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Short communication

Measuring water stress in *Eucalyptus grandis* Hill ex Maiden seedlings planted into pots

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Abstract

A pot trial incorporating various watering regimes was initiated to assess: 1) water stress in *Eucalyptus grandis* seedlings, and 2) the efficacy of different types of research equipment in quantifying these levels of water stress for applied research. There were two dry soil treatments differing in terms of seedling root plug moisture at transplanting, dry (DD) and wet (WD), respectively, and three treatments consisting of well watered seedlings transplanted into wet soil (WWD, WWW and control). Treatment WWW was re-watered when seedlings were water stressed. The control was maintained at field capacity for the entire trial period. Seedling physiology was assessed by shoot water potential, stomatal conductance and chlorophyll fluorescence. Seedlings with dry root plugs, planted into dry soil were dead one to two days after planting. A wet root plug at the time of transplanting increased seedling shoot water potential and survival for up to three days in dry soil. Planting into wet soil increased shoot water potential for the duration of the trial and was also associated with new root growth. This study indicated that both the pressure chamber and the porometer provided simple and easy to interpret measures of water stress in *E. grandis* seedlings. Measurements of chlorophyll fluorescence did not significantly reflect treatment effects.

Keywords: Chlorophyll fluorescence; *Eucalyptus grandis*; Re-establishment; Shoot water potential; Stomatal conductance

1. Introduction

In South Africa hardwoods are grown primarily for pulpwood over an eight to twelve year rotation. Most hardwoods are planted along the eastern seaboard and adjacent escarpment where rainfall ranges from approximately 600 to 1100 mm (Smith et al., 2005). To optimise productivity on a site specific basis, a variety of eucalypt species and hybrids are planted including: *Eucalyptus grandis*, *E. nitens*, *E. smithii*, *E dunnii*, *E. grandis* × *E. camaldulensis* and *E. grandis* × *E. urophylla* (Smith et al., 2005). Even when planted on highly productive sites mortality following planting can exceed 10%, resulting in sub-optimal stocking which affects final yield in a pulpwood stand (Morris, 1995; Chambers and Borralho, 1997). Survival and initial growth can be associated with one or a combination of the following factors: silvicultural practices (Turvey, 1996; Little and Van Staden, 2003), seedling quality and age (Zwolinski et al., 1995; Bayley and Kietzka, 1997) and incorrect site-species matching (Darrow, 1995). These factors may become more critical if plants are grown under adverse environmental conditions that negatively affect the ability of the seedling to utilize the site's resources.

Worldwide, water deficits are one of the major causes of failure during re-establishment, and a seedlings ability to use water efficiently is crucial to post-planting survival (Burdett, 1990; Margolis and Brand, 1990). If the seedling does not receive water during the period of new root development, its internal water deficits will increase considerably (Burdett, 1990). The natural distribution of *Eucalyptus* species in Australia is strongly influenced by their ability to manage water stress, either as seedlings, or as they continue to grow (Myers and Landsberg, 1989). Water stress following planting is considered one of the major causes of eucalypt seedling death in South Africa (Viero and Little, 2006) and Ethiopia (Gindaba et al., 2004). The availability of soil water as a function of rainfall is thus important when planting eucalypts in South Africa and most planting is confined to the summer months

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when higher rainfall is expected (Viero and Little, 2006). Even during the rainy season, however, rainfall can be erratic often forcing foresters to plant in less than ideal conditions. In addition, in regions with high mid-summer temperatures, such as the sub-tropical coastal regions in KwaZulu Natal, planting is carried out during the cooler, winter months when the potential for rainfall is lower (Viero and Little, 2006). The planting of eucalypts in South Africa is thus often characterized by the inclusion of water into the planting hole, as this has been shown to reduce the risk of early mortality (Viero et al., 2002; Viero and Little, 2006).

Since eucalypt seedling mortality may be partly attributed to soil water deficits, an understanding of the physiological response to water stress following planting is necessary if measures to alleviate water stress are to be developed. While some physiological research on eucalypts has been conducted (Myers and Landsberg, 1989; Stoneman, 1994; Gindaba et al., 2004), little could be found for species locally grown (*E. grandis*, *E. smithii*, *E. dunnii* and *E. nitens*), especially during establishment. Parameters that can be assessed to identify water stress include changes in photosynthetic rate, shoot water potential, transpiration rate, stomatal conductance and chlorophyll fluorescence (Dye, 1996; Roden and Ball, 1996; Rolando and Little, 2003; Stokes, 2004). Factors that limit the number of parameters that can be assessed include manpower, researcher competence and available funding. In addition, for applied studies, physiological data that are accurate, easy to obtain and simple to interpret have the benefit of increasing the value of the study without compromising the objectives of the research. Since there are few references to the measurement of physiological parameters of eucalypt seedlings planted in South Africa, the efficacy and simplicity of different methods in quantifying water stress needs to be assessed. A pot trial incorporating various watering regimes was initiated to assess: 1) water stress in *E. grandis* seedlings and 2) the efficacy of different types of research equipment in quantifying these levels of water stress, particularly for applied research.

2. Materials and methods

The pot trial was carried out at the Institute for Commercial Forestry Research (ICFR) nursery, Pietermaritzburg. The pots (25 cm diameter × 15 cm deep), chosen to reflect the dimensions of standard planting pits prepared for planting, were filled with the equivalent of 4 L of a dry, silty clay soil (47% silt, 45% clay and 8% sand). The pots were sheltered during rainfall events to prevent inadvertent wetting of the soil (and seedlings), as all watering in the trial was controlled. The *E. grandis* seedlings were first grown in a pine bark medium in polystyrene trays with 128 cavities each with a capacity of 36 ml.

The trial consisted of five watering treatments arranged in a randomised complete block design of eight single tree replications (Table 1). The treatments were designed to simulate different levels of water stress at the time of planting and immediately thereafter. Extra seedlings were planted for destructive sampling in treatments WD and WWD. Before planting, all of the seedlings were watered twice daily except the seedlings in treatment DD which did not receive any water on the day before planting. The seedlings were transplanted into the pots on the 10th September 2004. Since eucalypts respond rapidly to water deficits, the trial was terminated 11 days later on the 21st September 2004 when sufficient information had been collected to meet the objectives of the trial.

Table 1
Description of treatments used in a pot trial to determine the effect of water availability at, and after, planting on water stress in *E. grandis* seedlings

No.	Moisture content of root plug at planting	Water applied at planting (L)	Water applied after planting (L)	Treatment aim	Treatment name in text
1	Dry	0	0	– Water stressed before, at and after planting	DD
2	Wet	0	0	– Water stressed at and after planting	WD
3	Wet	1.5	0	– Water stressed after planting	WWD
4	Wet	1.5	1 L at 9 days	– Partial water stress after planting	WWW
5	Wet	1.5	1 L every 1–2 days	– Never water stressed	Control

Air temperature (1.5 m above ground) and relative humidity were measured for the duration of the trial with an Onset Hobo® temperature logger (Onset Computer Corporation) housed in a Stevenson Screen. Vapour pressure deficit (kPa) was calculated from measurements of air temperature and relative humidity (Unwin, 1980). Measurements of soil temperature 10 cm below the soil surface were made with thermocouples (Campbell Scientific, 1997) from the time of planting until trial termination in all treatments, except in treatment DD, as it was assumed that pot soil temperature would be similar to that in treatment WD. Measurements of the volumetric water content ($m^3\ m^{-3}$) of the top 6 cm of soil in the pots were made with a Delta-T Theta Probe type ML2 (Delta-T Devices Ltd) (Little et al., 1996). Using the probe, measurements of soil water content were taken in each pot for all treatments on the day of planting, and at 3, 5, 7, 9 and 11 days after planting (after the application of water in the respective treatments).

The height (ht, cm), groundline diameter (gld, mm), root and shoot dry mass (g) of the seedlings were measured on the day of planting ($n=10$) and at treatment termination ($n=8$). The oven dry mass of new roots emerging from the root plug, as well as the length of the longest new root (cm), was also determined at termination. Stomatal conductance, shoot water potential and chlorophyll fluorescence measurements were taken at mid-day to meet periods when water stress was assumed to be the greatest. Measurements were made on all seedlings on the day before planting, and at 1, 3, 5, 7, 9 and 11 days after planting, except for water potential, where only 4 seedlings were destructively sampled. Stomatal conductance measurements (g_s, mmol $m^{-2}\ s^{-1}$) were made with a LI-1600 Steady State porometer (LI-COR, 1984) and measurements of shoot water potential (Ψ, MPa) were made using the pressure chamber technique (Scholander et al., 1965). Since the leaves and shoots