Comparison of the combined biomass of rows 1 and 2 versus rows 7 and 8 in an 8-row belt on the same property at Sites 18 and 19 in this publication. This is an *E. polybractea* site measured when the trees were approximately 9 years old. Below is the output from the pair-wise t-test. The solid lines are the natural log–transformed biomass at row 1 and 2 compared to rows 7 and 8. The broken line represents the average biomass.



| N | Mean | Std Dev | Std Err | Minimum | Maximum |
|---|---|---|---|---|---|
| 5 | -0.0420 | 0.1300 | 0.0581 | -0.2297 | 0.1122 |



| Mean | 95% CL Mean | | Std Dev | 95% CL Std Dev | |
|---|---|---|---|---|---|
| -0.0420 | -0.2034 | 0.1193 | 0.1300 | 0.0779 | 0.3734 |



| DF | t Value | Pr > \|t\| |
|---|---|---|
| 4 | -0.72 | 0.5097 |

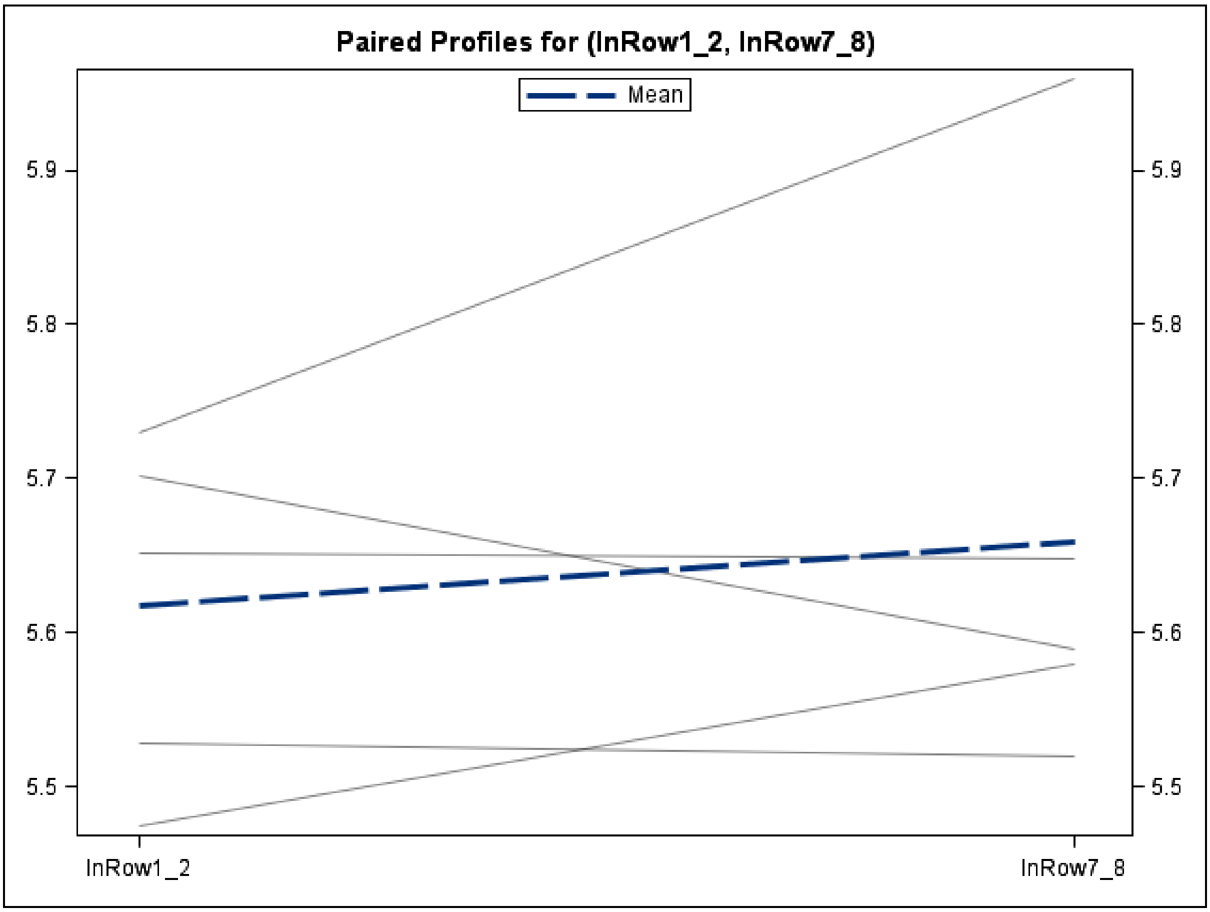
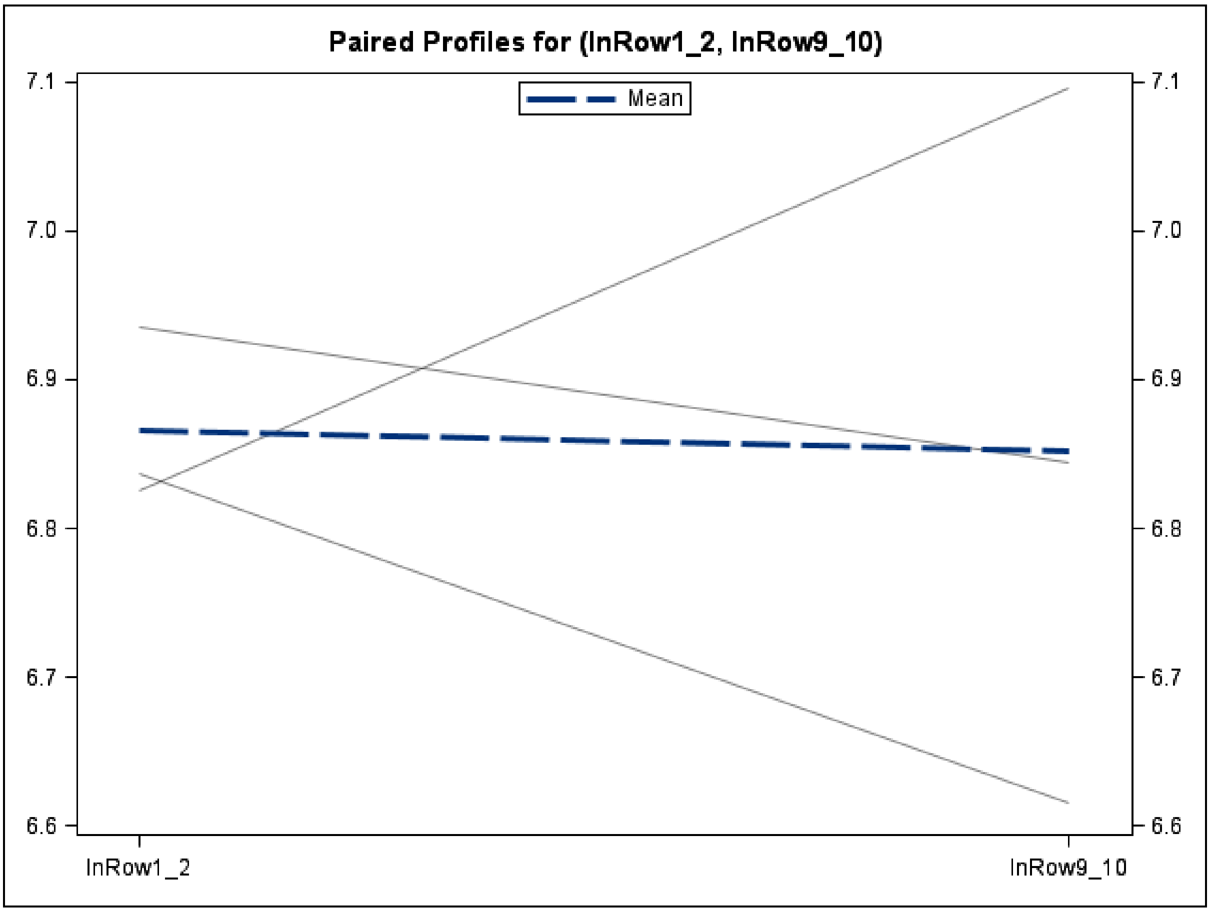


**Figure S20** – Comparison of the combined biomass of rows 1 and 2 versus rows 9 and 10 in a 10-row belt located near Harrowsmith. This is an *E. loxophleba* spp *lissophloia* site measured when the trees were approximately 13 years old. Below is the output from the pair-wise t-test. The solid lines are the natural log–transformed biomass at row 1 and 2 compared to rows 9 and 10. The broken line represents the average biomass.

| N | Mean | Std Dev | Std Err | Minimum | Maximum |
|---|---|---|---|---|---|
| 3 | 0.0143 | 0.2550 | 0.1472 | -0.2702 | 0.2224 |

| Mean | 95% CL Mean | | Std Dev | 95% CL Std Dev | |
|---|---|---|---|---|---|
| 0.0143 | -0.6191 | 0.6478 | 0.2550 | 0.1328 | 1.6026 |

| DF | t Value | Pr > \|t\| |
|---|---|---|
| 2 | 0.10 | 0.9315 |



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Peck, A., Sudmeyer, R., Huxtable, D., Bartle, J., & Mendham, D. (2012). Productivity of mallee agroforestry systems under various harvest and competition management regimes. RIRDC. Publication No 11/162. Rural Industries Research and Development Corporation, Canberra. Australia.

Sudmeyer, R., Daniels, T., Jones, H., & Huxtable, D. (2012). The extent and cost of mallee–crop competition in unharvested carbon sequestration and harvested mallee biomass agroforestry systems. *Crop and Pasture Science, 63*(6), 555-569.

Spencer, B., Bartle, J., Huxtable, D., Mazanec, R., Abadi, A., Gibberd, M., and Zerihun, A. (2019). A decadal multi-site study of the effects of frequency and season of harvest on biomass production from mallee eucalypts. *Unpublished*.

# Appendix B:

## Supplementary material- Chapter 4

## Supplementary Material

# Flowering phenology in a *Eucalyptus loxophleba* seed orchard, heritability and genetic correlation with biomass production and cineole – breeding strategy implications

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Primary research manuscript

Table S1 – The duration of a flowering phase in the 2014 assessment determined by the month flowering commenced. Separate assessments were made for trees that underwent two reproductive events.

| Month start flowering | No. trees flowering | Average flowering duration (weeks) |
|---|---|---|
| February | 2 | 21.5 |
| March | 13 | 22.4 |
| April | 151 | 22.3 |
| May | 240 | 20.4 |
| June | 189 | 17.8 |
| July | 127 | 15.7 |
| August | 120 | 12.7 |
| September | 77 | 10.9 |

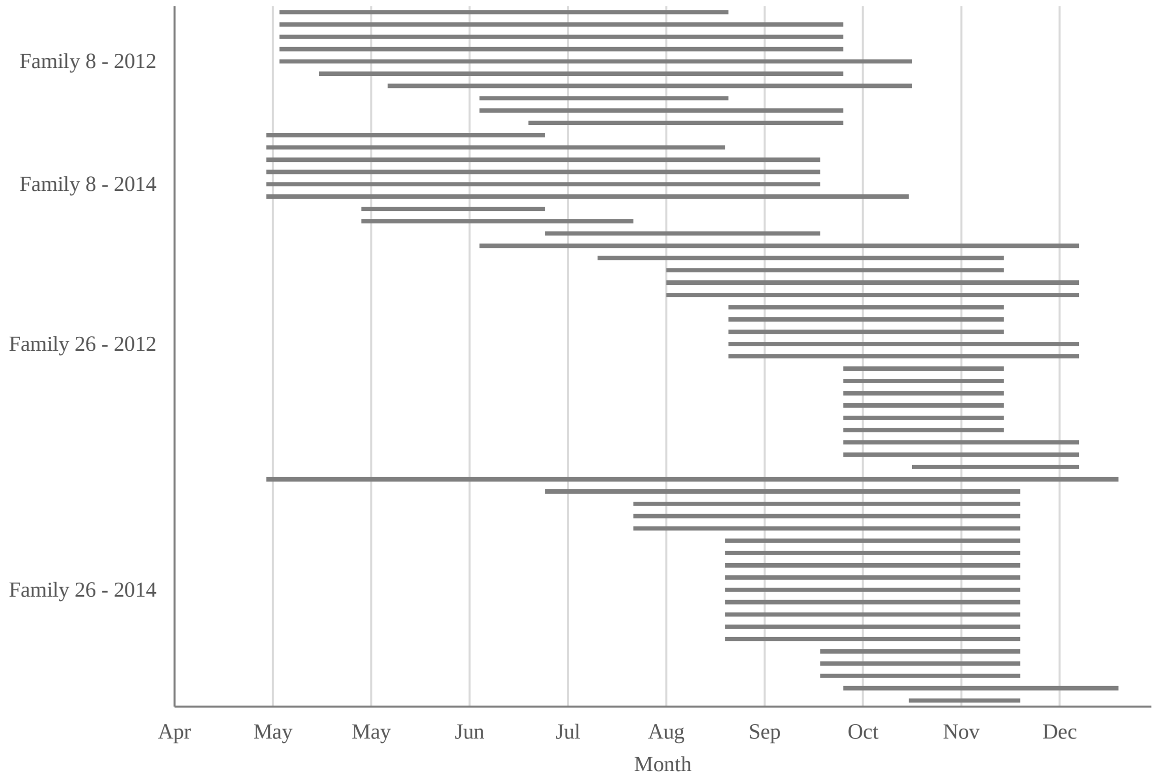

Fig. S1 Variation in flowering time for individual trees from two families (8 and 26) over two years (2012 and 2014). Family 8 was the earliest flowering family and Family 26 was the latest.

Table S2 – Least square means of each family for start flowering week, end flowering week and duration of flowering with the standard error in brackets (SE). Untransformed proportion or individuals flowering (%) in 2012 and 2014, and the proportion of trees that flowered twice (dual flowering) from each family. DUM = Dumbleyung, LGR = Lake Grace, COO = Coolgardie, GOG = Goongarrie, NOR = Norseman, NAR = Narembeen, SNC = Southern Cross, TYG = Trayning, WES = Westonia.

| Provenance | Family | Start flowering week (SE) | End flowering week (SE) | Duration flowering (SE) | % flowered 2012 | % flowered 2014 | % dual flowering |
|---|---|---|---|---|---|---|---|
| DUM | 1 | 24.3 (1.2) | 46.4 (0.6) | 23.2 (1.2) | 84.2 | 78.9 | 36.8 |
| DUM | 2 | 26.0 (1.3) | 46.1 (0.6) | 21.2 (1.3) | 83.3 | 77.8 | 16.7 |
| DUM | 3 | 26.0 (1.2) | 48.0 (0.6) | 23.3 (1.2) | 75.0 | 85.0 | 45.0 |
| DUM | 4 | 26.2 (1.5) | 44.2 (0.7) | 19.0 (1.5) | 73.3 | 73.3 | 0.0 |
| DUM | 5 | 28.9 (1.2) | 45.5 (0.6) | 17.5 (1.2) | 85.0 | 85.0 | 5.0 |
| DUM | 6 | 30.9 (1.2) | 46.8 (0.5) | 17.1 (1.2) | 100.0 | 84.2 | 10.5 |
| DUM | 7 | 31.8 (1.2) | 45.1 (0.6) | 14.5 (1.2) | 71.4 | 76.2 | 4.8 |
| LGR | 8 | 20.4 (1.6) | 40.2 (0.7) | 21.1 (1.6) | 62.5 | 56.3 | 0.0 |
| LGR | 9 | 26.3 (1.1) | 45.9 (0.5) | 20.6 (1.1) | 95.0 | 100.0 | 0.0 |
| LGR | 10 | 26.7 (1.3) | 43.7 (0.6) | 18.1 (1.3) | 81.0 | 61.9 | 0.0 |
| LGR | 11 | 27.3 (1.2) | 46.5 (0.6) | 20.3 (1.2) | 84.2 | 78.9 | 31.6 |
| LGR | 12 | 27.4 (1.2) | 47.4 (0.5) | 21.0 (1.2) | 95.0 | 80.0 | 50.0 |
| LGR | 13 | 28.2 (1.2) | 46.4 (0.5) | 19.3 (1.2) | 95.0 | 80.0 | 40.0 |
| LGR | 14 | 28.2 (1.2) | 46.5 (0.6) | 19.4 (1.2) | 94.1 | 94.1 | 35.3 |
| LGR | 15 | 29.3 (1.2) | 45.5 (0.6) | 17.0 (1.2) | 85.0 | 70.0 | 20.0 |
| LGR | 16 | 29.4 (1.3) | 45.4 (0.6) | 17.0 (1.3) | 65.0 | 85.0 | 0.0 |
| LGR | 17 | 29.6 (1.1) | 47.5 (0.5) | 18.9 (1.2) | 85.7 | 85.7 | 28.6 |
| LGR | 18 | 30.7 (1.2) | 46.9 (0.6) | 17.1 (1.2) | 85.0 | 70.0 | 25.0 |
| LGR | 19 | 31.0 (1.1) | 48.3 (0.5) | 18.3 (1.1) | 90.9 | 90.9 | 54.5 |
| LGR | 20 | 31.5 (1.2) | 46.1 (0.6) | 15.6 (1.2) | 70.0 | 90.0 | 5.0 |
| LGR | 21 | 32.9 (1.6) | 45.4 (0.7) | 13.5 (1.6) | 64.7 | 47.1 | 0.0 |
| COO | 22 | 20.0 (1.2) | 44.8 (0.6) | 26.1 (1.2) | 83.3 | 94.4 | 22.2 |
| COO | 23 | 20.8 (1.1) | 49.4 (0.5) | 29.7 (1.2) | 94.7 | 94.7 | 57.9 |
| COO | 24 | 24.6 (1.3) | 46.1 (0.6) | 22.5 (1.3) | 81.3 | 93.8 | 0.0 |
| GOG | 25 | 29.0 (1.2) | 45.6 (0.6) | 17.7 (1.2) | 70.0 | 90.0 | 5.0 |
| GOG | 26 | 37.7 (1.1) | 51.2 (0.5) | 14.5 (1.1) | 100.0 | 100.0 | 5.3 |
| NOR | 27 | 20.6 (1.1) | 46.9 (0.5) | 28.1 (1.1) | 95.0 | 90.0 | 45.0 |
| NOR | 28 | 25.1 (1.2) | 47.3 (0.5) | 23.2 (1.2) | 90.0 | 85.0 | 5.0 |
| NOR | 29 | 25.2 (1.2) | 46.5 (0.6) | 22.3 (1.2) | 86.4 | 68.2 | 9.1 |
| NOR | 30 | 25.8 (1.2) | 50.0 (0.5) | 25.7 (1.2) | 100.0 | 94.4 | 50.0 |
| NOR | 31 | 27.4 (1.2) | 49.8 (0.6) | 23.7 (1.2) | 94.4 | 88.9 | 55.6 |
| NOR | 32 | 28.2 (1.1) | 49.3 (0.5) | 22.3 (1.2) | 100.0 | 100.0 | 33.3 |
| NOR | 33 | 29.1 (1.1) | 50.9 (0.5) | 23.4 (1.1) | 95.0 | 95.0 | 50.0 |
| NOR | 34 | 30.2 (1.2) | 50.1 (0.6) | 20.9 (1.2) | 84.2 | 84.2 | 42.1 |
| NOR | 35 | 31.6 (1.1) | 52.3 (0.5) | 21.9 (1.1) | 100.0 | 90.0 | 35.0 |
| NOR | 36 | 33.7 (1.1) | 50.9 (0.5) | 18.6 (1.1) | 95.0 | 95.0 | 25.0 |
| NOR | 37 | 34.5 (1.1) | 52.0 (0.5) | 18.7 (1.1) | 85.7 | 100.0 | 33.3 |
| NAR | 38 | 27.7 (1.2) | 45.7 (0.6) | 18.9 (1.2) | 95.0 | 75.0 | 0.0 |
| NAR | 39 | 27.8 (1.1) | 44.6 (0.5) | 17.8 (1.1) | 95.2 | 100.0 | 4.8 |
| NAR | 40 | 29.7 (1.2) | 46.4 (0.6) | 17.8 (1.2) | 94.4 | 77.8 | 0.0 |
| NAR | 41 | 30.9 (1.2) | 46.4 (0.6) | 16.4 (1.2) | 94.7 | 84.2 | 0.0 |
| SNC | 42 | 21.5 (1.2) | 44.7 (0.6) | 24.1 (1.2) | 77.8 | 100.0 | 5.6 |
| SNC | 43 | 24.1 (1.1) | 45.6 (0.5) | 22.7 (1.2) | 85.0 | 95.0 | 0.0 |
| SNC | 44 | 27.2 (1.3) | 44.2 (0.6) | 17.9 (1.3) | 68.4 | 73.7 | 5.3 |
| SNC | 45 | 29.7 (1.2) | 45.8 (0.6) | 17.0 (1.2) | 88.9 | 88.9 | 0.0 |
| SNC | 46 | 33.7 (1.3) | 51.3 (0.6) | 18.9 (1.3) | 100.0 | 100.0 | 13.3 |
| TYG | 47 | 21.9 (1.2) | 42.3 (0.5) | 21.6 (1.2) | 89.5 | 94.7 | 0.0 |
| TYG | 48 | 24.9 (1.1) | 45.5 (0.5) | 21.6 (1.1) | 100.0 | 100.0 | 5.0 |
| TYG | 49 | 26.8 (1.2) | 46.1 (0.5) | 20.3 (1.2) | 89.5 | 94.7 | 0.0 |
| TYG | 50 | 28.3 (1.2) | 45.4 (0.6) | 18.1 (1.2) | 100.0 | 100.0 | 0.0 |
| TYG | 51 | 28.7 (1.3) | 46.7 (0.6) | 19.0 (1.3) | 100.0 | 92.9 | 0.0 |
| TYG | 52 | 28.8 (1.1) | 47.3 (0.5) | 19.5 (1.1) | 81.0 | 100.0 | 4.8 |
| TYG | 53 | 29.4 (1.1) | 44.0 (0.5) | 15.6 (1.2) | 85.0 | 95.0 | 0.0 |
| TYG | 54 | 29.5 (1.2) | 48.3 (0.5) | 19.8 (1.2) | 76.2 | 95.2 | 4.8 |
| TYG | 55 | 29.6 (1.2) | 46.8 (0.6) | 18.2 (1.2) | 88.9 | 88.9 | 0.0 |
| TYG | 56 | 29.7 (1.2) | 48.1 (0.6) | 19.3 (1.3) | 68.4 | 94.7 | 0.0 |
| TYG | 57 | 29.7 (1.1) | 45.9 (0.5) | 17.2 (1.1) | 95.0 | 95.0 | 0.0 |
| WES | 58 | 24.1 (1.4) | 44.7 (0.7) | 21.8 (1.4) | 85.7 | 85.7 | 14.3 |
| WES | 59 | 30.6 (1.2) | 48.3 (0.6) | 18.6 (1.2) | 94.4 | 88.9 | 11.1 |
| WES | 60 | 32.7 (1.2) | 47.1 (0.5) | 15.5 (1.2) | 94.4 | 100.0 | 0.0 |

# Appendix C:

## Supplementary material- Chapter 5

# Sapling and coppice biomass heritabilities and potential gains from *Eucalyptus polybractea* progeny trials

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Keywords: coppice biomass, Eucalyptus polybractea, net breeding values, heritability, sapling biomass, genetic and phenotypic correlation, selection simulation

Supplementary material starts on page 2

Table S1 The total number of *E. polybractea* saplings and coppice across the three progeny trials at Newdegate, Drummartin and Condobolin and the proportion of the saplings that reshot (coppiced) after harvest.

| Population | Family | No. saplings | No. coppice | % coppiced |
|---|---|---|---|---|
| Charcoal Tank Rd | 25 | 54 | 54 | 1.00 |
| Kerrs Lane | 5 | 63 | 59 | 0.94 |
| | 6 | 55 | 55 | 1.00 |
| Talimba Rd East | 44 | 60 | 59 | 0.98 |
| Talimba Rd West | 45 | 51 | 50 | 0.98 |
| West Mid-West Hwy | 28 | 64 | 64 | 1.00 |
| | 29 | 59 | 59 | 1.00 |
| | 30 | 60 | 59 | 0.98 |
| | 31 | 63 | 62 | 0.98 |
| West Wyalong Town | 1 | 55 | 55 | 1.00 |
| | 2 | 51 | 50 | 0.98 |
| | 3 | 56 | 56 | 1.00 |
| | 4 | 57 | 55 | 0.96 |
| | 26 | 58 | 58 | 1.00 |
| | 27 | 55 | 54 | 0.98 |
| | 36 | 58 | 57 | 0.98 |
| | 37 | 56 | 56 | 1.00 |
| | 38 | 58 | 57 | 0.98 |
| | 39 | 62 | 62 | 1.00 |
| | 40 | 56 | 56 | 1.00 |
| | 41 | 60 | 58 | 0.97 |
| | 42 | 53 | 53 | 1.00 |
| | 43 | 57 | 57 | 1.00 |
| | 46 | 60 | 60 | 1.00 |
| | 47 | 64 | 62 | 0.97 |
| | 48 | 56 | 56 | 1.00 |
| | 49 | 53 | 53 | 1.00 |
| | 56 | 53 | 53 | 1.00 |
| | 57 | 61 | 61 | 1.00 |
| | 58 | 51 | 49 | 0.96 |
| | 59 | 60 | 60 | 1.00 |
| | 60 | 61 | 60 | 0.98 |
| | 61 | 57 | 54 | 0.95 |
| | 62 | 57 | 56 | 0.98 |
| | 63 | 54 | 54 | 1.00 |
| | 64 | 62 | 62 | 1.00 |
| | 65 | 58 | 57 | 0.98 |
| | 66 | 47 | 47 | 1.00 |
| West Yalgogrin | 32 | 56 | 54 | 0.96 |
| | 33 | 54 | 54 | 1.00 |
| | 34 | 57 | 57 | 1.00 |
| | 35 | 61 | 61 | 1.00 |
| Winters Lane | 7 | 52 | 52 | 1.00 |
| | 8 | 53 | 52 | 0.98 |
| | 9 | 57 | 58 | 1.02 |
| | 10 | 61 | 61 | 1.00 |
| | 11 | 59 | 58 | 0.98 |
| | 12 | 51 | 49 | 0.96 |
| | 13 | 54 | 54 | 1.00 |
| | 14 | 62 | 59 | 0.95 |
| | 15 | 61 | 61 | 1.00 |
| | 16 | 56 | 55 | 0.98 |
| | 17 | 58 | 58 | 1.00 |
| | 18 | 61 | 60 | 0.98 |
| | 19 | 50 | 48 | 0.96 |
| | 20 | 54 | 54 | 1.00 |
| | 21 | 60 | 60 | 1.00 |
| | 22 | 57 | 56 | 0.98 |
| | 23 | 62 | 60 | 0.97 |
| | 24 | 63 | 63 | 1.00 |
| | 50 | 56 | 56 | 1.00 |
| | 51 | 55 | 54 | 0.98 |
| | 52 | 59 | 59 | 1.00 |
| | 53 | 53 | 52 | 0.98 |
| | 54 | 56 | 56 | 1.00 |
| | 55 | 56 | 55 | 0.98 |

Table S2 Best Linear Unbiased Estimates (± standard errors) for three biomass assessment at the Newdegate *E. polybractea* progeny trial.

| Provenance | Family | 3-year sapling | 6-year sapling | Coppice |
|---|---|---|---|---|
| Charcoal Tank Rd | 25 | 1.6860 ± 0.0950 | 3.2679 ± 0.1644 | 1.8692 ± 0.0920 |
| Kerrs Lane | 5 | 1.4633 ± 0.0873 | 3.1734 ± 0.1436 | 1.8500 ± 0.0862 |
| | 6 | 1.5764 ± 0.0983 | 3.2548 ± 0.1707 | 1.8294 ± 0.0948 |
| Talimba Rd East | 44 | 1.5859 ± 0.0886 | 3.3229 ± 0.1470 | 1.8047 ± 0.0863 |
| Talimba Rd West | 45 | 1.7119 ± 0.0886 | 3.5819 ± 0.1548 | 1.9617 ± 0.0888 |
| West Mid-West Hwy | 28 | 1.5238 ± 0.0873 | 2.9563 ± 0.1436 | 1.6691 ± 0.0851 |
| | 29 | 1.4976 ± 0.0873 | 2.7640 ± 0.1470 | 1.6687 ± 0.0862 |
| | 30 | 1.5341 ± 0.0886 | 3.3781 ± 0.1509 | 1.9074 ± 0.0876 |
| | 31 | 1.8357 ± 0.0861 | 3.1139 ± 0.1404 | 1.7891 ± 0.0840 |
| West Wyalong Town | 1 | 1.5173 ± 0.0873 | 2.8809 ± 0.1470 | 1.7525 ± 0.0862 |
| | 2 | 1.5561 ± 0.0898 | 2.9539 ± 0.1547 | 1.8041 ± 0.0887 |
| | 3 | 1.4738 ± 0.0921 | 3.0980 ± 0.1553 | 1.7398 ± 0.0892 |
| | 4 | 1.7367 ± 0.0901 | 3.2627 ± 0.1509 | 1.8936 ± 0.0889 |
| | 26 | 1.4421 ± 0.0934 | 3.3931 ± 0.1649 | 1.8864 ± 0.0925 |
| | 27 | 1.6070 ± 0.0933 | 3.3251 ± 0.1596 | 1.8966 ± 0.0920 |
| | 36 | 1.6184 ± 0.0901 | 3.2703 ± 0.1550 | 1.9025 ± 0.0904 |
| | 37 | 1.5909 ± 0.0886 | 3.0229 ± 0.1470 | 1.7114 ± 0.0862 |
| | 38 | 1.7166 ± 0.0899 | 3.4003 ± 0.1507 | 1.9977 ± 0.0876 |
| | 39 | 1.5948 ± 0.0861 | 3.3090 ± 0.1435 | 1.8528 ± 0.0851 |
| | 40 | 1.5303 ± 0.0912 | 3.0810 ± 0.1470 | 1.8289 ± 0.0863 |
| | 41 | 1.9072 ± 0.0915 | 3.6836 ± 0.1549 | 2.0094 ± 0.0889 |
| | 42 | 1.6164 ± 0.0929 | 3.3587 ± 0.1593 | 1.8673 ± 0.0903 |
| | 43 | 1.5924 ± 0.0922 | 3.3428 ± 0.1553 | 1.9548 ± 0.0893 |
| | 46 | 1.8176 ± 0.0899 | 3.8434 ± 0.1549 | 2.0690 ± 0.0889 |
| | 47 | 1.6226 ± 0.0873 | 3.2580 ± 0.1435 | 1.8077 ± 0.0851 |
| | 48 | 1.4918 ± 0.0900 | 2.9198 ± 0.1597 | 1.7352 ± 0.0907 |
| | 49 | 1.8240 ± 0.0929 | 3.4287 ± 0.1644 | 1.9592 ± 0.0919 |
| | 56 | 1.8981 ± 0.0873 | 3.6750 ± 0.1435 | 1.9625 ± 0.0851 |
| | 57 | 1.6459 ± 0.0863 | 3.2388 ± 0.1404 | 1.9000 ± 0.0841 |
| | 58 | 1.4573 ± 0.0956 | 3.0301 ± 0.1649 | 1.8118 ± 0.0925 |
| | 59 | 1.5650 ± 0.0888 | 2.9294 ± 0.1509 | 1.8719 ± 0.0876 |
| | 60 | 1.7884 ± 0.0916 | 3.5006 ± 0.1550 | 1.9244 ± 0.0907 |
| | 61 | 1.6837 ± 0.0861 | 3.4732 ± 0.1403 | 1.7966 ± 0.0841 |
| | 62 | 1.5642 ± 0.0930 | 3.2691 ± 0.1594 | 1.7966 ± 0.0903 |
| | 63 | 1.5493 ± 0.0887 | 3.0509 ± 0.1471 | 1.6992 ± 0.0863 |
| | 64 | 1.6580 ± 0.0874 | 3.2221 ± 0.1436 | 1.8457 ± 0.0852 |
| | 65 | 1.7106 ± 0.0918 | 3.3503 ± 0.1551 | 1.9088 ± 0.0905 |
| | 66 | 1.4598 ± 0.0918 | 3.0184 ± 0.1551 | 1.6791 ± 0.0891 |
| West Yalgogrin | 32 | 1.5536 ± 0.0902 | 3.0276 ± 0.1509 | 1.7158 ± 0.0889 |
| | 33 | 1.7399 ± 0.0912 | 3.6052 ± 0.1548 | 1.9004 ± 0.0887 |
| | 34 | 1.7215 ± 0.0887 | 3.4155 ± 0.1471 | 1.9131 ± 0.0864 |
| | 35 | 1.6495 ± 0.0873 | 3.1724 ± 0.1435 | 1.9609 ± 0.0851 |
| Winters Lane | 7 | 1.7521 ± 0.0899 | 3.3618 ± 0.1507 | 1.9217 ± 0.0874 |
| | 8 | 1.6204 ± 0.0931 | 3.2780 ± 0.1645 | 1.8522 ± 0.0921 |
| | 9 | 1.6419 ± 0.0921 | 3.2329 ± 0.1552 | 1.8651 ± 0.0877 |
| | 10 | 1.7345 ± 0.0900 | 3.6195 ± 0.1508 | 1.9042 ± 0.0876 |
| | 11 | 1.8069 ± 0.0933 | 3.5382 ± 0.1595 | 1.9769 ± 0.0905 |
| | 12 | 1.7256 ± 0.0915 | 3.4520 ± 0.1550 | 1.9157 ± 0.0889 |
| | 13 | 1.4390 ± 0.0885 | 3.0361 ± 0.1470 | 1.7021 ± 0.0862 |
| | 14 | 1.6461 ± 0.0872 | 3.5379 ± 0.1469 | 1.8884 ± 0.0862 |
| | 15 | 1.5610 ± 0.0932 | 2.8792 ± 0.1511 | 1.7900 ± 0.0879 |
| | 16 | 1.7121 ± 0.0885 | 3.2626 ± 0.1470 | 1.8285 ± 0.0861 |
| | 17 | 1.5071 ± 0.0874 | 2.9860 ± 0.1436 | 1.8339 ± 0.0851 |
| | 18 | 1.4677 ± 0.0884 | 3.1774 ± 0.1507 | 1.7419 ± 0.0873 |
| | 19 | 1.8276 ± 0.0970 | 3.7747 ± 0.1700 | 2.1256 ± 0.0940 |
| | 20 | 1.5983 ± 0.0921 | 3.4461 ± 0.1553 | 1.8543 ± 0.0893 |
| | 21 | 1.6150 ± 0.0872 | 3.1200 ± 0.1434 | 1.8201 ± 0.0850 |
| | 22 | 1.6516 ± 0.0916 | 3.4471 ± 0.1550 | 1.9620 ± 0.0889 |
| | 23 | 1.6538 ± 0.0902 | 3.2971 ± 0.1510 | 1.9009 ± 0.0876 |
| | 24 | 1.7075 ± 0.0863 | 3.4196 ± 0.1404 | 1.8378 ± 0.0841 |
| | 50 | 1.6126 ± 0.0887 | 3.2334 ± 0.1471 | 1.7976 ± 0.0864 |
| | 51 | 1.7056 ± 0.0889 | 3.4068 ± 0.1472 | 1.8361 ± 0.0866 |
| | 52 | 1.7333 ± 0.0873 | 3.4120 ± 0.1435 | 1.8914 ± 0.0851 |
| | 53 | 1.5601 ± 0.0977 | 3.2828 ± 0.1705 | 1.8226 ± 0.0944 |
| | 54 | 1.6371 ± 0.0873 | 3.1089 ± 0.1435 | 1.8927 ± 0.0850 |
| | 55 | 1.7183 ± 0.0885 | 3.4792 ± 0.1470 | 1.8303 ± 0.0862 |

Table S3 Best Linear Unbiased Estimates (± standard errors) for three biomass estimates at the Drummartin *E. polybractea* progeny trial

| Provenance | Family | 3-year sapling | 6-year sapling | Coppice |
|---|---|---|---|---|
| Charcoal Tank Rd | 25 | 1.9339 ± 0.1005 | 3.2720 ± 0.1582 | 2.3132 ± 0.0850 |
| Kerrs Lane | 5 | 1.7258 ± 0.0983 | 2.8390 ± 0.1507 | 2.0592 ± 0.0849 |
|  | 6 | 1.8025 ± 0.0984 | 3.0850 ± 0.1508 | 2.1102 ± 0.0807 |
| Talimba Rd East | 44 | 1.8822 ± 0.0984 | 3.1089 ± 0.1543 | 2.1915 ± 0.0834 |
| Talimba Rd West | 45 | 1.7797 ± 0.1003 | 3.0419 ± 0.1623 | 2.2234 ± 0.0862 |
| West Mid-West Hwy | 28 | 1.8170 ± 0.0962 | 2.9256 ± 0.1474 | 2.1764 ± 0.0805 |
|  | 29 | 1.7521 ± 0.1029 | 2.8062 ± 0.1582 | 2.1958 ± 0.0848 |
|  | 30 | 1.9213 ± 0.1005 | 3.1064 ± 0.1582 | 2.2533 ± 0.0833 |
|  | 31 | 1.9060 ± 0.0983 | 3.1350 ± 0.1508 | 2.3565 ± 0.0818 |
| West Wyalong Town | 1 | 1.7318 ± 0.1030 | 2.8469 ± 0.1624 | 2.1763 ± 0.0866 |
|  | 2 | 1.5361 ± 0.1145 | 2.5225 ± 0.1773 | 2.1524 ± 0.0926 |
|  | 3 | 1.9449 ± 0.0963 | 3.2835 ± 0.1474 | 2.2965 ± 0.0807 |
|  | 4 | 1.9447 ± 0.0944 | 3.1758 ± 0.1474 | 2.2770 ± 0.0806 |
|  | 26 | 2.0437 ± 0.0962 | 3.2675 ± 0.1474 | 2.2598 ± 0.0792 |
|  | 27 | 1.8160 ± 0.0943 | 3.0195 ± 0.1507 | 2.2504 ± 0.0817 |
|  | 36 | 1.9701 ± 0.0983 | 3.0966 ± 0.1508 | 2.2457 ± 0.0819 |
|  | 37 | 1.8454 ± 0.1031 | 2.9442 ± 0.1624 | 2.1906 ± 0.0867 |
|  | 38 | 1.8597 ± 0.0925 | 3.1171 ± 0.1474 | 2.2626 ± 0.0803 |
|  | 39 | 1.7412 ± 0.0983 | 2.9590 ± 0.1507 | 2.1838 ± 0.0818 |
|  | 40 | 1.7464 ± 0.0984 | 3.0615 ± 0.1669 | 2.2868 ± 0.0885 |
|  | 41 | 1.9800 ± 0.0962 | 3.3609 ± 0.1507 | 2.5169 ± 0.0832 |
|  | 42 | 1.8389 ± 0.1051 | 3.2101 ± 0.1668 | 2.1714 ± 0.0880 |
|  | 43 | 1.9242 ± 0.0983 | 3.2548 ± 0.1581 | 2.3124 ± 0.0846 |
|  | 46 | 2.0562 ± 0.0964 | 3.1336 ± 0.1474 | 2.3076 ± 0.0807 |
|  | 47 | 1.8519 ± 0.1005 | 3.0017 ± 0.1507 | 2.2090 ± 0.0819 |
|  | 48 | 2.0426 ± 0.0964 | 3.3888 ± 0.1508 | 2.3985 ± 0.0819 |
|  | 49 | 1.9748 ± 0.0985 | 3.1974 ± 0.1508 | 2.3451 ± 0.0821 |
|  | 56 | 2.0603 ± 0.1280 | 3.4413 ± 0.1980 | 2.3014 ± 0.1023 |
|  | 57 | 1.9375 ± 0.1011 | 3.0666 ± 0.1545 | 2.3363 ± 0.0837 |
|  | 58 | 1.7514 ± 0.1005 | 3.0576 ± 0.1581 | 2.2190 ± 0.0847 |
|  | 59 | 1.7115 ± 0.1006 | 2.7729 ± 0.1544 | 2.1775 ± 0.0833 |
|  | 60 | 1.7870 ± 0.0962 | 2.8594 ± 0.1474 | 2.1641 ± 0.0804 |
|  | 61 | 1.9484 ± 0.1003 | 3.0821 ± 0.1581 | 2.1553 ± 0.0887 |
|  | 62 | 1.7310 ± 0.0983 | 2.8431 ± 0.1544 | 2.0549 ± 0.0834 |
|  | 63 | 1.7378 ± 0.1056 | 3.0040 ± 0.1719 | 2.2019 ± 0.0906 |
|  | 64 | 2.0206 ± 0.0944 | 3.2068 ± 0.1443 | 2.3360 ± 0.0792 |
|  | 65 | 1.9893 ± 0.1027 | 3.0259 ± 0.1581 | 2.2043 ± 0.0847 |
|  | 66 | 1.6321 ± 0.1027 | 2.9888 ± 0.1717 | 2.0073 ± 0.0900 |
| West Yalgogrin | 32 | 1.9592 ± 0.1028 | 3.1903 ± 0.1581 | 2.0670 ± 0.0848 |
|  | 33 | 2.3114 ± 0.1110 | 3.6439 ± 0.1718 | 2.2971 ± 0.0904 |
|  | 34 | 1.8718 ± 0.1054 | 3.1504 ± 0.1623 | 2.1694 ± 0.0868 |
|  | 35 | 1.8680 ± 0.0983 | 3.1435 ± 0.1543 | 2.2242 ± 0.0833 |
| Winters Lane | 7 | 1.9598 ± 0.1079 | 3.2954 ± 0.1668 | 2.4231 ± 0.0881 |
|  | 8 | 1.9323 ± 0.0984 | 3.1202 ± 0.1543 | 2.2054 ± 0.0834 |
|  | 9 | 1.9731 ± 0.0983 | 3.3150 ± 0.1582 | 2.3496 ± 0.0849 |
|  | 10 | 1.9411 ± 0.0962 | 3.1413 ± 0.1474 | 2.2192 ± 0.0804 |
|  | 11 | 1.9897 ± 0.0944 | 3.3237 ± 0.1507 | 2.3576 ± 0.0818 |
|  | 12 | 2.1065 ± 0.1080 | 3.3252 ± 0.1668 | 2.2479 ± 0.0908 |
|  | 13 | 1.8909 ± 0.1004 | 3.1765 ± 0.1543 | 2.2916 ± 0.0832 |
|  | 14 | 1.8763 ± 0.0944 | 3.1107 ± 0.1474 | 2.2628 ± 0.0817 |
|  | 15 | 1.8318 ± 0.0963 | 3.0841 ± 0.1474 | 2.3998 ± 0.0807 |
|  | 16 | 1.9914 ± 0.1054 | 3.1346 ± 0.1624 | 2.1888 ± 0.0865 |
|  | 17 | 1.8536 ± 0.0943 | 3.1030 ± 0.1507 | 2.2321 ± 0.0817 |
|  | 18 | 2.0561 ± 0.0963 | 3.4396 ± 0.1507 | 2.3077 ± 0.0818 |
|  | 19 | 2.0716 ± 0.1027 | 3.3006 ± 0.1581 | 2.3916 ± 0.0847 |
|  | 20 | 1.9598 ± 0.1027 | 3.3809 ± 0.1668 | 2.2863 ± 0.0881 |
|  | 21 | 1.8609 ± 0.0982 | 3.1063 ± 0.1543 | 2.1847 ± 0.0831 |
|  | 22 | 1.8654 ± 0.0963 | 3.1247 ± 0.1507 | 2.4055 ± 0.0833 |
|  | 23 | 1.9282 ± 0.0945 | 3.1445 ± 0.1443 | 2.2347 ± 0.0808 |
|  | 24 | 2.0370 ± 0.1004 | 3.4031 ± 0.1543 | 2.3616 ± 0.0831 |
|  | 50 | 1.7933 ± 0.1056 | 3.1635 ± 0.1719 | 2.2644 ± 0.0904 |
|  | 51 | 1.9678 ± 0.1055 | 3.0188 ± 0.1624 | 2.3390 ± 0.0867 |
|  | 52 | 1.9468 ± 0.1004 | 3.1138 ± 0.1543 | 2.2810 ± 0.0833 |
|  | 53 | 2.1663 ± 0.1004 | 3.5431 ± 0.1581 | 2.4250 ± 0.0848 |
|  | 54 | 1.7718 ± 0.1080 | 3.0931 ± 0.1668 | 2.3728 ± 0.0884 |
|  | 55 | 1.9014 ± 0.0982 | 3.2003 ± 0.1623 | 2.3164 ± 0.0880 |

Table S4 Best Linear Unbiased Estimates (± standard errors) for three biomass estimates at the Condobolin *E. polybractea* progeny trial

| Provenance | Family | 3-year sapling | 6-year sapling | Coppice |
|---|---|---|---|---|
| Charcoal Tank Rd | 25 | 1.9591 ± 0.0864 | 3.0739 ± 0.1231 | 1.8346 ± 0.0645 |
| Kerrs Lane | 5 | 1.8820 ± 0.0849 | 3.0382 ± 0.1178 | 1.6973 ± 0.0635 |
| | 6 | 1.8562 ± 0.0849 | 3.0653 ± 0.1261 | 1.7750 ± 0.0656 |
| Talimba Rd East | 44 | 2.1082 ± 0.0864 | 3.2972 ± 0.1203 | 1.8486 ± 0.0645 |
| Talimba Rd West | 45 | 2.1897 ± 0.0935 | 3.2921 ± 0.1366 | 1.9220 ± 0.0713 |
| West Mid-West Hwy | 28 | 2.1275 ± 0.0836 | 3.3408 ± 0.1178 | 1.8806 ± 0.0625 |
| | 29 | 2.0336 ± 0.0864 | 2.9906 ± 0.1203 | 1.7472 ± 0.0635 |
| | 30 | 1.8376 ± 0.0850 | 2.8768 ± 0.1178 | 1.7312 ± 0.0635 |
| | 31 | 2.2693 ± 0.0864 | 3.3671 ± 0.1203 | 1.8997 ± 0.0645 |
| West Wyalong Town | 1 | 1.8883 ± 0.0914 | 2.7853 ± 0.1293 | 1.7774 ± 0.0669 |
| | 2 | 2.0058 ± 0.0881 | 2.9527 ± 0.1261 | 1.7995 ± 0.0669 |
| | 3 | 2.1975 ± 0.0917 | 3.2545 ± 0.1328 | 1.8794 ± 0.0683 |
| | 4 | 1.9299 ± 0.0897 | 3.0949 ± 0.1328 | 1.8980 ± 0.0696 |
| | 26 | 2.0182 ± 0.0864 | 3.0432 ± 0.1203 | 1.7336 ± 0.0635 |
| | 27 | 2.2661 ± 0.0902 | 3.4233 ± 0.1294 | 1.9338 ± 0.0670 |
| | 36 | 2.1645 ± 0.0881 | 3.1189 ± 0.1231 | 1.7628 ± 0.0646 |
| | 37 | 2.0899 ± 0.0879 | 3.1787 ± 0.1261 | 1.7773 ± 0.0656 |
| | 38 | 2.0575 ± 0.0914 | 3.1505 ± 0.1293 | 1.8052 ± 0.0682 |
| | 39 | 1.9668 ± 0.0849 | 3.1518 ± 0.1203 | 1.8833 ± 0.0635 |
| | 40 | 2.0403 ± 0.0879 | 3.1042 ± 0.1231 | 1.7339 ± 0.0645 |
| | 41 | 2.3330 ± 0.0836 | 3.3713 ± 0.1178 | 1.9578 ± 0.0635 |
| | 42 | 1.9537 ± 0.0881 | 3.1384 ± 0.1231 | 1.8012 ± 0.0646 |
| | 43 | 2.2061 ± 0.0850 | 3.3282 ± 0.1203 | 1.9062 ± 0.0635 |
| | 46 | 2.6214 ± 0.0864 | 3.8335 ± 0.1203 | 2.0495 ± 0.0635 |
| | 47 | 2.1517 ± 0.0836 | 3.2463 ± 0.1153 | 1.8002 ± 0.0635 |
| | 48 | 2.1430 ± 0.0895 | 3.1807 ± 0.1261 | 1.8997 ± 0.0656 |
| | 49 | 2.1544 ± 0.0917 | 3.3085 ± 0.1328 | 1.8346 ± 0.0682 |
| | 56 | 2.0784 ± 0.0849 | 3.2988 ± 0.1203 | 1.8191 ± 0.0635 |
| | 57 | 2.1403 ± 0.0864 | 3.3907 ± 0.1231 | 1.8205 ± 0.0645 |
| | 58 | 1.7361 ± 0.0917 | 2.7374 ± 0.1328 | 1.6536 ± 0.0713 |
| | 59 | 1.9186 ± 0.0849 | 2.8248 ± 0.1178 | 1.6924 ± 0.0625 |
| | 60 | 2.2691 ± 0.0849 | 3.3555 ± 0.1178 | 1.9031 ± 0.0625 |
| | 61 | 2.1132 ± 0.0938 | 3.3594 ± 0.1328 | 1.6681 ± 0.0697 |
| | 62 | 1.9905 ± 0.0864 | 3.1909 ± 0.1203 | 1.7595 ± 0.0645 |
| | 63 | 1.9468 ± 0.0864 | 3.2410 ± 0.1261 | 1.7367 ± 0.0656 |
| | 64 | 2.2797 ± 0.0881 | 3.4203 ± 0.1261 | 1.9180 ± 0.0657 |
| | 65 | 2.3341 ± 0.0836 | 3.5006 ± 0.1178 | 1.8306 ± 0.0625 |
| | 66 | 1.6893 ± 0.0955 | 2.9358 ± 0.1452 | 1.6389 ± 0.0729 |
| West Yalgogrin | 32 | 1.9764 ± 0.0864 | 3.2298 ± 0.1261 | 1.8088 ± 0.0668 |
| | 33 | 2.0313 ± 0.0850 | 3.3195 ± 0.1203 | 1.8163 ± 0.0635 |
| | 34 | 2.1727 ± 0.0879 | 3.3365 ± 0.1231 | 1.9060 ± 0.0645 |
| | 35 | 2.0900 ± 0.0864 | 3.2428 ± 0.1203 | 1.7772 ± 0.0635 |
| Winters Lane | 7 | 2.1224 ± 0.0914 | 3.2684 ± 0.1328 | 1.9016 ± 0.0682 |
| | 8 | 1.9454 ± 0.0912 | 3.0278 ± 0.1293 | 1.7062 ± 0.0682 |
| | 9 | 1.9499 ± 0.0864 | 2.9385 ± 0.1203 | 1.7491 ± 0.0635 |
| | 10 | 2.1078 ± 0.0864 | 3.3287 ± 0.1231 | 1.6759 ± 0.0635 |
| | 11 | 2.3260 ± 0.0849 | 3.4501 ± 0.1203 | 1.9207 ± 0.0635 |
| | 12 | 1.9902 ± 0.0935 | 3.0478 ± 0.1328 | 1.7760 ± 0.0696 |
| | 13 | 2.0063 ± 0.0985 | 3.2113 ± 0.1408 | 1.7858 ± 0.0714 |
| | 14 | 2.1587 ± 0.0849 | 3.1589 ± 0.1203 | 1.8148 ± 0.0656 |
| | 15 | 2.2804 ± 0.0864 | 3.1860 ± 0.1203 | 1.9048 ± 0.0635 |
| | 16 | 1.9587 ± 0.0881 | 2.9997 ± 0.1261 | 1.7974 ± 0.0670 |
| | 17 | 2.0615 ± 0.0914 | 3.0007 ± 0.1328 | 1.8653 ± 0.0682 |
| | 18 | 1.9759 ± 0.0850 | 3.1071 ± 0.1178 | 1.7204 ± 0.0635 |
| | 19 | 2.0784 ± 0.0879 | 3.3563 ± 0.1328 | 1.8619 ± 0.0712 |
| | 20 | 2.1546 ± 0.0864 | 3.3111 ± 0.1231 | 1.8441 ± 0.0645 |
| | 21 | 2.1139 ± 0.0864 | 3.2006 ± 0.1231 | 1.7263 ± 0.0645 |
| | 22 | 2.2015 ± 0.0866 | 3.3653 ± 0.1261 | 1.9164 ± 0.0657 |
| | 23 | 1.9902 ± 0.0864 | 3.1050 ± 0.1203 | 1.8294 ± 0.0645 |
| | 24 | 2.2992 ± 0.0850 | 3.5578 ± 0.1178 | 1.8964 ± 0.0625 |
| | 50 | 1.9098 ± 0.0866 | 2.8342 ± 0.1204 | 1.6773 ± 0.0635 |
| | 51 | 2.2416 ± 0.0915 | 3.3764 ± 0.1293 | 1.8879 ± 0.0682 |
| | 52 | 2.0801 ± 0.0879 | 3.3740 ± 0.1261 | 1.9551 ± 0.0656 |
| | 53 | 2.2433 ± 0.0879 | 3.3679 ± 0.1231 | 1.9481 ± 0.0656 |
| | 54 | 1.8995 ± 0.0879 | 2.8248 ± 0.1261 | 1.8082 ± 0.0656 |
| | 55 | 2.0018 ± 0.0879 | 3.3077 ± 0.1261 | 1.8760 ± 0.0656 |

Table S5 Mean Net Breeding Values (NBV) ± standard errors from the three biomass assessments; 2013 (3-year sapling), 2016 (6-year sapling) and 2020 (coppice) assessments for orchard thinning scenario at the Newdegate *E. polybractea* progeny trial and the number of individuals selected from each family at each assessment.

| Population | Family | Mean NBV 2013 | No. selected 2013 | Mean NBV 2016 | No. selected 2016 | Mean NBV 2020 | No. selected 2020 |
|---|---|---|---|---|---|---|---|
| Charcoal Tank Rd | 25 | 1.685 ± 0.132 | 4 | 3.267 ± 0.250 | 3 | 1.858 ± 0.109 | 4 |
| Kerrs Lane | 5 | 1.492 ± 0.122 | | 3.186 ± 0.232 | 1 | 1.828 ± 0.097 | |
| | 6 | 1.540 ± 0.124 | | 3.216 ± 0.237 | 3 | 1.849 ± 0.098 | 4 |
| Talimba Rd East | 44 | 1.590 ± 0.128 | 2 | 3.341 ± 0.242 | 4 | 1.818 ± 0.105 | |
| Talimba Rd West | 45 | 1.725 ± 0.128 | 4 | 3.601 ± 0.246 | 4 | 1.966 ± 0.106 | 4 |
| West Mid-West Hwy | 28 | 1.571 ± 0.118 | 1 | 3.087 ± 0.226 | | 1.765 ± 0.091 | |
| | 29 | 1.538 ± 0.118 | | 2.887 ± 0.227 | | 1.724 ± 0.091 | |
| | 30 | 1.538 ± 0.118 | | 3.102 ± 0.227 | | 1.766 ± 0.091 | |
| | 31 | 1.735 ± 0.118 | 4 | 3.172 ± 0.226 | 1 | 1.795 ± 0.091 | |
| West Yalgogrin | 32 | 1.601 ± 0.118 | 2 | 3.166 ± 0.228 | 2 | 1.836 ± 0.091 | 2 |
| | 33 | 1.720 ± 0.119 | 4 | 3.446 ± 0.228 | 4 | 1.880 ± 0.091 | 4 |
| | 34 | 1.691 ± 0.118 | 4 | 3.360 ± 0.227 | 4 | 1.903 ± 0.091 | 4 |
| | 35 | 1.642 ± 0.118 | 4 | 3.221 ± 0.227 | 3 | 1.878 ± 0.091 | 4 |
| Winters Lane | 7 | 1.693 ± 0.115 | 4 | 3.361 ± 0.224 | 4 | 1.896 ± 0.087 | 4 |
| | 8 | 1.613 ± 0.115 | 3 | 3.244 ± 0.224 | 3 | 1.828 ± 0.087 | 1 |
| | 9 | 1.631 ± 0.115 | 3 | 3.190 ± 0.223 | 2 | 1.842 ± 0.087 | 2 |
| | 10 | 1.683 ± 0.115 | 4 | 3.492 ± 0.223 | 4 | 1.825 ± 0.087 | |
| | 11 | 1.757 ± 0.115 | 4 | 3.503 ± 0.223 | 4 | 1.917 ± 0.087 | 4 |
| | 12 | 1.679 ± 0.116 | 4 | 3.322 ± 0.224 | 4 | 1.860 ± 0.087 | 4 |
| | 13 | 1.551 ± 0.115 | | 3.188 ± 0.224 | 2 | 1.816 ± 0.087 | |
| | 14 | 1.655 ± 0.115 | 4 | 3.404 ± 0.223 | 4 | 1.865 ± 0.087 | 4 |
| | 15 | 1.643 ± 0.115 | 3 | 3.125 ± 0.223 | | 1.872 ± 0.087 | 4 |
| | 16 | 1.653 ± 0.115 | 4 | 3.230 ± 0.223 | 4 | 1.847 ± 0.087 | 4 |
| | 17 | 1.577 ± 0.115 | 1 | 3.095 ± 0.223 | | 1.864 ± 0.087 | 4 |
| | 18 | 1.582 ± 0.115 | 2 | 3.239 ± 0.223 | 4 | 1.808 ± 0.087 | |
| | 19 | 1.731 ± 0.116 | 4 | 3.551 ± 0.225 | 4 | 1.924 ± 0.087 | 4 |
| | 20 | 1.649 ± 0.115 | 4 | 3.420 ± 0.223 | 4 | 1.874 ± 0.087 | 4 |
| | 21 | 1.632 ± 0.115 | 4 | 3.237 ± 0.222 | 3 | 1.827 ± 0.087 | 1 |
| | 22 | 1.662 ± 0.115 | 4 | 3.434 ± 0.224 | 4 | 1.913 ± 0.087 | 4 |
| | 23 | 1.634 ± 0.115 | 4 | 3.280 ± 0.223 | 3 | 1.874 ± 0.087 | 4 |
| | 24 | 1.718 ± 0.115 | 4 | 3.484 ± 0.222 | 4 | 1.879 ± 0.087 | 4 |
| | 50 | 1.592 ± 0.115 | 1 | 3.169 ± 0.223 | 2 | 1.806 ± 0.087 | |
| | 51 | 1.700 ± 0.115 | 4 | 3.413 ± 0.223 | 4 | 1.874 ± 0.087 | 4 |
| | 52 | 1.685 ± 0.115 | 4 | 3.433 ± 0.223 | 4 | 1.908 ± 0.087 | 4 |
| | 53 | 1.679 ± 0.116 | 4 | 3.372 ± 0.224 | 4 | 1.891 ± 0.087 | 4 |
| | 54 | 1.598 ± 0.115 | 2 | 3.097 ± 0.223 | | 1.865 ± 0.087 | 4 |
| | 55 | 1.659 ± 0.115 | 4 | 3.441 ± 0.223 | 4 | 1.876 ± 0.087 | 4 |
| West Wyalong Town | 1 | 1.534 ± 0.115 | | 2.946 ± 0.223 | | 1.817 ± 0.087 | |
| | 2 | 1.544 ± 0.115 | | 3.040 ± 0.223 | | 1.841 ± 0.087 | |
| | 3 | 1.603 ± 0.115 | 2 | 3.207 ± 0.224 | 2 | 1.852 ± 0.087 | 3 |
| | 4 | 1.649 ± 0.115 | 4 | 3.224 ± 0.223 | 3 | 1.885 ± 0.087 | 4 |
| | 26 | 1.574 ± 0.115 | 2 | 3.254 ± 0.224 | 4 | 1.835 ± 0.087 | 3 |
| | 27 | 1.641 ± 0.115 | 4 | 3.357 ± 0.224 | 4 | 1.896 ± 0.087 | 4 |
| | 36 | 1.654 ± 0.115 | 4 | 3.234 ± 0.223 | 3 | 1.851 ± 0.087 | 4 |
| | 37 | 1.614 ± 0.115 | 4 | 3.142 ± 0.223 | | 1.810 ± 0.087 | |
| | 38 | 1.648 ± 0.115 | 4 | 3.289 ± 0.223 | 4 | 1.882 ± 0.087 | 4 |
| | 39 | 1.571 ± 0.115 | 1 | 3.252 ± 0.222 | 3 | 1.872 ± 0.086 | 4 |
| | 40 | 1.569 ± 0.115 | 1 | 3.147 ± 0.223 | 2 | 1.821 ± 0.087 | |
| | 41 | 1.788 ± 0.115 | 4 | 3.485 ± 0.223 | 4 | 1.929 ± 0.087 | 4 |
| | 42 | 1.594 ± 0.115 | 2 | 3.272 ± 0.223 | 4 | 1.855 ± 0.087 | 4 |
| | 43 | 1.651 ± 0.115 | 4 | 3.347 ± 0.223 | 4 | 1.906 ± 0.087 | 4 |
| | 46 | 1.820 ± 0.115 | 4 | 3.726 ± 0.223 | 4 | 1.970 ± 0.087 | 4 |
| | 47 | 1.640 ± 0.115 | 4 | 3.275 ± 0.222 | 4 | 1.843 ± 0.086 | 3 |
| | 48 | 1.613 ± 0.115 | 3 | 3.098 ± 0.224 | | 1.857 ± 0.087 | 4 |
| | 49 | 1.735 ± 0.115 | 4 | 3.366 ± 0.224 | 4 | 1.881 ± 0.087 | 4 |
| | 56 | 1.743 ± 0.115 | 4 | 3.471 ± 0.222 | 4 | 1.878 ± 0.086 | 4 |
| | 57 | 1.647 ± 0.115 | 4 | 3.296 ± 0.222 | 4 | 1.863 ± 0.086 | 4 |
| | 58 | 1.491 ± 0.115 | | 3.013 ± 0.224 | | 1.804 ± 0.087 | |
| | 59 | 1.548 ± 0.115 | | 2.978 ± 0.222 | | 1.818 ± 0.086 | |
| | 60 | 1.708 ± 0.115 | 4 | 3.404 ± 0.223 | 4 | 1.893 ± 0.087 | 4 |
| | 61 | 1.663 ± 0.115 | 4 | 3.406 ± 0.222 | 4 | 1.806 ± 0.087 | |
| | 62 | 1.572 ± 0.115 | 2 | 3.270 ± 0.223 | 4 | 1.829 ± 0.087 | |
| | 63 | 1.558 ± 0.115 | | 3.177 ± 0.223 | 2 | 1.798 ± 0.087 | |
| | 64 | 1.701 ± 0.115 | 4 | 3.327 ± 0.222 | 4 | 1.886 ± 0.087 | 4 |
| | 65 | 1.715 ± 0.115 | 4 | 3.387 ± 0.223 | 4 | 1.865 ± 0.087 | 4 |
| | 66 | 1.461 ± 0.115 | | 3.061 ± 0.224 | | 1.767 ± 0.087 | |
| Totals | | 1.636 ± 0.116 | 185 | 3.274 ± 0.225 | 183 | 1.854 ± 0.088 | 171 |

Table S6 Mean Net Breeding Values (NBV) ± standard errors from the three biomass assessments; 2013 (3-year sapling), 2016 (6-year sapling) and 2020 (coppice) assessments for orchard thinning scenario at the Condobolin *E. polybractea* progeny trial and the number of individuals selected from each family at each assessment.

| Population | Family | Mean NBV 2013 | No. selected 2013 | Mean NBV 2016 | No. selected 2016 | Mean NBV 2020 | No. selected 2020 |
|---|---|---|---|---|---|---|---|
| Charcoal Tank Rd | 25 | 1.960 ± 0.170 | 1 | 3.074 ± 0.230 | 1 | 1.834 ± 0.103 | 6 |
| Kerrs Lane | 5 | 1.863 ± 0.167 | | 3.039 ± 0.223 | | 1.714 ± 0.101 | |
| | 6 | 1.883 ± 0.167 | | 3.065 ± 0.225 | 1 | 1.760 ± 0.101 | 1 |
| Talimba Rd East | 44 | 2.115 ± 0.170 | 6 | 3.297 ± 0.229 | 6 | 1.848 ± 0.103 | 6 |
| Talimba Rd West | 45 | 2.184 ± 0.172 | 6 | 3.292 ± 0.235 | 6 | 1.921 ± 0.107 | 6 |
| West Mid-West Hwy | 28 | 2.093 ± 0.166 | 6 | 3.237 ± 0.220 | 6 | 1.843 ± 0.099 | 6 |
| | 29 | 2.023 ± 0.166 | 4 | 2.991 ± 0.220 | | 1.758 ± 0.100 | |
| | 30 | 1.908 ± 0.166 | | 3.053 ± 0.220 | | 1.782 ± 0.100 | |
| | 31 | 2.258 ± 0.166 | 6 | 3.291 ± 0.220 | 6 | 1.871 ± 0.100 | 6 |
| West Yalgogrin | 32 | 1.983 ± 0.166 | 4 | 3.192 ± 0.221 | 6 | 1.792 ± 0.100 | 4 |
| | 33 | 2.087 ± 0.166 | 6 | 3.372 ± 0.221 | 6 | 1.826 ± 0.100 | 6 |
| | 34 | 2.136 ± 0.167 | 6 | 3.336 ± 0.221 | 6 | 1.879 ± 0.100 | 6 |
| | 35 | 2.056 ± 0.166 | 4 | 3.226 ± 0.220 | 6 | 1.809 ± 0.100 | 6 |
| Winters Lane | 7 | 2.129 ± 0.166 | 6 | 3.244 ± 0.219 | 6 | 1.878 ± 0.099 | 6 |
| | 8 | 1.981 ± 0.166 | 3 | 3.090 ± 0.220 | 2 | 1.754 ± 0.100 | |
| | 9 | 1.998 ± 0.165 | 2 | 3.024 ± 0.218 | 1 | 1.779 ± 0.099 | 2 |
| | 10 | 2.113 ± 0.165 | 5 | 3.337 ± 0.218 | 6 | 1.737 ± 0.099 | |
| | 11 | 2.282 ± 0.165 | 6 | 3.392 ± 0.218 | 6 | 1.902 ± 0.099 | 6 |
| | 12 | 2.055 ± 0.166 | 5 | 3.140 ± 0.220 | 5 | 1.805 ± 0.100 | 4 |
| | 13 | 1.988 ± 0.167 | 2 | 3.127 ± 0.220 | 2 | 1.775 ± 0.100 | 2 |
| | 14 | 2.125 ± 0.165 | 6 | 3.224 ± 0.218 | 6 | 1.821 ± 0.099 | 6 |
| | 15 | 2.195 ± 0.165 | 6 | 3.092 ± 0.218 | 1 | 1.864 ± 0.099 | 6 |
| | 16 | 2.013 ± 0.166 | 4 | 3.071 ± 0.219 | 1 | 1.802 ± 0.099 | 4 |
| | 17 | 2.033 ± 0.166 | 3 | 3.000 ± 0.219 | | 1.841 ± 0.099 | 6 |
| | 18 | 1.996 ± 0.165 | 2 | 3.117 ± 0.218 | 2 | 1.743 ± 0.099 | |
| | 19 | 2.124 ± 0.166 | 6 | 3.376 ± 0.220 | 6 | 1.884 ± 0.100 | 6 |
| | 20 | 2.127 ± 0.165 | 6 | 3.293 ± 0.219 | 6 | 1.841 ± 0.099 | 6 |
| | 21 | 2.092 ± 0.165 | 6 | 3.153 ± 0.218 | 6 | 1.760 ± 0.099 | |
| | 22 | 2.153 ± 0.165 | 6 | 3.321 ± 0.219 | 6 | 1.896 ± 0.099 | 6 |
| | 23 | 2.016 ± 0.165 | 3 | 3.138 ± 0.218 | 6 | 1.834 ± 0.099 | 6 |
| | 24 | 2.247 ± 0.165 | 6 | 3.428 ± 0.218 | 6 | 1.867 ± 0.098 | 6 |
| | 50 | 1.946 ± 0.165 | 3 | 2.971 ± 0.218 | | 1.724 ± 0.099 | |
| | 51 | 2.199 ± 0.166 | 6 | 3.314 ± 0.219 | 6 | 1.858 ± 0.099 | 5 |
| | 52 | 2.101 ± 0.165 | 6 | 3.324 ± 0.219 | 6 | 1.910 ± 0.099 | 6 |
| | 53 | 2.208 ± 0.166 | 6 | 3.293 ± 0.219 | 6 | 1.895 ± 0.099 | 6 |
| | 54 | 1.940 ± 0.166 | 2 | 2.933 ± 0.219 | | 1.819 ± 0.099 | 4 |
| | 55 | 2.031 ± 0.165 | 5 | 3.303 ± 0.219 | 6 | 1.854 ± 0.099 | 6 |
| West Wyalong Town | 1 | 1.922 ± 0.166 | 1 | 2.879 ± 0.219 | | 1.773 ± 0.099 | 2 |
| | 2 | 1.980 ± 0.166 | 2 | 2.993 ± 0.219 | | 1.800 ± 0.099 | 3 |
| | 3 | 2.147 ± 0.166 | 6 | 3.198 ± 0.220 | 6 | 1.839 ± 0.099 | 6 |
| | 4 | 2.010 ± 0.166 | 3 | 3.146 ± 0.219 | 5 | 1.869 ± 0.100 | 6 |
| | 26 | 2.040 ± 0.165 | 3 | 3.136 ± 0.219 | 2 | 1.769 ± 0.099 | 2 |
| | 27 | 2.200 ± 0.166 | 6 | 3.343 ± 0.219 | 6 | 1.891 ± 0.099 | 6 |
| | 36 | 2.152 ± 0.165 | 6 | 3.159 ± 0.219 | 6 | 1.792 ± 0.099 | 3 |
| | 37 | 2.084 ± 0.165 | 6 | 3.134 ± 0.219 | 3 | 1.769 ± 0.099 | 2 |
| | 38 | 2.081 ± 0.166 | 5 | 3.202 ± 0.219 | 6 | 1.832 ± 0.099 | 6 |
| | 39 | 1.979 ± 0.165 | 3 | 3.182 ± 0.218 | 5 | 1.856 ± 0.099 | 6 |
| | 40 | 2.027 ± 0.165 | 5 | 3.108 ± 0.218 | 2 | 1.759 ± 0.099 | |
| | 41 | 2.321 ± 0.165 | 6 | 3.389 ± 0.218 | 6 | 1.926 ± 0.099 | 6 |
| | 42 | 1.992 ± 0.166 | 2 | 3.186 ± 0.219 | 6 | 1.810 ± 0.099 | 5 |
| | 43 | 2.175 ± 0.165 | 6 | 3.302 ± 0.218 | 6 | 1.890 ± 0.099 | 6 |
| | 46 | 2.514 ± 0.165 | 6 | 3.701 ± 0.218 | 6 | 1.994 ± 0.099 | 6 |
| | 47 | 2.135 ± 0.165 | 6 | 3.232 ± 0.217 | 6 | 1.802 ± 0.099 | 4 |
| | 48 | 2.129 ± 0.166 | 6 | 3.114 ± 0.219 | 3 | 1.853 ± 0.099 | 6 |
| | 49 | 2.190 ± 0.166 | 6 | 3.302 ± 0.220 | 6 | 1.842 ± 0.099 | 6 |
| | 56 | 2.146 ± 0.165 | 6 | 3.355 ± 0.218 | 6 | 1.833 ± 0.099 | 5 |
| | 57 | 2.131 ± 0.165 | 6 | 3.299 ± 0.218 | 6 | 1.824 ± 0.099 | 6 |
| | 58 | 1.808 ± 0.166 | | 2.898 ± 0.220 | | 1.720 ± 0.100 | |
| | 59 | 1.938 ± 0.165 | | 2.907 ± 0.218 | | 1.738 ± 0.099 | |
| | 60 | 2.230 ± 0.165 | 6 | 3.342 ± 0.218 | 6 | 1.879 ± 0.099 | 6 |
| | 61 | 2.115 ± 0.166 | 6 | 3.344 ± 0.219 | 6 | 1.725 ± 0.099 | |
| | 62 | 1.995 ± 0.165 | 3 | 3.206 ± 0.218 | 6 | 1.776 ± 0.099 | |
| | 63 | 1.967 ± 0.165 | 1 | 3.177 ± 0.219 | 6 | 1.744 ± 0.099 | |
| | 64 | 2.249 ± 0.165 | 6 | 3.326 ± 0.218 | 6 | 1.879 ± 0.099 | 6 |
| | 65 | 2.291 ± 0.165 | 6 | 3.394 ± 0.218 | 6 | 1.829 ± 0.099 | 6 |
| | 66 | 1.765 ± 0.166 | | 3.002 ± 0.221 | | 1.686 ± 0.100 | |
| Totals | | 2.083 ± 0.166 | 284 | 3.201 ± 0.220 | 276 | 1.821 ± 0.099 | 262 |

Table S7 Mean Net Breeding Values (NBV) ± standard errors from the 2020 (coppice) assessment for orchard thinning scenario at the Drummartin *E. polybractea* progeny trial and the number of individuals selected from each family at each assessment.

| Population | Family | Mean NBV 2013 | No. selected 2013 | Mean NBV 2020 | No. selected 2020 |
|---|---|---|---|---|---|
| Charcoal Tank Rd | 25 | 1.930 ± 0.124 | 6 | 2.311 ± 0.104 | 6 |
| Kerrs Lane | 5 | 1.744 ± 0.103 | | 2.079 ± 0.095 | |
| | 6 | 1.769 ± 0.103 | | 2.096 ± 0.094 | |
| | 44 | 1.884 ± 0.123 | 2 | 2.182 ± 0.103 | |
| Talimba Rd West | 45 | 1.788 ± 0.124 | | 2.233 ± 0.105 | 1 |
| West Mid-West Hwy | 28 | 1.842 ± 0.093 | | 2.219 ± 0.089 | |
| | 29 | 1.814 ± 0.093 | | 2.233 ± 0.090 | |
| | 30 | 1.785 ± 0.093 | | 2.248 ± 0.090 | 4 |
| | 31 | 1.955 ± 0.093 | 6 | 2.284 ± 0.090 | 6 |
| West Yalgogrin | 32 | 1.941 ± 0.094 | 6 | 2.146 ± 0.090 | |
| | 33 | 2.011 ± 0.094 | 6 | 2.222 ± 0.091 | |
| | 34 | 2.019 ± 0.094 | 6 | 2.183 ± 0.091 | |
| | 35 | 1.980 ± 0.094 | 6 | 2.200 ± 0.090 | |
| Winters Lane | 7 | 1.974 ± 0.082 | 6 | 2.342 ± 0.086 | 6 |
| | 8 | 1.906 ± 0.083 | 6 | 2.273 ± 0.086 | 6 |
| | 9 | 1.916 ± 0.082 | 6 | 2.319 ± 0.086 | 6 |
| | 10 | 1.967 ± 0.082 | 6 | 2.274 ± 0.086 | 6 |
| | 11 | 2.035 ± 0.082 | 6 | 2.324 ± 0.086 | 6 |
| | 12 | 1.948 ± 0.083 | 6 | 2.289 ± 0.087 | 6 |
| | 13 | 1.876 ± 0.083 | 1 | 2.298 ± 0.086 | 6 |
| | 14 | 1.958 ± 0.082 | 6 | 2.292 ± 0.086 | 6 |
| | 15 | 1.968 ± 0.082 | 6 | 2.339 ± 0.086 | 6 |
| | 16 | 1.931 ± 0.082 | 6 | 2.267 ± 0.086 | 6 |
| | 17 | 1.899 ± 0.082 | 6 | 2.279 ± 0.086 | 6 |
| | 18 | 1.886 ± 0.082 | 2 | 2.308 ± 0.086 | 6 |
| | 19 | 1.986 ± 0.083 | 6 | 2.335 ± 0.086 | 6 |
| | 20 | 1.951 ± 0.082 | 6 | 2.301 ± 0.086 | 6 |
| | 21 | 1.939 ± 0.082 | 6 | 2.262 ± 0.086 | 6 |
| | 22 | 1.966 ± 0.082 | 6 | 2.339 ± 0.086 | 6 |
| | 23 | 1.923 ± 0.082 | 6 | 2.282 ± 0.086 | 6 |
| | 24 | 2.006 ± 0.082 | 6 | 2.326 ± 0.086 | 6 |
| | 50 | 1.893 ± 0.082 | 5 | 2.296 ± 0.087 | 6 |
| | 51 | 1.990 ± 0.082 | 6 | 2.317 ± 0.086 | 6 |
| | 52 | 1.965 ± 0.082 | 6 | 2.299 ± 0.086 | 6 |
| | 53 | 1.975 ± 0.083 | 6 | 2.350 ± 0.086 | 6 |
| | 54 | 1.895 ± 0.082 | 5 | 2.326 ± 0.086 | 6 |
| | 55 | 1.940 ± 0.082 | 6 | 2.309 ± 0.086 | 6 |
| West Wyalong Town | 1 | 1.791 ± 0.082 | | 2.224 ± 0.086 | |
| | 2 | 1.811 ± 0.082 | | 2.217 ± 0.087 | |
| | 3 | 1.861 ± 0.082 | | 2.261 ± 0.086 | 6 |
| | 4 | 1.858 ± 0.082 | | 2.255 ± 0.086 | 6 |
| | 26 | 1.821 ± 0.082 | | 2.252 ± 0.085 | 5 |
| | 27 | 1.897 ± 0.082 | 6 | 2.247 ± 0.086 | 2 |
| | 36 | 1.887 ± 0.082 | 4 | 2.247 ± 0.086 | 3 |
| | 37 | 1.859 ± 0.082 | | 2.226 ± 0.086 | |
| | 38 | 1.876 ± 0.082 | 1 | 2.252 ± 0.086 | 5 |
| | 39 | 1.821 ± 0.082 | | 2.226 ± 0.086 | |
| | 40 | 1.829 ± 0.082 | | 2.254 ± 0.086 | 6 |
| | 41 | 1.989 ± 0.082 | 6 | 2.340 ± 0.086 | 6 |
| | 42 | 1.831 ± 0.082 | | 2.221 ± 0.086 | |
| | 43 | 1.892 ± 0.082 | 6 | 2.266 ± 0.086 | 6 |
| | 46 | 2.038 ± 0.082 | 6 | 2.268 ± 0.086 | 6 |
| | 47 | 1.882 ± 0.082 | 4 | 2.233 ± 0.086 | |
| | 48 | 1.858 ± 0.082 | | 2.304 ± 0.086 | 6 |
| | 49 | 1.935 ± 0.082 | 6 | 2.281 ± 0.086 | 6 |
| | 56 | 1.933 ± 0.082 | 5 | 2.258 ± 0.087 | 4 |
| | 57 | 1.881 ± 0.082 | 1 | 2.276 ± 0.086 | 5 |
| | 58 | 1.747 ± 0.082 | | 2.233 ± 0.086 | 1 |
| | 59 | 1.802 ± 0.082 | | 2.220 ± 0.086 | |
| | 60 | 1.938 ± 0.082 | 6 | 2.208 ± 0.086 | |
| | 61 | 1.887 ± 0.082 | 4 | 2.214 ± 0.086 | |
| | 62 | 1.825 ± 0.082 | | 2.177 ± 0.086 | |
| | 63 | 1.811 ± 0.082 | | 2.231 ± 0.087 | |
| | 64 | 1.927 ± 0.082 | 6 | 2.276 ± 0.085 | 6 |
| | 65 | 1.946 ± 0.082 | 6 | 2.230 ± 0.086 | |
| | 66 | 1.727 ± 0.082 | | 2.171 ± 0.086 | |
| Totals | | 1.899 ± 0.086 | 238 | 2.259 ± 0.088 | 240 |

Table S8 The selections of families for the clonal selection scenario from each of the three biomass assessment across the three *E. polybractea* progeny trials.

9

| | | | Newdegate | | Drummartin | | | Condobolin | | |
|---|---|---|---|---|---|---|---|---|---|---|
| Population | Family | 3-year sapling | 6-year sapling | Coppice | 3-year sapling | Coppice | 3-year sapling | 6-year sapling | Coppice | Total selected |
| Talimba Rd West | 45 | 1 | 1 | 1 | | | | | 1 | 4 |
| West Mid-West Hwy | 31 | 1 | | | | | | | | 1 |
| West Yalgogrin | 33 | 1 | | | 1 | | | 1 | | 3 |
| | 34 | | | 1 | 1 | | | | | 2 |
| | 35 | | | | 1 | | | | | 1 |
| Winters Lane | 7 | | | | | 1 | | | | 1 |
| | 9 | | | | | 1 | | | | 1 |
| | 10 | | 1 | | | | 1 | | | 2 |
| | 11 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 8 |
| | 15 | | | | | 1 | | | | 1 |
| | 19 | 1 | 1 | 1 | 1 | 1 | | 1 | 1 | 7 |
| | 20 | | 1 | | | | | | | 1 |
| | 22 | | 1 | 1 | | 1 | | 1 | 1 | 5 |
| | 24 | | | | 1 | 1 | 1 | 1 | | 4 |
| | 51 | | | | 1 | | 1 | | | 2 |
| | 52 | | | 1 | | | | | 1 | 2 |
| | 53 | | | | 1 | 1 | | | 1 | 3 |
| | 54 | | | | | 1 | | | | 1 |
| | 55 | | 1 | | | | | 1 | | 2 |
| West Wyalong Town | 27 | | | | | | | | 1 | 1 |
| | 38 | | | 1 | | | | | | 1 |
| | 41 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 8 |
| | 43 | | | 1 | | | | | 1 | 2 |
| | 46 | 1 | 1 | 1 | 1 | | 1 | 1 | 1 | 7 |
| | 49 | 1 | | | | | 1 | 1 | | 3 |
| | 56 | 1 | | | | | | | | 1 |
| | 60 | | | | | | 1 | | | 1 |
| | 61 | | 1 | | | | | | | 1 |
| | 64 | | | | | | 1 | | | 1 |
| | 65 | 1 | | | | | 1 | 1 | | 3 |

# Appendix D:

## Supplementary material- Chapter 6

Determinants of the economic viability of mallee eucalypts as a short rotation coppice crop integrated into farming systems of Western Australia

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## Methods – additional information

### *Above ground mallee biomass assessment*

Every mallee in the first two rows of each plot were measured annually from 2006 to 2012 in the allocated season of harvest. Unharvested treatments were measured in autumn. For the unharvested mallee, each stem was measured 10 cm above ground level with diameter tape or callipers in which two perpendicular measurements were recorded. These data were converted to Equivalent Diameter near Root Collar (EDRC), to provide a single aggregated diameter estimate (Chojnacky & Milton, 2008). Crown Volume Index (CVI) method was used for coppice by measuring the height and two perpendicular crown widths in cm. CVI is the volume of the mallee in $m^3$.

### *Mallee biomass estimate*

CVI or EDCR were converted to green biomass using methods and site specific allometric equations presented in the supplementary material in Spencer *et al.* (2019). At Site 20, a general species equation was used.

Plot biomass was estimated by measuring two rows of mallee in a belt, an outer-row and the adjacent row. This approach accurately estimated the standing biomass of 2-row sites. Yield symmetry between rows within a belt is observed in belts of mallee for 3, 4 and 6 rows (Spencer *et al.*, 2019). These principles were used to standardise biomass of sites with more than two rows. For example, the plot biomass of a 4-row belt was estimated by doubling the biomass of the measured two rows to account for the two unmeasured rows.

Across all sites, the within belt inter-row spacing was approximately 2 m. There was also a ~2 m uncropped area adjacent to the belts. Plot-level biomass was calculated in green biomass per hectare per harvest ($Mg\ ha^{-1}$) which included the uncropped distance on each side of the belt.

### *Crop and pasture yield*

To calculate grain yield, strips 1.7- or 1.8-m wide were machine harvested. Canola crops were hand harvested in five 0.5 $m^2$ quadrates and above ground green biomass was weighed in the field just prior to the crop harvest. At each site, subsamples were oven dried at 70 $^{o}C$ and harvest index determined. This was used to estimate dry grain weight.

Above ground pasture biomass was assessed annually in September using a visual assessment outlined by Campbell and Arnold (1973). Each measurement strip consisted of either 20 (2006-09) or 10 (2010-11) assessments. The above ground pasture biomass was averaged for each measurement strip at each treatment.

### *Production Costs and Prices for grain and sheep production*

The operational costs associated with crop and sheep production were estimated using regional data for each experimental year (Planfarm-Bankwest, 2007, 2008, 2009, 2010, 2011, 2012). This included all relevant variable and fixed operating costs: labour wages, contracts, cartage, crop insurance, fertiliser, fuel and oil, chemicals for control of weeds, diseases and pests, repairs and maintenance, fencing, levies, transport, storage and handling. Additional costs for pasture/sheep production were fodder and agistment, livestock expenses, shearing, rams and sheep purchases. Farm management and costs including rates, utilities and administration expenses were excluded from the analysis.

Table S1 Crop or pasture type yield at each site with names as defined in Spencer *et al.* (2019) and Sudmeyer *et al.* (2012) respectively. Open paddock yield is yield without competition from mallee belt for each growing season (2006/07 to 2011/12), and the relative yield in the competition zone, expressed as percentage of open yield, adjacent to each harvest treatment, 2 – 20 m adjacent to the mallee belts. Harvest treatments are long and short frequencies (3 to 6 years) in autumn or spring.

| Site number (Spencer) | Site number (Sudmeyer) | Year | Crop | Open yield (Mg/ha) | Autumn short (% yield) | Autumn long (% yield) | Spring short (% yield) | Spring long (% yield) | Unharvested (% yield) |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 1 | 2006 | Wheat | 2.1 | 77 | | 55 | | 58 |
| | | 2007 | Pasture | 3 | 90 | | 89 | | 75 |
| | | 2008 | Wheat | 1.8 | 84 | | 84 | | 91 |
| | | 2009 | Pasture | 2.1 | 93 | 88 | 80 | 75 | 73 |
| | | 2010 | Pasture | 1.3 | 106 | 88 | 97 | 90 | 71 |
| | | 2011 | Oats | 3.9 | 81 | 95 | 77 | 89 | 85 |
| 3 | 2 | 2006 | Barley | 3 | 85 | | 81 | | 79 |
| | | 2007 | Pasture | 7.5 | 90 | | 97 | | 82 |
| | | 2008 | Pasture | 6.4 | 87 | | 93 | | 84 |
| | | 2009 | Pasture | 3.3 | 92 | 88 | 89 | 93 | 92 |
| | | 2010 | Pasture | 0.9 | 103 | 79 | 99 | 70 | 95 |
| | | 2011 | Pasture | 1.6 | 91 | 96 | 93 | 90 | 81 |
| 8 | 4 | 2006 | Wheat | 1 | 69 | | 47 | | 36 |
| | | 2007 | Lupins | 2.2 | 92 | | 78 | | 69 |
| | | 2008 | Wheat | 1.7 | 70 | 70 | 62 | 56 | 51 |
| | | 2009 | Canola | 0.7 | 65 | 52 | 58 | 48 | 49 |
| | | 2010 | Wheat | 0.4 | 59 | 65 | 46 | 30 | 39 |
| | | 2011 | Wheat | 3.4 | 66 | 71 | 67 | 68 | 61 |
| 13 | 5 | 2006 | Wheat | 0.6 | 69 | | 40 | | 35 |
| | | 2007 | Pasture | 1.5 | 98 | | 97 | | 71 |
| | | 2008 | Wheat | 2.4 | 67 | | 65 | | 56 |
| | | 2009 | Pasture | 0.9 | 114 | 106 | 124 | 93 | 70 |
| | | 2010 | Wheat | Crop failed | | | | | |
| | | 2011 | Wheat | 2.1 | 68 | 70 | 65 | 58 | 55 |
| 18 | 8 | 2006 | Pasture | 4.36 | 90 | | 94 | | 89 |
| | | 2007 | Pasture | 3.8 | 87 | | 80 | | 81 |
| | | 2008 | Pasture | 2.6 | 86 | | 87 | | 81 |
| | | 2009 | Canola | 1.5 | 68 | 55 | 64 | 68 | 61 |
| | | 2010 | Wheat | 3.3 | 80 | 78 | 79 | 77 | 74 |
| | | 2011 | Pasture | 1.6 | 86 | 88 | 93 | 84 | 67 |
| 19 | 9 | 2006 | Canola | 1.8 | 92 | | 96 | | 80 |
| | | 2007 | Wheat | 4.8 | 88 | | 92 | | 90 |
| | | 2008 | Pasture | 3.8 | 88 | | 95 | | 78 |
| | | 2009 | Wheat | 4.2 | 88 | 79 | 85 | 77 | 76 |
| | | 2010 | Canola | 1.1 | 84 | 102 | 107 | 88 | 84 |
| | | 2011 | Wheat | 4 | 73 | 75 | 72 | 65 | 56 |
| Site number (Spencer) | Site number (Sudmeyer) | Year | Crop | Open yield (Mg/ha) | Autumn short (% yield) | Autumn long (% yield) | Spring short (% yield) | Spring long (% yield) | Unharvested (% yield) |
| 5 | 10 | 2006 | Wheat | 1.7 | | 48 | | 29 | 44 |
| | | 2007 | Wheat | 1 | | 50 | | 75 | 26 |
| | | 2008 | Wheat | 1.9 | | 114 | | 128 | 83 |
| | | 2009 | Wheat | 1.6 | | 76 | | 82 | 57 |

| | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | 2010 | Canola | 0.7 | | 30 | | 35 | 20 |
| | | 2011 | Wheat | 3.1 | | 76 | | 79 | 48 |
| 16 | 6 | 2006 | Wheat | 0.5 | 65 | | 46 | | 52 |
| | | 2007 | Pasture | 0.5 | 73 | | 71 | | 35 |
| | | 2008 | Wheat | 1.4 | 65 | | 82 | | 50 |
| 12 | 12 | 2006 | Pasture | 1.6 | 93 | | 67 | | 78 |
| | | 2007 | Pasture | 5 | 77 | | 78 | | 73 |
| | | 2008 | Oats | 1 | 84 | | 78 | | 69 |
| | | 2009 | Pasture | 1.5 | 58 | | 77 | | 72 |
| | | 2010 | Pasture | 2.6 | 58 | | 52 | | 61 |
| | | 2011 | Pasture | 6.1 | 71 | | 68 | | 70 |
| 15 | 14 | 2006 | Pasture | 0.6 | 60 | | 54 | | 79 |
| | | 2007 | Wheat | 1 | 76 | | 79 | | 31 |
| | | 2008 | Wheat | 3 | 82 | | 88 | | 51 |
| | | 2009 | Wheat | 1.6 | 82 | | 102 | | 64 |
| | | 2010 | Wheat | 0.9 | 49 | | 53 | | 29 |
| | | 2011 | Canola | 0.7 | 63 | | 58 | | 34 |
| 20 | 15 | 2006 | Wheat | 2.6 | 86 | | 82 | | 77 |
| | | 2007 | Barley | 2.6 | 85 | | 81 | | 82 |
| | | 2008 | Pasture | 3.1 | 72 | | 85 | | 56 |
| | | 2009 | Canola | 1.6 | 73 | | 76 | | 70 |
| | | 2010 | Wheat | 3.7 | 65 | | 78 | | 69 |
| | | 2011 | Barley | 2.3 | 53 | | 64 | | 48 |

Table S2 Crop and pasture costs per hectare (AUD/ha) for each growing season across all study sites. Regions (H5, M2, M4 and L2) refer to PlanFarm Bankwest Benchmarks (Planfarm-Bankwest, 2007, 2008, 2009, 2010, 2011, 2012). Cost incurred include: wages, contracts, cartage, insurance, fertiliser, fuel/oil, weeds and pest control, plant repairs, building/fencing/water and commodity handling fees and levies. Additionally, for cropping, seed/grading is included, and for pasture, fodder & agistment

| Planfarm index | Sites in each region | 2006 ($/ha) | 2007 ($/ha) | 2008 ($/ha) | 2009 ($/ha) | 2010 ($/ha) | 2011 ($/ha) |
|---|---|---|---|---|---|---|---|
| H5 Crop costs | 18, 19, 20 | 174 | 218 | 300 | 246 | 283 | 322 |
| H5 Pasture costs | | 226 | 265 | 353 | 332 | 324 | 373 |
| M2 Crop costs | 5 | 179 | 208 | 293 | 320 | 274 | 335 |
| M2 Pasture costs | | 191 | 216 | 298 | 339 | 280 | 340 |
| M4 Crop costs | 3, 8, 12, 13 | 155 | 168 | 236 | 220 | 183 | 212 |
| M4 Pasture costs | | 175 | 192 | 254 | 259 | 203 | 235 |
| L2 Crop costs | 15, 16 | 102 | 96 | 186 | 186 | 140 | 190 |
| L2 Pasture costs | | 108 | 102 | 185 | 197 | 144 | 196 |

Table S3 Grain prices (AUD) for each growing season (2006 – 2011) sourced from Grain and Graze 3 (G&G3) (Grain & Graze3, 2020) or ABARES (ABARE, 2015). Pasture prices (AUD), which included both wool and sheep sales, were obtained for each growing season in each region (H5, M2, M4 and L2) from PlanFarm Bankwest Benchmarks (Planfarm-Bankwest, 2007, 2008, 2009, 2010, 2011, 2012).

| Year | 2006 ($) | 2007 ($) | 2008 ($) | 2009 ($) | 2010 ($) | 2011 ($) | Source |
|---|---|---|---|---|---|---|---|
| Canola ($/t) | 480 | 659 | 546 | 427 | 638 | 528 | G&G3 |
| Lupins ($/t) | 286 | 339 | 275 | 223 | 330 | 184 | G&G3 |
| Oats ($/t) | 281 | 216 | 160 | 196 | 202 | 236 | ABARES |
| Wheat ($/t) | 239 | 392 | 306 | 226 | 340 | 236 | G&G3 |
| Barley ($/t) | 264 | 352 | 222 | 193 | 281 | 229 | G&G3 |
| Pasture H5 ($/ha) | 139 | 157 | 157 | 159 | 152 | 191 | PlanFarm |
| Pasture M4 ($/ha) | 78 | 72 | 66 | 70 | 85 | 86 | PlanFarm |
| Pasture M2 ($/ha) | 37 | 36 | 32 | 49 | 43 | 44 | PlanFarm |
| Pasture L2 ($/ha) | 19 | 20 | 16 | 24 | 26 | 25 | PlanFarm |

Table S4 The levelised cost for above ground green biomass (AUD/Mg) using low discount rate of 7% (Low DR), medium discount rate of 10% (Med DR) and high discount rate or 13% (High DR). Treatments varied between sites with either a 3- and/or 4-year harvest or a 6-year harvest regime. Site 16 only has only one 3-year harvest cycle.

| Site | Frequency of harvest (years) | Season of harvest | Levelised cost ($/Mg) Low DR | Levelised cost ($/Mg) Med DR | Levelised cost ($/Mg) High DR |
|---|---|---|---|---|---|
| 1 | 3 | Autumn | 42.0 | 42.7 | 43.5 |
| | 3 | Spring | 50.7 | 52.2 | 53.9 |
| | 4 | Autumn | 39.3 | 40.1 | 41.0 |
| | 4 | Spring | 56.8 | 58.8 | 61.0 |
| | Unharvested | | 30.3 | 33.0 | 35.9 |

| | | | | | |
|---|---|---|---|---|---|
| 3 | 3 | Autumn | 47.5 | 50.1 | 52.9 |
| | 3 | Spring | 51.0 | 54.2 | 57.6 |
| | 4 | Autumn | 54.5 | 57.9 | 61.6 |
| | 4 | Spring | 50.4 | 53.3 | 56.4 |
| | Unharvested | | 50.8 | 58.4 | 67.0 |
| 8 | 3 | Autumn | 81.2 | 82.4 | 83.6 |
| | 3 | Spring | 95.0 | 97.3 | 99.6 |
| | 4 | Autumn | 79.6 | 81.4 | 83.2 |
| | 4 | Spring | 90.7 | 93.1 | 95.7 |
| | Unharvested | | 128.2 | 138.0 | 148.5 |
| 13 | 3 | Autumn | 87.3 | 88.4 | 89.4 |
| | 3 | Spring | 108.0 | 110.3 | 112.6 |
| | 4 | Autumn | 104.2 | 106.1 | 108.2 |
| | 4 | Spring | 114.7 | 116.3 | 118.1 |
| | Unharvested | | 80.9 | 86.6 | 92.9 |
| 18 | 3 | Autumn | 48.3 | 48.5 | 48.7 |
| | 3 | Spring | 48.3 | 48.6 | 48.9 |
| | 4 | Autumn | 48.6 | 49.0 | 49.5 |
| | 4 | Spring | 45.2 | 45.6 | 46.0 |
| | Unharvested | | 29.6 | 30.9 | 32.3 |
| 19 | 3 | Autumn | 106.3 | 109.4 | 112.6 |
| | 3 | Spring | 91.5 | 94.1 | 96.7 |
| | 4 | Autumn | 103.7 | 107.9 | 112.4 |
| | 4 | Spring | 99.1 | 103.0 | 107.0 |
| | Unharvested | | 80.6 | 87.0 | 94.1 |
| 16 | 3 | Autumn | 53.6 | 54.1 | 54.5 |
| | 3 | Spring | 61.9 | 62.9 | 63.9 |
| | Unharvested | | 33.1 | 33.5 | 34.0 |
| 5 | 4 | Autumn | 80.7 | 82.7 | 84.7 |
| | 4 | Spring | 113.7 | 117.9 | 122.5 |
| | Unharvested | | 76.9 | 82.4 | 88.3 |
| 12 | 6 | Autumn | 64.3 | 70.7 | 77.7 |
| | 6 | Spring | 59.2 | 64.7 | 71.0 |
| | Unharvested | | 78.0 | 89.3 | 101.9 |
| 15 | 6 | Autumn | 148.2 | 156.3 | 164.8 |
| | 6 | Spring | 243.3 | 256.9 | 271.4 |
| | Unharvested | | 212.4 | 227.0 | 242.6 |
| 20 | 6 | Autumn | 179.4 | 190.1 | 201.5 |
| | 6 | Spring | 244.7 | 261.0 | 278.7 |
| | Unharvested | | 221.5 | 237.0 | 253.7 |
| Average | | | 89.2 | 93.6 | 98.3 |
| SD | | | 54.8 | 58.5 | 62.5 |
| CV (%) | | | 61.4 | 62.5 | 63.6 |

Table S5 The break-even mallee income (AUD) required to offset the total costs incurred by mallee from the agroforestry paddock compared to agricultural paddock over the 6-year trial and the proportion of mallee costs attributable to direct and indirect costs from the agroforestry paddock for each site and treatment. The direct cost of mallee includes establishment, maintenance and harvest. The indirect costs consist of the opportunity cost, being the area foregone to agriculture, and the competition cost, being income lost from lower crop yields in the competition zone.

| | | | | Direct Costs (%) | | Indirect Costs (%) | |
|---|---|---|---|---|---|---|---|
| Site | Frequency of harvest (years) | Season of harvest | Total Costs | Establishment and maintenance cost | Harvest costs | Opportunity cost | Competition cost |
| 1 | 3 | Autumn | 67,653 | 5 | 54 | 12 | 29 |
| | 3 | Spring | 73,097 | 5 | 46 | 11 | 38 |
| | 4 | Autumn | 61,187 | 6 | 59 | 13 | 23 |
| | 4 | Spring | 58,700 | 6 | 42 | 13 | 39 |
| | Unharvested | | 34,345 | 9 | 0 | 23 | 68 |
| 3 | 3 | Autumn | 37,203 | 26 | 53 | -5 | 27 |
| | 3 | Spring | 38,656 | 25 | 50 | -5 | 30 |
| | 4 | Autumn | 35,248 | 27 | 46 | -6 | 32 |
| | 4 | Spring | 41,843 | 23 | 50 | -5 | 32 |
| | Unharvested | | 21,604 | 41 | 0 | -9 | 68 |
| 8 | 3 | Autumn | 34,239 | 11 | 28 | 23 | 38 |
| | 3 | Spring | 38,075 | 10 | 24 | 21 | 45 |
| | 4 | Autumn | 34,501 | 11 | 29 | 23 | 37 |
| | 4 | Spring | 40,571 | 9 | 26 | 19 | 46 |
| | Unharvested | | 32,707 | 11 | 0 | 24 | 65 |
| 13 | 3 | Autumn | 53,783 | 9 | 26 | 6 | 59 |
| | 3 | Spring | 56,644 | 9 | 21 | 6 | 64 |
| | 4 | Autumn | 50,708 | 10 | 22 | 7 | 61 |
| | 4 | Spring | 60,503 | 8 | 20 | 6 | 66 |
| | Unharvested | | 57,008 | 8 | 0 | 6 | 86 |
| 18 | 3 | Autumn | 74,939 | 17 | 46 | 12 | 25 |
| | 3 | Spring | 77,074 | 16 | 46 | 12 | 25 |
| | 4 | Autumn | 81,789 | 16 | 46 | 11 | 27 |
| | 4 | Spring | 83,290 | 15 | 50 | 11 | 24 |
| | Unharvested | | 45,256 | 26 | 0 | 20 | 54 |
| 19 | 3 | Autumn | 86,257 | 10 | 22 | 44 | 23 |
| | 3 | Spring | 82,043 | 11 | 26 | 47 | 17 |
| | 4 | Autumn | 85,522 | 10 | 23 | 45 | 22 |
| | 4 | Spring | 89,605 | 10 | 24 | 43 | 23 |
| | Unharvested | | 75,349 | 11 | 0 | 51 | 38 |
| 16 | 3 | Autumn | 16,231 | 8 | 42 | 7 | 44 |
| | 3 | Spring | 12,107 | 10 | 37 | 9 | 44 |
| | Unharvested | | 12,486 | 9 | 0 | 9 | 82 |
| 5 | 4 | Autumn | 29,761 | 10 | 29 | 18 | 43 |
| | 4 | Spring | 23,850 | 13 | 21 | 22 | 44 |
| | Unharvested | | 29,759 | 9 | 0 | 18 | 73 |
| 12 | 6 | Autumn | 40,371 | 45 | 42 | -7 | 19 |
| | 6 | Spring | 45,348 | 40 | 45 | -6 | 20 |
| | Unharvested | | 26,313 | 66 | 0 | -10 | 44 |
| 15 | 6 | Autumn | 40,439 | 6 | 17 | 22 | 56 |
| | 6 | Spring | 32,246 | 8 | 10 | 27 | 55 |
| | Unharvested | | 58,451 | 4 | 0 | 15 | 81 |
| 20 | 6 | Autumn | 51,757 | 12 | 14 | 35 | 40 |
| | 6 | Spring | 45,671 | 13 | 10 | 39 | 37 |
| | Unharvested | | 46,138 | 13 | 0 | 39 | 49 |

Table S6 The levelised cost of each harvest treatments at a discount rate of 10%. Sensitivities were performed for carbon dioxide equivalent price per Mg $CO_2$e of AUD15 and AUD30. Sensitivities were also performed on the below-ground carbon biomass estimates with the three categories of minimum below-ground biomass (Min BGB), average below-ground biomass (Avg BGB) and maximum below-ground biomass (Max BGB) per Mg $CO_2$e across the 6-years of experimental data. The methodology for estimating minimum, maximum and average below-ground biomass is detailed in methods section.

| | | Season of | $0 $CO_2$e | | $15 Mg $CO_2$e | | | $30 Mg $CO_2$e | |
|---|---|---|---|---|---|---|---|---|---|
| Site | Frequency of harvest | harvest (years) | Scenario b ($/Mg) | Min BGB ($/Mg) | Avg BGB ($/Mg) | Max BGB ($/Mg) | Min BGB ($/Mg) | Avg BGB ($/Mg) | Max BGB ($/Mg) |
| 1 | 3 | Autumn | 43 | 41 | 40 | 39 | 39 | 37 | 36 |
| | 3 | Spring | 52 | 51 | 49 | 48 | 50 | 46 | 43 |
| | 4 | Autumn | 40 | 38 | 37 | 36 | 36 | 34 | 31 |
| | 4 | Spring | 59 | 58 | 55 | 54 | 56 | 52 | 49 |
| 3 | 3 | Autumn | 50 | 44 | 42 | 40 | 38 | 35 | 31 |
| | 3 | Spring | 54 | 49 | 46 | 44 | 44 | 38 | 33 |
| | 4 | Autumn | 58 | 52 | 48 | 45 | 45 | 39 | 32 |
| | 4 | Spring | 53 | 49 | 46 | 43 | 44 | 39 | 33 |
| 8 | 3 | Autumn | 82 | 77 | 75 | 74 | 72 | 69 | 65 |
| | 3 | Spring | 97 | 92 | 91 | 89 | 88 | 84 | 81 |
| | 4 | Autumn | 81 | 77 | 74 | 72 | 72 | 67 | 63 |
| | 4 | Spring | 93 | 89 | 87 | 85 | 85 | 81 | 76 |
| 13 | 3 | Autumn | 88 | 85 | 83 | 81 | 81 | 77 | 74 |
| | 3 | Spring | 110 | 106 | 104 | 102 | 101 | 97 | 93 |
| | 4 | Autumn | 106 | 102 | 99 | 96 | 98 | 91 | 86 |
| | 4 | Spring | 116 | 111 | 109 | 107 | 106 | 101 | 97 |
| 18 | 3 | Autumn | 49 | 44 | 42 | 40 | 39 | 35 | 31 |
| | 3 | Spring | 49 | 44 | 43 | 41 | 40 | 37 | 33 |
| | 4 | Autumn | 49 | 44 | 43 | 41 | 40 | 37 | 33 |
| | 4 | Spring | 46 | 41 | 39 | 38 | 37 | 33 | 30 |
| 19 | 3 | Autumn | 109 | 104 | 102 | 100 | 98 | 94 | 91 |
| | 3 | Spring | 94 | 89 | 88 | 86 | 84 | 82 | 79 |
| | 4 | Autumn | 108 | 101 | 100 | 98 | 95 | 91 | 88 |
| | 4 | Spring | 103 | 97 | 95 | 94 | 91 | 87 | 85 |
| 16 | 3 | Autumn | 54 | 48 | 45 | 43 | 41 | 36 | 31 |
| | 3 | Spring | 63 | 55 | 51 | 47 | 47 | 39 | 32 |
| 5 | 4 | Autumn | 83 | 81 | 79 | 76 | 79 | 75 | 70 |
| | 4 | Spring | 118 | 116 | 112 | 109 | 114 | 105 | 99 |
| 12 | 6 | Autumn | 71 | 57 | 50 | 44 | 43 | 29 | 16 |
| | 6 | Spring | 65 | 56 | 48 | 43 | 46 | 32 | 21 |
| 15 | 6 | Autumn | 156 | 153 | 151 | 149 | 150 | 146 | 141 |
| | 6 | Spring | 257 | 253 | 250 | 245 | 250 | 243 | 234 |
| 20 | 6 | Autumn | 190 | 166 | 154 | 146 | 141 | 117 | 102 |
| | 6 | Spring | 261 | 233 | 211 | 195 | 206 | 162 | 128 |
| Average | | | 91 | 85 | 82 | 79 | 79 | 73 | 67 |
| SD | | | 54 | 51 | 49 | 48 | 49 | 46 | 44 |
| CV (%) | | | 59 | 60 | 60 | 60 | 62 | 63 | 66 |

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# Appendix E:

## Statement of attribution by co-authors

## Statement of attribution by co-authors

I, Beren Spencer-Grayling, have devised this thesis with data collected from many established mallee trials. With assistance from those that established the trials, I have developed a narrative for each chapter. I collected most of the data, processed all the datasets, interpreted the datasets and have written each manuscript. Supervisors (Ayalsew Zerihun, Richard Mazanec, Amir Abadi and Mark Gibberd) and former colleagues (John Bartle, Robert Sudmeyer, Dan Huxtable and Sarah Van Gent) have assisted with manuscript editing which has improved the scope and quality of the manuscripts. Ayalsew Zerihun and Richard Mazanec have assisted me with data analysis. Ayalsew Zerihun has also helped me through the peer-review process.

Listed below are the attributions for each of the chapters contained in this thesis. Below this is a signed statement from each co-author.

Chapter 2

Spencer B, Bartle J, Huxtable D, Mazanec R, Abadi A, Gibberd M, Zerihun A (2019). A decadal multi-site study of the effects of frequency and season of harvest on biomass production from mallee eucalypts. Forest Ecology and Management 453:117576. https://doi.org/10.1016/j.foreco.2019.117576

J.B. and D.H. established the experiment. B.S. and D.H. carried out the experiment. B.S. drafted the manuscript. R.M. and A.Z. commented on the statistical models. A.Z., J.B., R.M., A.A., D.H. and M.G. contributed to the final version of the manuscript.

Chapter 3

Spencer B, Bartle J, Abadi A, Gibberd M, Zerihun A (2021). Planting configuration affects productivity, tree form and survival of mallee eucalypts in agroforestry systems. Agroforestry Systems 95(1):71–84 https://doi:10.1007/s10457-020-00543-0

J.B. conceived the idea of the experiment. B.S. carried out the experiment and drafted the manuscript. A.Z. commented on the statistical models. A.Z., J.B., A.A. and M.G. contributed to the final version of the manuscript.

Chapter 4

Spencer B, Mazanec R, Abadi A, Gibberd M, Zerihun A (2020). Flowering phenology in a *Eucalyptus loxophleba* seed orchard, heritability and genetic correlation with biomass production and cineole: breeding strategy implications. Scientific Reports 10:15303 https://doi:10.1038/s41598-020-72346-3

R.M. conceived the idea of the experiment. B.S. carried out the experiment and drafted the manuscript. R.M. and A.Z. commented on the statistical models. R.M., A.Z., A.A. and M.G. contributed to the final version of the manuscript.

Chapter 5

Spencer B, Mazanec R, Gibberd M, Zerihun A (in press). Sapling and coppice biomass heritabilities and potential gains from *Eucalyptus polybractea* progeny trials. Tree Genetics & Genomes.

R.M. established the trials. B.S. and R.M. conceived the idea of the experiment. B.S. carried out the experiment and drafted the manuscript. B.S. and R.M. completed the statistical analysis. R.M., A.Z., and M.G. contributed to the final version of the manuscript.

Chapter 6

Spencer B, Abadi A, Bartle J, Sudmeyer R, Van Gent S, Gibberd M, Zerihun A (2021). Determinants of the economic viability of mallee eucalypts as a short rotation coppice crop integrated into farming systems of Western Australia. GCB Bioenergy 13:242-256 doi: https://doi.org/10.1111/gcbb.12775

J.B. and R.S. designed the experiment. B.S. and A.A. conceived the conceptual design of the study. S.V.G. and B.S. built the economic model. B.S. drafted the manuscript. R.S., S.V.G, J.B., A.Z., A.A. and M.G. contributed to the final version of the manuscript.

I, as a co-author, agree that my contribution to each chapter has been accurately assessed above:

Ayalsew Zerihun ____________________________

Richard Mazanec ____________________________

John Bartle _________________________________

Amir Abadi ________________________________

Mark Gibberd ______________________________

Robert Sudmeyer ____________________________

Dan Huxtable _______________________________

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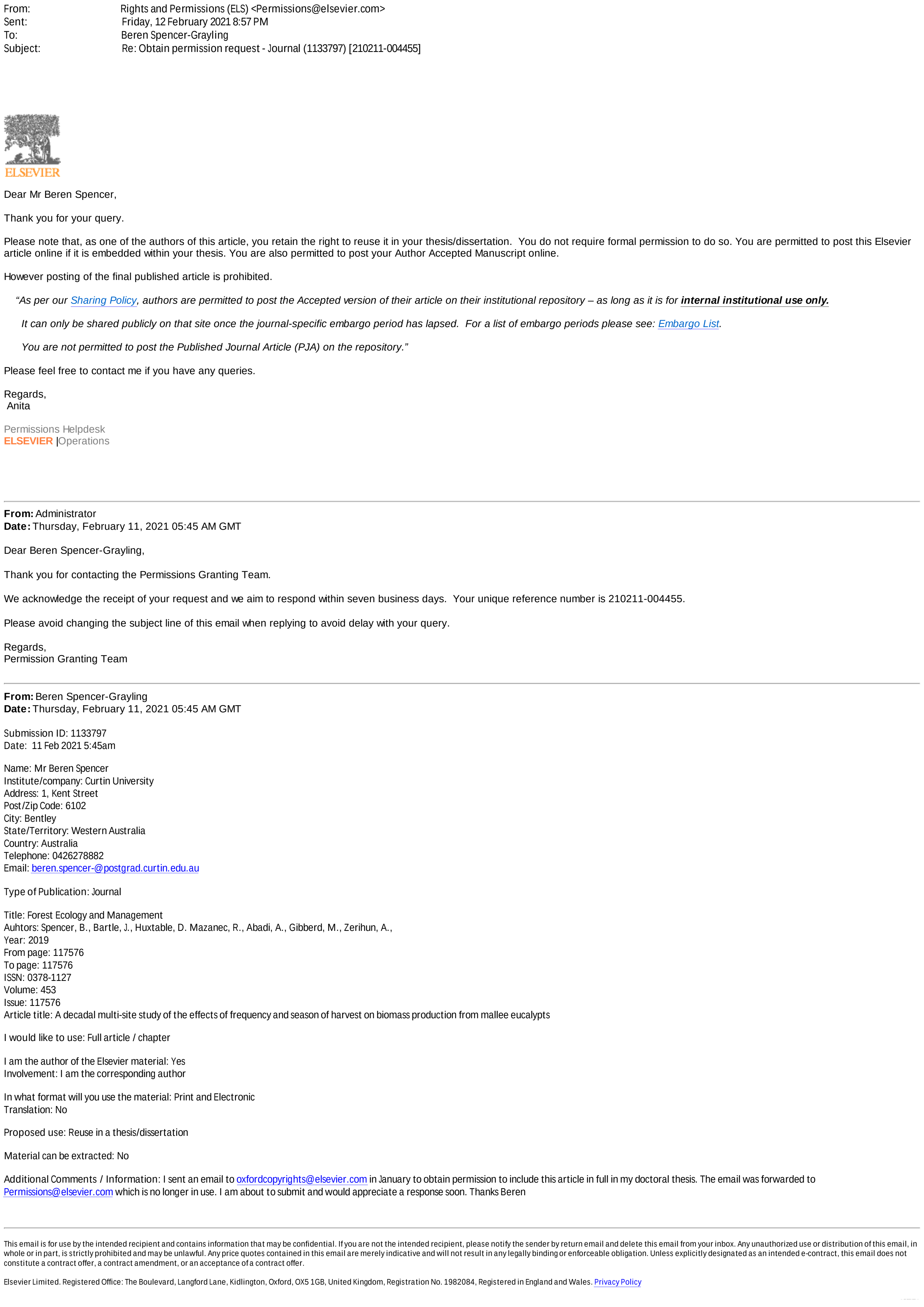

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Type of Publication: Journal

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