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Assessing genetic variation within *Eucalyptus camaldulensis* for survival and growth on two spatially variable saline sites in southern Australia

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ABSTRACT

Eucalyptus camaldulensis Dehnh. provenance–family trials were assessed on two southern Australian saline discharge sites, each of which demonstrated spatial and temporal heterogeneity for salinity and groundwater-table depth. Ninety-six seedlots (from 29 Australian provenances) including 82 individual, open-pollinated families, 14 bulked provenances and one clone of *E. camaldulensis* as well as one seedlot each of *E. grandis* and *E. occidentalis* were evaluated for survival and growth up to age 34 months. Significant differences in growth were found among provenances and families within provenances. The best provenances at both sites were all from north-western Victoria, particularly those from the terminal wetland of the Wimmera River around Lake Albacutya. Inclusion of statistical model terms for soil salinity (EC_e), spatially-oriented incomplete blocking and autoregressive spatial error terms improved the partitioning of within-site variance and were valuable for making selections on these heterogeneous sites.

1. Introduction

Eucalyptus camaldulensis Dehnh. (river red gum) is the most widely distributed of all eucalypts, occurring between approximately 11°S and 38°S and 114°E and 152°E. It is typically a riverine species, found along watercourses and on flood plains in the arid and semi-arid zones of mainland Australia (Boland et al., 2006). It is well adapted to nutrient-poor, low-rainfall sites and is amongst the most tolerant of eucalypts to moderately-saline and waterlogged conditions (Marcar and Crawford, 2004).

E. camaldulensis is planted for a variety of applications including household and industrial energy, pulp fibre, solid timber and environmental remediation. Under favourable plantation conditions, it can grow rapidly and will coppice well for several rotations (Eldridge et al., 1993). It can be readily mass-propagated by vegetative cuttings, which is a preferred method of deployment in many places, as well as by tissue culture. These traits, together with its tolerance of drought, salinity and waterlogged soils make it an attractive hybrid parent. Control-pollinated crosses with *E. grandis* and *E. globulus* have been tested in Australia and overseas (e.g. Hardner et al., 2011; Madhibha et al., 2013) and clones of *E. urophylla* × *camaldulensis* have been deployed commercially in southern China.

The species' diverse uses and ability to adapt to a wide range of climates has resulted in it being one of the most widely planted eucalypts in the world, particularly in countries with long dry seasons (Eldridge et al., 1993). More recently, studies have suggested dividing the species into seven subspecies (McDonald et al., 2009) based on its considerable molecular and morphological variation (Butcher et al., 2009). In the humid tropics, *E. camaldulensis* subsp. *simulata* and *obtusa* are grown, mainly for pulp production, whereas in Mediterranean environments *E. camaldulensis* subsp. *camaldulensis* is usually planted, often for fuelwood or environmental remediation. In Australia, *E. camaldulensis* is a prime candidate for remediating saline discharge areas, which are typically low in the landscape with relatively abundant, but saline, water usually associated with shallow groundwater that has risen close to the soil surface under either dryland or irrigated conditions. Selection of particularly salt-tolerant genotypes is therefore of interest.

Considerable variation is known to exist among and within provenances (and among and within subspecies) for growth, form and tolerance to drought, frost, salinity and susceptibility to defoliating insects (Midgley et al., 1989; Eldridge et al., 1993; Floyd et al., 1994; Bennett and George, 1996; Marcar et al., 2002; Mahmood et al., 2003; Marcar and Crawford, 2004). Provenances of *E. camaldulensis* ssp. *camaldulensis* from north-western Victoria such as those from terminal wetlands of the Wimmera River (e.g. Lake Hindmarsh and Lake Albacutya) are known to perform well in southern Australia, particularly on sites which are saline and experience seasonal waterlogging (Marcar and Crawford, 2004).

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The source of the Wimmera River is in the Grampians mountain range in western Victoria, and it runs inland through a series of shallow, terminal lakes. These fill sequentially only in years when rainfall is sufficient with those at the terminus (Lake Albert and Lake Wirrengen) not having had water since 1918 and 1874 respectively (Durham, 2001). The water in the terminal lakes and underlying aquifer is saline, and the salinity increases (up to 7.9 dS m^{-1} has been reported in Lake Hindmarsh) as the water levels in the lakes fall (Wimmera Catchment Management Authority, 2004). Irrigation water extraction from the river system has made the replenishment of water even less frequent, and dieback of *E. camaldulensis* populations has been noted in the last two decades (Wouters, 1993; Marcar, 1997).

A challenge in assessing the relative performance of plants growing on saline and waterlogged sites is dealing with within-site variation (Corwin and Lesch, 2005). Soil salinity is a dynamic property that varies temporally and spatially with depth and across the landscape and is typically very variable at the field trial-site scale (Corwin et al., 2003; Zohar et al., 2010). Accounting for this intra-site variance is likely to improve the accuracy of genotypic evaluations. One approach is to attempt to directly measure the intra-site variability by sampling at a fine scale. A drawback is that it is time consuming and costly to gather enough data to capture temporal variation caused by factors such as rising and falling groundwater and salinity levels. Nevertheless Zohar et al. (2010) effectively used measures of soil salinity to categorize tree species growth responses in a range from 'salt-sensitive' to 'salt-tolerant'. Another approach is to use statistical methods to try and capture and partition the observed intra-site growth variance into genetic and environmental components. Though most properties vary spatially in an erratic way, where sampling is spatially continuous, values at sites that are close together in space are more likely to be similar than those further from one another, i.e., they are statistically inter-dependent. Approaches such as kriging (e.g. Oliver and Webster, 1990) and autoregressive modelling of residual variance (e.g. Gilmour et al., 1997; Costa e Silva et al., 2001) exploit this important feature of spatial data. Though computationally demanding and more complex, statistical methods are generally less expensive to apply than those that involve direct data gathering.

This paper reports genetic parameter estimates at the provenance and family-within-provenance levels for survival and growth of *E. camaldulensis.* The growth of *E. camaldulensis*, which was expected to be moderately salt-tolerant and capable of moderate growth (Marcar and Crawford, 2004), was benchmarked against two other eucalypts, *E. grandis* (low salt tolerance and fast growth rate) and *E. occidentalis* (highly salt tolerant with moderate growth rate), at two saline trial sites in southern Australia. We have compared two methods that adjust for between- and within-site variation estimated (i) directly using salinity (EC_e – estimated soil electrical conductivity of a saturated soil extract determined at a plot-level at a single point in time) and (ii) indirectly, using mixed models incorporating incomplete blocking and/or an autoregressively-correlated residual variance model.

2. Materials and methods

2.1. Seed sources and preparation of planting stock

Eighty-two family seedlots from individual parent trees, fourteen bulked multiple-parent seedlots and a single clone of *E. camaldulensis* were assembled from a set of provenances spread across the southern part of the species' natural range (Table 1). These accessions were mainly from *E. camaldulensis* subsp. *camaldulensis* and subsp. *arida* and originated from a major, individual-tree collection in north-western Victoria. This collection sampled *E. camaldulensis* subsp. *camaldulensis* from the headwaters to the terminal lakes of the Wimmera River/Outlet Creek, Yarriambiack Creek and Avon/Richardson River systems, and the southwest Wimmera Lakes (Thomson and Merwin, 1987). Single, bulked, multiple-parent seedlots of subsp. *simulata*, from the northern part of the species range, and subsp. *minima*, from the Flinders Ranges in South Australia, were also included along with one bulked multiple-parent seed source each of *E. occidentalis* (flat-topped yate) and *E. grandis* (flooded gum). These latter species are respectively highly salt-tolerant (Marcar and Crawford, 2004) and capable of rapid growth under favourable conditions (Arnold et al., 2005) and are prospective for afforestation on dryland sites in temperate Australia and other regions of the world (Harwood et al., 2007; Marcar et al., 2011).

E. camaldulensis and *E. grandis* seedlings were propagated in commercial seedling containers by Narromine Transplants (Narromine, NSW). Seedlings of *E. occidentalis* were obtained from State Forests (Wagga, NSW) and clonal ramets of *E. camaldulensis* were sourced from the Natural Resources Conservation League (Melbourne).

2.2. Trial site details

Location and climate data for the two trial sites are given in Table 2.

The Deniliquin site is located about 7 km N of Deniliquin in southern NSW in a natural saline depression (discharge area) nearby to fields that were, until a few years earlier, irrigated during summer for rice cultivation. Soil texture was sandy to sandy loam, mean root-zone soil salinity slight to moderate (EC_e 2–8 dS m^{-1}) with a shallow watertable that fluctuates seasonally (i.e. shallower and less saline in winter). At the time of planting the site was largely devoid of vegetation except for a few scattered plants of the perennial legume *Medicago sativa* (lucerne).

The Wellington site is located about 12 km SE of Wellington, central-west NSW, on a slight to moderately saline discharge area that slopes gently towards a creek. Soils varied from clay loam to light clay (pH around 7.4) near the surface to light medium/medium clay (pH around 8.9) at about 1 m, were well drained and showed little evidence of mottling. Root-zone (0–0.6 m) soil salinity (EC_e) ranged from 2–9 dS m^{-1}, and the water table depth ranged from about 1.5 to 4.5 m with a salinity (EC_e) range of 2–12 dS m^{-1}, fluctuating seasonally (shallower and less saline in winter-spring). At the time of planting most of the site was covered with *Cynodon dactylon* (couch grass) *Hordeum marianum* (sea barley grass) and *Polypogon monspeliensis* (annual beard grass).

2.3. Site preparation, planting and trial management

In early September 1996, the Deniliquin site was ripped to a depth of 0.6 m in rows 3.5 m apart, cultivated with 1.2 m wide crowder discs to form a small, shallow bank along each rip line. Glyphosate herbicide (1 L ha^{-1}) and NPK starter fertiliser (20 g per plant) was applied along planting lines prior to planting. The trial was planted on 27 and 28 September 1996. All seedlings were hand watered at the time of planting and again two weeks later. In the first year, weeds around trees were manually sprayed with glyphosate herbicide on three occasions. Weeds between rows were slashed in November 1997 and October 1998.

In early September 1996, the Wellington site was ripped to 0.7 m depth in lines 3.5 metres apart, cultivated with 1.2 m wide discs and small mounds created with mounding-cultivation equipment to form a small, shallow bank along each rip line. Initial weed control was with 1 L ha^{-1} glyphosate. The trial was planted on 29–31 October 1996. All seedlings were hand watered at planting and