Physical and nutritional characteristics and performance after planting of *Eucalyptus globulus* Labill. seedlings from ten nurseries: implications for seedling specifications

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Summary

We measured the physical and nutritional characteristics of seedlings of *Eucalyptus globulus* Labill. (Tasmanian blue gum) in the nursery, and examined their effects on growth and mortality after planting in the field. Container-grown seedlings of widely varying specifications from ten nurseries were assessed immediately before planting. After planting, seedling height and collar diameter were recorded at monthly intervals for six months.

Variation in seedling height, collar diameter, number of leaf pairs, visually estimated root development or foliar concentrations of P, K, S, Ca, Mg, Na, Cl, Fe, Mn, Zn, Cu, B and Al did not account for observed height growth after planting. Seedling foliar N and (to a lesser extent) the size of the root ball largely explained the height growth observed after planting. The stems of seedlings with collar diameter $\leqslant$2.2 mm were susceptible to breakage.

From this work, we recommend that nurseries producing *E. globulus* seedlings note the following points if they aim for optimal survival and performance of seedlings after planting on favourable sites:

- foliar N is a good indicator of potential early growth after planting, and concentrations of 15–20 g kg^{-1} are associated with best field performance;
- average seedling collar diameters $\geqslant$2.3 mm seem to prevent breakage of the stem of seedlings of a height typically produced by nurseries (180–270 mm).

Keywords: planting stock; forest nurseries; foliar nutrition; nitrogen; root shoot ratio; hardiness; seedling growth; *Eucalyptus globulus*; Australia

Introduction

Eucalyptus globulus Labill. (Tasmanian blue gum) is the most widely planted plantation species in the world (Tibbits *et al*. 1997). It exhibits rapid early growth but is relatively drought tolerant and consequently suited to a wide range of planting sites. *Eucalyptus globulus* at present accounts for most of the commercial plantings in southern and western Australia.

The area of *E. globulus* plantations in Australia is rapidly expanding, particularly in eastern South Australia, western Victoria and south-west Western Australia. For example, during the 2000 planting season, one forestry company planted 28 million *E. globulus* seedlings on over 23 000 ha. However, much of the planting is in areas where blue gum has not been grown previously on a commercial scale.

Seedling deployers (e.g. a forestry company) send nurseries seedling specifications that list desirable physical, chemical and/or physiological characteristics. This is done to ensure that expectations of the final product are satisfied. Seedling specifications are widely employed (Menzies 1988; Shroeder 1988; Chaisurisi *et al*. 1994; Thompson 1995; Hudson and Carlson 1998). However, for eucalypts, the specifications may differ between regions (Knight and Nicholas 1996) and within regions (Close *et al*. 2000), depending on the prevailing environmental factors that limit the growth of the species at the planting site.

In Tasmania, substantial research has been conducted on specifications for seedlings of *E. globulus* and *E. nitens* for optimal survival and performance on cold sites (Close *et al*. 2000, 2001). However, this research is not readily transferable to mainland sites now being selected for the planting of *E. globulus* seedlings, because establishment at those sites is generally not limited by low temperature and frost. There is little published research addressing seedling specifications of *E. globulus*, a crop worth more than $US5 million in 2001.

Some forestry companies in Australia provide contracted nurseries with a specification for *E. globulus* seedlings, based on seedling height, collar diameter, number of leaf pairs, root system development, foliar nutrient concentrations and disease status. The values prescribed for these characteristics have not been well researched, and their effect on subsequent tree growth is unknown. To gain a better understanding of the effects of various seedling characteristics, a trial was initiated in July 2000 to compare the growth, after planting, of seedlings from ten nurseries. Seedlings from the nurseries exhibited large differences in seedling height and collar diameter, two- to four-fold differences in foliar concentrations of N, P, Na and Cl, and large differences in the ratios of nutrients (i.e. nutrient balance).

Materials and methods

Plant material

Seedlings were raised at ten nurseries under varying conditions of germination timing, climate, potting mix and plug size (Table 1). Seedlings were transported from each nursery to the trial site within one or two days, in shipping containers or under enclosed conditions on trucks. Routine quality assurance checks on delivery ensured that the root balls of seedlings were in a fully hydrated condition. Concentrations of foliar nutrients (N, P, K, S, Ca, Mg, Na, Cl, Al, Fe, Mn, Zn, Cu and B) were determined in 30 randomly-selected seedlings from each nursery. Height, collar diameter and number of leaf pairs of a further 60 randomly-selected seedlings from each nursery were measured just before planting. In addition, apparent hardiness and root quality were assessed visually. Hardiness was ranked 1–4, 4 being 'hardy'. Factors that contributed, to the same degree, to the visual appearance of hardiness were leaf size (large and thin = soft, small and thick = hard), leaf colour (green = soft, red/purple = hard) and internode length (long = soft, short = hard). Root development was ranked 1, 2, 3 or 4, for 'good', 'above average', 'average' or 'poor', respectively. Factors contributing to root development were the degree to which root mass retained the shape of the plug when removed from the container, and the amount of healthy, white, young root visible.

Nutrient analyses

The three most recently expanded leaf pairs of each of the 30 seedlings were dried at 70ºC for 72 h. Total N was determined by the Dumas combustion method (Schindler and Knighton 1999) and S, P, K, Ca, Mg, Na, Al, Fe, Mn, Zn, Cu and B were measured by Inductively Coupled Plasma Atomic Emission Spectroscopy (ICP-AES) (Long and Davis 1989) on a nitric–perchloric acid digest of plant material. Soluble Cl was determined on an acidified aqueous extract of plant tissue by titration with silver nitrate.

Trial site

The trial was established about 13 km south-west of Hamilton, Victoria (37º45'S, 142º02'E), at the 'Foran' treefarm, 180 m asl. Mean annual rainfall and temperature are 691 mm and 12.7ºC at the Hamilton climate research station, 12 km east of Foran. The soil is a Brown Dermosol (Isbell 1996) and consists of a clay loam over light clay over Quaternary basalt. Prior to mounding, the site was sprayed with glyphosate (1 L ha^{-1}) and metsulfuron (5 g ha^{-1}). Planting rows were ripped to a depth of 600 mm and mounded to a wide profile. The mounds were sprayed with glyphosate (0.5 L ha^{-1}), metsulfuron (5 g ha^{-1}), atrazine (3.3 kg ha^{-1}), sulfometuron (1 L ha^{-1}), simazine (3.3 kg ha^{-1}) and ammonium sulphate (1 L ha^{-1}).

The natural distribution of *E. globulus* (ssp. *globulus*) is predominantly coastal or near-coastal, and $<$400 m asl (Williams and Potts 1996). Planting sites are characterised by mean rainfall and annual temperatures of 500–2000 mm and 11–16ºC, respectively (Tibbits *et al*. 1997). Thus in terms of altitude, mean annual rainfall and temperature, the trial site was well suited for the growth of *E. globulus*. Rainfall and temperature during the period of the trial were favourable (Table 2). The site is ex-pasture and of high inherent fertility. On similar ex-pasture soils, *E. globulus* has shown no response to additions of fertiliser (I. Bail, *pers. comm.*). Sites with similar climates and soils in the area typically produce about 2.2 m height growth after one year (P. Smale, *pers. comm.*).

Trial layout

The experiment was in four randomised blocks each containing ten plots. Each plot consisted of three rows of five seedlings (12 m × 10.5 m) from one of the ten nurseries with between- and within-row spacings of 3.5 m and 2.5 m, respectively. There were no buffer seedlings between plots but the trial was surrounded by a buffer of four rows of seedlings.

Climate

A tipping-bucket rain gauge (Davis Rain Collector 2, Davis Instruments, USA) was located on the site. Air temperature was monitored with a thermocouple at a reference height of 1.3 m. Climatic data were logged every hour on a GPSE 101 Series logger (AR Harris GPSE Ltd, New Zealand).

Seedling mortality and performance

All seedlings were measured for height and collar diameter at planting on 20 July, and on 28 August (month 1), 27 September (month 2), 24 October (month 3), 24 November (month 4), 20 December 2000 (month 5) and 23 January 2001 (month 6). Seedling mortality was also assessed, and broken stems and exposure of the seedling root ball to air were noted. Data from 28 August 2000 were used as the first measure of field performance as variation in planting depth produced an inaccurate measure of seedling height growth between the time of planting and that date.

Statistical analyses

The relationships between the full range of measured, quantifiable variables (i.e. excluding visually assessed hardiness) prior to planting and growth after planting were investigated using Pearson's Correlation Coefficients (Proc. Corr.) and stepwise regression (Proc. Reg.) analyses in SAS (SAS Institute Inc. 1989). Variables were not included in the stepwise regression model unless they met a significance level of $P < 0.05$. Relationships between the concentration of foliar N at planting and visually estimated hardiness, and the concentration of foliar N at planting and incremental seedling growth, were explained using regression (Proc. Reg.) analyses in SAS (SAS Institute Inc. 1989). Regression of the concentration of foliar N at planting and incremental seedling growth was done including data from all nurseries, and

Table 1. Location, germination pre-treatments, potting mix and container type at the ten nurseries providing seedlings for this trial. Seedlings were germinated either with or without pre-treatment.

Nursery[1]	Latitude, longitude	Germination pre-treatment[1]	Potting mix	Container type	Container volume (cm^3)	Container depth (mm)	Container cells (No. m^{-2})
Bacchus Marsh	37º41'S 147º11'E	None	Unknown	Hiko	93	87	526
Floriana	37º49'S 144º58'E	None	Debco pinebark ‘Triplemix’	Lannen 81F	85	73	549
Forestry Tasmania	41º34'S 147º11'E	Imbibition pre-treatment, partial drying and stored at 5ºC until germination for 3–5 days at 21ºC	Pine bark, Canadian peat, rock gypsum, Mini Osmocote, Limil lime, Dolomite lime, Micromax, ferrous sulphate, Saturaid®	Lannen 81F	85	73	549
Forrest	33º35'S 115º49'E	Water imbibed, 2 days of warm, moist conditions	100% composted pine bark	Colmax 72	45	50	736
Jayfields	35º44'S 147º19'E	None	65% pinebark, 10% coir peat, 25% peat	Hiko	93	87	526
Moriac	38º15'S 144º10'E	Four days of warm, moist conditions	Fine pine peat, lignum peat, brown coal, sand	Lannen 81F	85	73	549
Narromine Transplants	32º14'S 148º15'E	Controlled environment: 2 days at 25ºC	60% peat moss, 30% vermiculite, 10% polystyrene, Saturaid®	Hiko	93	87	526
North Forest Products	41º03'S 145º49'E	Glasshouse: 14 days at about 21ºC	75% pine bark, 25% peat	Lannen 121	50	73	820
Virginia Nursery	34º56'S 138º36'E	4–6 days at 20–25ºC	Peat moss and vermiculite	Virginia	49	65	700
Woodlea Nursery	41º10'S 147º31'E	Pre-imbibed, controlled environment at 19ºC for 4–5 days	Pinebark 97%, sand 3%	Lannen 81F	85	73	549

[1] In all cases, the seed was sown directly into the containers.

Table 2. Mean monthly climatic variables at the Foran trial site during the period of study. Measurements were logged hourly. January measurements were made until the 16th only.

Variable	August	September	October	November	December	January
Maximum temperature (ºC)	21.1	22.5	28.4	37.2	41.1	42.6
Minimum temperature (ºC)	1.4	3.5	1.7	6.2	4.8	10.5
Average temperature (ºC)	9.9	12.0	13.3	18.6	19.0	23.8
Rainfall (mm)	54.2	76.4	73.6	29.0	19.6	4.2

then after omission of the lowest concentration of foliar N. All data sets passed tests of normality.

Results and discussion

Seedling specifications at planting and field performance

Seedling height, collar diameter, number of leaf pairs, root development or foliar concentrations of P, K, S, Ca, Mg, Na, Cl, Fe, Mn, Zn, Cu and B at planting did not explain the observed growth after planting (Tables 3, 4). Concentrations of all foliar nutrients at planting, except N, were considered above levels that limit *E. globulus* seedling growth (Judd *et al*. 1996).

Carbohydrate and nutrient reserves need to be above a critical level if new growth is not to be limited (Balneaves and Fredric 1983; Balneaves *et al*. 1985; Lauer 1987; Jinks and Kerr 1999; South and Mitchell 1999). It appeared that the minimum biomass of seedlings in this trial was not below a critical level as relative growth after planting was independent of seedling height or collar diameter at planting.

Seedling foliar N and root volume at planting and field performance

Seedling foliar N concentration at planting explained 75, 57, 54, 57 and 51% of variation ($P < 0.05$) in height growth 2, 3, 4, 5 and 6 months after planting, respectively (Fig. 1) or 67 ($P < 0.01$), 26, 22, 28 and 22% (all $P > 0.05$) of variation in height growth 2, 3, 4, 5 and 6 months after planting, respectively, when the lowest concentration of foliar N was omitted from the analysis. Height increment increased logarithmically with increasing concentration of foliar N at planting.

Internal recycling of nutrients in recently transplanted seedlings can contribute significantly to the annual requirement for growth (van den Driessche 1985). Stored foliar N is retranslocated for new leaf growth, and particularly root growth, in *E. nitens* after transplanting (Close 2001). Foliar N content was the only variable of various morphological, physiological and nutritional

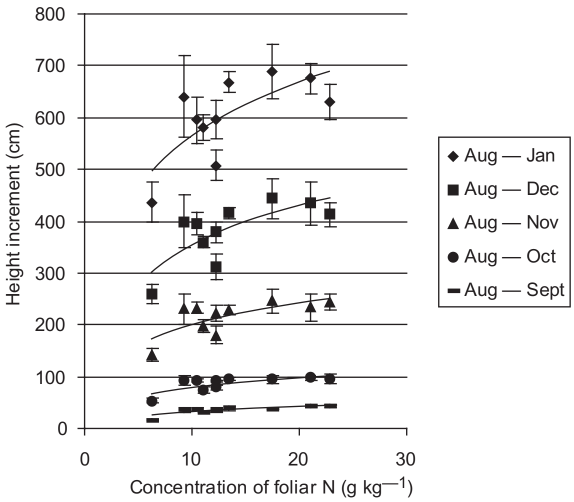

Figure 1. Concentration of foliar N at planting, and height increment during August and September 2000, for *E. globulus* seedlings from ten nurseries

characteristics of *Pinus taeda* seedlings from twenty nurseries in Alabama that was correlated with initial growth following planting (Larsen *et al*. 1988).

Root growth potential (RGP) is defined as the increase in root weight per unit initial root weight of seedlings, growing under favourable conditions during a specific (species dependent) period (Ritchie and Dunlap 1980). RGP is correlated to seedling growth after transplanting (Larsen *et al*. 1988; Williams *et al*. 1988). N stores in roots and root mass also have significant effects on RGP (Carlson 1986; Thaler and Pages 1996). RGP was not measured in this trial but a stepwise regression analysis which incorporated all the variables listed in Tables 3 and 4 was undertaken. The combination of concentration of foliar N and plug depth had a greater correlation with seedling growth than did concentration of foliar N and plug volume. Concentration of foliar N and plug depth were found to be positively correlated with height growth and explained 80, 83, 84, 79 and 73% of variation ($P<0.004$, 0.002, 0.002, 0.004 and 0.01) in growth 2, 3, 4, 5 and 6 months after planting, respectively. A possible explanation for the better correlation of seedling growth to plug depth than to plug volume may be that the root architecture of seedlings in deeper plugs enables a greater initiation of primary roots (Nelson 1996). This may enable faster root establishment and thus greater RGP.

Whilst root depth (in conjunction with the concentration of foliar N) provided the best correlation with seedling growth in this trial, plug volume was significantly correlated (0.80, $P < 0.005$) with root depth. Increased container volume increased the root:shoot ratio for a given seedling height, and (because of the greater root volume) increased the amount of N available for retranslocation. A high RGP soon after planting is desirable in eucalypt and other seedlings because the root:shoot ratio is relatively low in plants raised in plugs, and this restricts acquisition of sufficient nutrients and water to support shoot growth (Ledig 1983; Reitveld 1989; Close 2001).

Height growth after planting was positively correlated with the root:shoot ratio at planting in *P. taeda* (Larsen *et al*. 1988). A study of N-sufficient and N-deficient *E. nitens* seedlings (Close, unpublished data) indicated that RGP increases with increasing foliar N concentration. A similar result was reported in *P. taeda* seedlings (Williams *et al*. 1995). These findings support the correlation between concentration of seedling foliar N at planting and after-planting performance found in the current trial.

Visual estimation of seedling hardiness

Visual hardiness was linearly related to the concentration of foliar N ($r^2 = 0.88$, $P < 0.0001$; Fig. 2). Plants with high N concentrations were considered 'soft'; and those with low N concentrations 'hard'. Low concentrations of foliar N harden eucalypt seedlings to photodamage but have no effect on tolerance to mechanical frost damage (Close *et al*. 2000; Close 2001). Resistance to mechanical frost damage occurs naturally in response to lowering ambient temperatures (Close *et al*. 2000; Greer *et al*. 2000; Close 2001). Thus, a traditional subjective assessment of hardiness in the nursery does not provide a useful guide to frost tolerance of *E. globulus* seedlings after planting.

Table 3. Measured variables of baseline data of seedlings from the ten nurseries at planting on 20 July 2000

Nursery[1]	Height (cm)	Collar diameter (mm)	Height: collar diameter	No. leaf pairs	Hardiness score (1 = poor, 4 = good)	Seedling dry mass (g)
1	23	2.1	11.0	9	2	0.66
2	27	2.3	11.7	11	3	0.65
3	20	2.2	9.1	10	2	0.61
4	27	2.8	9.6	12	3	0.92
5	18	2.1	8.6	14	1	0.47
6	22	2.2	10.0	17	3	0.65
7	20	2.6	7.7	11	2	0.58
8	19	2.2	8.6	12	1	0.62
9	23	2.4	9.6	14	3	0.57
10	25	2.4	10.4	12	4	0.67

[1] Nursery number is randomly allocated and does not correspond to order of presentation in Table 1.

Table 4. Concentrations of foliar nutrient in seedlings from the ten nurseries at planting on 20 July 2000

Nursery[1]	Concentration ($g\ kg^{-1}$)								Concentration ($mg\ kg^{-1}$)					
	N	P	K	S	Ca	Mg	Na	Cl	Al	Fe	Mn	Zn	B	Cu
1	12.2	2.3	17.0	1.4	12.4	4.0	9.9	12.3	30	66	296	58	20	8.8
2	10.5	1.0	16.1	1.2	11.7	2.5	2.9	4.1	89	66	146	40	21	57.4[2]
3	13.5	1.3	16.1	1.3	8.4	3.8	2.2	3.2	35	115	429	60	18	11.0
4	11.0	2.3	15.1	1.2	9.8	3.6	5.4	6.5	30	49	138	23	17	6.0
5	22.8	2.4	18.6	2.3	15.1	3.4	2.4	5.3	39	112	137	62	25	8.6
6	12.3	2.3	12.1	1.4	13.9	3.1	3.0	4.9	62	302	329	34	16	132.3[2]
7	17.4	2.6	13.9	1.9	10.6	4.3	6.6	8.2	90	127	122	62	15	14.6
8	21.0	3.5	18.9	2.1	10.8	3.1	2.5	4.7	76	87	1234	43	20	114.7[2]
9	9.3	2.1	13.1	1.0	9.9	3.6	3.2	5.1	76	80	974	51	15	7.2
10	6.2	1.3	12.4	0.8	8.1	3.0	6.0	5.8	114	136	108	48	14	0.9

[1] Nursery number is randomly allocated and does not correspond to the order of presentation in Table 1.
[2] High Cu levels were linked to the application of Cu-based fungicides that were probably present on leaf surfaces.

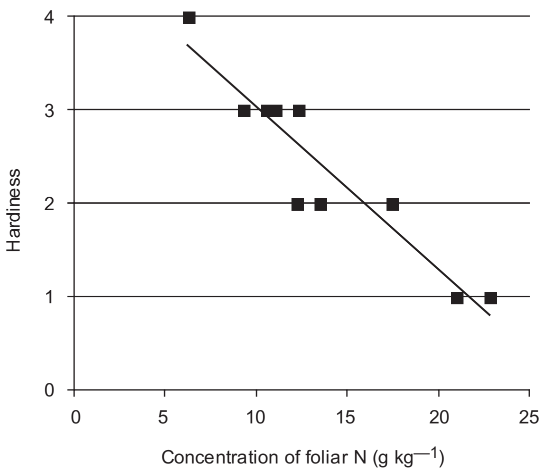

Figure 2. Visually estimated seedling hardiness *vs* concentration of foliar N of *E. globulus* seedlings from ten nurseries at planting

Monitoring of temperature during seedling development, particularly during the month before planting, is recommended to nursery managers as the best measure for gauging hardiness to mechanical frost damage.

Seedling hardiness and field performance

The strong correlation between the concentration of foliar N at planting and field performance indicated that hardy seedlings perform relatively poorly in the field. Hardy seedlings have traditionally been regarded as 'desirable' by nursery managers for both in-nursery management (e.g. to minimise fungal attack of 'soft' tissues or mortality due to drought) and field survival (e.g. to ensure tolerance of drought or frost).

Historically in Australia, seedlings have not been planted on a commercial scale on the high quality ex-agricultural sites of low frost risk that are currently being converted to *E. globulus* plantations. Previously, hardy seedlings have been demanded of nursery managers by forestry companies to ensure seedling survival on their climatically sub-optimal sites. Some compromise may be necessary. Nursery managers need to produce seedlings sufficiently hardy to minimise risk of damage or impaired growth during their development in the nursery, but the seedlings at planting must have a high concentration of foliar N and a good balance of other nutrients to maximise their growth after planting.

Critical concentrations of foliar N and maximum growth performance vs. transport and handling characteristics

It has been reported that a concentration of foliar N < 25 g kg^{-1} is deficient for 2–3-mo-old *E. globulus* seedlings (Dell *et al*. 1995) and <19 g kg^{-1} is deficient for 1–2-y-old *E. globulus* saplings in plantations (Dell *et al*. 2001), where deficiency is defined as concentrations below which visual symptoms become apparent. In this trial, only the seedlings with <10 g kg^{-1} had visual symptoms indicative of N deficiency. Severe N deficiency was observed at N <14 g kg^{-1} in 9-week-old *E. globulus* seedlings grown in sand culture (Shedley *et al*. 1995). In this experiment, at least 90% of maximum height increment occurred at a foliar N concentration of 15–20 g kg^{-1} (Fig. 1). This is substantially lower than the value of 26 g kg^{-1} required for 90% of maximum growth reported by Shedley *et al*. (1995). This divergence may have occurred because a controlled environment was used by Shedley *et al*. (1995).

Seedlings with a concentration of foliar N > 20 g kg^{-1} were estimated to be of very low hardiness (Fig. 2). In this trial seedlings were kept well watered before planting and were handled and planted carefully. This is not always the case in practice, however, and seedlings with 15–20 g kg^{-1} foliar N may be more robust to dehydration during transport and rough handling during planting (I. Bail, *pers. comm*.). Thus seedlings with a foliar N concentration within this range represent the best compromise between the requirements of the nursery, transport and planting on the one hand, and attaining maximum growth in the field after planting on the other.

Seedling stem breakage soon after planting

Seedlings from nurseries 1 and 5 (Table 3) had a higher incidence of stem breakage (causing mortality) after planting in the trial than did those from other nurseries. Although this loss was only about 8% and 7% of the seedlings from each nursery, respectively, such breakage may have implications for planting at exposed or windy sites. Seedlings from these nurseries had the lowest average collar diameters: 2.1 mm (Table 3). The greatest number of breakages occurred in seedlings from nursery 5 which were grown in the nursery over a shorter period (about 3 mo) than seedlings in the other nurseries and which had high concentrations of foliar N. Rapid growth in the nursery may have resulted in inadequate lignification of stem tissues. It is well established that plants with inadequately lignified stems or of relatively high height to stem diameter ratios are susceptible to stem breakage (Peltola and Kellomaki 1993; Essen 1994; Munishi and Chamshama 1994; Guo 1999; Peltola *et al*. 2000). Stem breakage in this trial, however, was not linked to the ratio of height to collar diameter (Table 3). Seedlings from nursery 5 had the lowest ratio, implying that inadequate lignification resulted in stem breakage. Nursery 1 seedlings had significantly higher concentrations of foliar Na and Cl. This indicates poor nutrient balance which may detrimentally affect lignification. Lack of, or abnormal, lignification has been reported to arise under conditions of nutrient imbalance in various plant species (Downes and Turvey 1990a,b; Turvey *et al*. 1992; Cachorro *et al*. 1993; Padu 1999; Santandrea *et al*. 2000) including eucalypts (Graham 1976; Dell 1994). Little stem breakage was observed in seedlings of average collar diameter ⩾2.3 mm.

Conclusions and implications

Low temperature limits growth of *E. globulus* seedlings (Close *et al*. 2000). In eastern South Australia, western Victoria and south-west Western Australia, however, the factor most limiting growth is probably moisture deficit. Thus in these regions rapid early growth of seedlings is desired to ensure that the expanding root system has the greatest possible access to deeper soil water during periods of otherwise limiting moisture levels in the upper soil profile. It is likely that the effects of nursery culture on growth after planting in this trial would be greater on sites across the region where more severe summer drought occurs. Thus the results from this trial can be used to formulate specifications for seedlings for sites elsewhere in these regions, and for *E. globulus* planted outside Australia under similar climatic and site conditions, with a reasonable level of confidence.

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