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Effect of Copper Coating Nursery Containers on Plant Growth and Root Morphology of *Eucalyptus globulus* Labill. Cuttings and Seedlings

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Abstract. Plant production in rigid plastic containers is operationally advantageous in nursery eucalyptus production, but root morphology differs from those grown under natural conditions. Root systems deformations resulting from nursery culture can affect post-planting growth performance and mechanical stability, being instability of eucalypts a major problem. Therefore, in order to improve post-planting response of *Eucalyptus globulus* Labill., we tested the effect of chemical root pruning with $CuCO_3xCu(OH)_2$ on root architecture and plant growth at the nursery and in the field. The concentration of 25 g/L applied to the interior walls of the containers could be suitable for chemical root pruning in cuttings and seedlings. At the end of the nursery growth period, the root morphology of copper-treated plants differed significantly from the untreated ones, but differences in root or shoot dry weights were not significant. In the field, copper-treated plants performed no better than untreated plants four years after planting, and the differences in root morphology observed in the nursery did not affect plant survival or growth. Therefore, these results do not recommend the use of chemical root pruning with copper in order to improve stability of *E. globulus*, at least for the first four years after outplanting.
Key words: chemical root-pruning; copper carbonate; out-planting response

Efeito do Revestimento em Cobre de Contentores de Viveiro sobre o Crescimento e Morfologia Radicular de Plantas Provenientes de Estacas e Sementes de *Eucalyptus globulus* Labill.

Sumário. A produção de plantas em contentores de plástico rígido é vantajosa na produção de eucaliptos em viveiro, mas a morfologia radicular difere da apresentada por plantas que cresceram em condições naturais. As deformações do sistema radicular resultantes da produção em viveiro podem afectar o comportamento pós-plantação relativamente ao crescimento e

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estabilidade mecânica, sendo a estabilidade dos eucaliptos uma questão importante. Assim, para melhorar a resposta pós-plantação do *Eucalyptus globulus* Labill., testámos o efeito da poda química da raiz pelo $CuCO_3xCu(OH)_2$, sobre a arquitectura da raiz e crescimento da planta em viveiro e no campo. A concentração de 25g/l aplicada nas paredes interiores dos contentores seria apropriada para a poda química da raiz de plantas provenientes de estacas e sementes. No final do período de crescimento em viveiro, a morfologia da raiz de plantas tratadas com cobre diferia significativamente da apresentada por plantas sem tratamento, mas as diferenças em peso seco de raízes e lançamentos não foram significativas. No campo, quatro anos após a plantação, o comportamento das plantas tratadas com cobre não foi melhor do que o das plantas sem tratamento, e as diferenças de morfologia da raiz observadas em viveiro não afectaram a sobrevivência ou o crescimento das plantas. Por conseguinte, estes resultados não recomendam o uso de poda química com cobre para melhorar a estabilidade do *E. globulus*, pelo menos nos primeiros quatro anos após a plantação.
Palavras-chave: poda química da raiz; carbonato de cobre; resposta à plantação

Effet du Revêtement par le Cuivre des Pots de Pépinière sur la Pousse et la Morphologie Racinaire des Plantes d'*Eucalyptus globulus* Labill. Provenant de Boutures et de Graines

Résumé. La production de plantes en pots de plastique rigide a des avantages dans la production d'eucalyptus en pépinière, mais la morphologie racinaire diffère de celle présentée par les plantes qui poussent en milieu naturel. Les déformations du système racinaire résultant de la production en pépinière peuvent avoir des effets sur le comportement post-plantation, en ce qui concerne la pousse et la stabilité mécanique, l'instabilité des eucalyptus étant une question majeure. Donc, dans le but d'améliorer la réponse post-plantation de l'*Eucalyptus globulus* Labill., nous avons testé l'effet de la taille chimique par le $CuCO_3xCu(OH)_2$, sur l'architecture du système racinaire et la pousse de la plante en pépinière et dans les champs. La concentration de 25g/l utilisée dans les parois intérieures des pots serait appropriée pour la taille chimique de la racine des plantes provenant de boutures et de graines. À la fin de la période de pousse en pépinière, la morphologie de la racine des plantes traitées avec le cuivre était significativement différente de celle présentée par les plantes sans traitement, mais les différences entre le poids sec des racines et celui des bourgeons n'ont pas été significatives. Quatre années après la plantation définitive, le comportement des plantes traitées avec le cuivre n'était pas supérieur à celui des plantes sans traitement, et les différences de morphologie du système racinaire observées en pépinière n'ont eu aucun effet sur la survie ou la pousse des plantes. Par conséquent, étant donné ces résultats l'utilisation de la taille chimique au cuivre pour améliorer la stabilité de l'*E. globulus*, du moins les quatre premières années après la plantation n'est pas recommandée.
Mots clés: taille chimique de la racine; carbonate de cuivre; réponse à la plantation

Introduction

Successful afforestation in arid and semiarid lands relies on selecting the appropriate genotype, planting adequately conditioned plants, and employing efficient and effective establishment techniques. Well-developed and well-structured root systems with numerous lateral roots are essential attributes of high-quality plants. The root form of nursery plants produced in containers is strongly influenced by container design, and containerisation has been singled out as a cause of plantation instability (SMITH and McCUBBIN, 1992; KRASOWSKI, 2003; CABAL *et al.*, 2005). Containerisation of plants in the forestry industries has advanced rapidly. Root growth is also influenced by the physical and chemical properties of the growing medium, nutrition and mechanical and chemical pruning of roots in plugs. Instability of eucalyptus is a major problem, and wide variations in stability between clones, provenances and species can be found (CHAMPS and MICHAUD, 1985; YU *et al.*, 2000; NEVES *et al.*, 2004b). Good stability in eucalyptus is related to a root system containing taproots (CHAMPS and MICHAUD, 1985), and taproots are present in the typical root morphology of seedlings (SASSE and SANDS, 1997). The root systems of rooted cuttings are fundamentally different from those of seedlings, with lesser number and total length of secondary roots, and total length of primary roots.

The eucalyptus is now the most widely grown commercial hardwood in the world. Eucalyptus plantations are grown in 70 or more countries on around 15 Mha of land and constitute approximately one quarter of all global plantations (BEADLE and SANDS, 2004). For *Eucalyptus globulus* alone, plantations cover more than 1 Mha in the Iberian Peninsula, located in a area with Atlantic climate incidence but with most of it mainly characterised by a typically Mediterranean climate (TOVAL, 2004).

In many cases, containerised roots exhibit weak tap root development with twisted lateral roots that often mat together. This can lead to girdling and strangling of the tap-root and reduced wind-firmness (HUTH *et al.*, 1996). Root coiling problems are particularly significant for fast-growing species in small pots. Nowadays, the containers design can, to some extent, control the incidences of root coiling. For instance, containers have lateral ribs and a drainage hole at the base to reduce root coiling and to encourage air pruning of the roots (CABAL *et al.*, 2005). Nevertheless, containerised roots usually grow in the plug with a "caged" form (i.e.: several outer "caged" roots and a lesser proportion of lateral root branching).

Chemical root pruning in containers eliminates "caging" and the characteristic effect of containers on root form resulting in a changed root distribution within the plug with fewer outer "caged" roots and more internal branching (WENNY *et al.*, 1988; LANDIS, 1990; ALDRETE *et al.*, 2002; NEVES *et al.*, 2004b). Therefore, stability after outplanting could be improved (BELL, 1978; NEVES *et al.*, 2004a). Moreover, arrested root tips are not killed and may resume growth when plants are planted outside the container (SMITH and McCUBBIN, 1992). Chemical root-pruning reduces incidences of damping-off diseases and improves nursery labour productivity by making seedlings easier to extract from containers (NELSON, 1992). It can also reduce water stress during immediate (21 days) post-transplantation (ARNOLD, 1996). Mycorrhizae are not adversely affected (McDONALD *et al.*, 1984).

Several heavy metals (e.g.: copper, lead, cadmium, arsenic, and mercury) may be used to prune roots but they are very toxic to plants and animals, except for copper (LANDIS, 1990; CRAWFORD, 2003). Since the 1960s, chemical root pruning with copper has been used in forestry and ornamental nurseries (SAUL, 1968; BEESON and NEWTON, 1992). When elemental copper is present in the environment it is not biologically active. The only form that is active is ionic Cu^{2+}. Copper in the ionic form quickly complexes with organic matter in the soil and container substrates, rendering it biologically inactive. Fixed copper salts used for root control, like $Cu(OH)_2$ or

$CuCO_3$, release active copper ions over a long period of time (CRAWFORD, 2003) and copper could be toxic to roots. The mode of action for copper-treated containers is the controlled release of copper ions along the container-substrate interface, where Cu ions inhibit root elongation at the root tip. Root inhibition is localised to the root tip, and very little to no excess copper is translocated to other plant parts. The effect is very specific to root tips.

Copper mixed with water based latex paint is applied to the internal surfaces of nursery containers. Therefore, copper is encapsulated in a latex matrix and is very resistant to leaking and dislodgement by rain or irrigation (CRAWFORD, 2003). Several products are already available on the market in South Africa, USA, Canada and Australia (e.g.: Styrodip®, Plazdip®, Spin Out® Root growth regulator, Copperblock®).

As far as we know, a few number of chemical pruning experiments have been carried out on eucalyptus seedlings (SMITH and McCUBBIN, 1992; HUTH *et al.*, 1996; SCHUCH and PITTENGER, 1996; DUNN *et al.*, 1997; PEZZUTTI and SCHUMACHER, 2000; FERNÁNDEZ *et al.*, 2001) and only one has been carried out on *Eucalyptus globulus* rooted cuttings (NEVES *et al.*, 2004b). This research deals with the effects of Cu-based root pruning treatments on the growth of young *Eucalyptus globulus* plants (cuttings and seedlings) and their effect on plant growth and survival for four years after outplanting.

Material and methods

The global experiment was divided into two assays; one for seedlings and the other for cuttings of *Eucalyptus globulus* Labill. Seeds were harvested from "Dehesa del Carmen" (Huelva, Spain) and cuttings belonged to clone nº 13 (Silvasur Agroforestal nursery, Huelva, Spain). This clone is extensively used in forest plantations in Spain and only one clone was studied to avoid genetic variability in root structure (SASSE and SANDS, 1997; NEVES *et al.*, 2004b). The separation into two assays was due to the different growth conditions needed for germinating and rooting. Both types of plants were initially placed under greenhouse conditions (23.5ºC mean temperature and relative humidity over 70%) in permanent moist growing media. The containers (Superleach® 125, 125 cm^3, 19.1 cm depth, 3.2 cm diameter at the top) were filled with a pine bark:peat mixture (1:1, Pons®), and transferred outdoors to 50% shade after germinating or rooting (approximately one-month-old plants), watered when necessary, and fertilised once a week with a soluble fertiliser (10-3-3 + 0,4 Fe + 0,05 B). The container internal surfaces were either coated with a water-based latex paint solution containing 25 or 50 g/L $CuCO_3 \times Cu(OH)_2$ or left uncoated as a test (0 g/L).

The design of the experiment was a randomised complete block with each copper treatment being represented by 63 plants/block. Due to the foreseeable decrease in the number of plants during rooting (i.e. through death, lack of rooting, etc.) we decided to increase the number of blocks for cuttings (Table 1). With regard to the cuttings, 150 plants were obtained from another treatment, which contained a smaller concentration of copper (12.5 g/L), because previous assays led us to suspect that copper was responsible for damaging the aerial part of many cuttings during rooting (TEJERO and PÉREZ, 2002). These were cultivated under the same growing conditions as the other cuttings.

Table 1 - Initial number of plants used for both nursery assays. The copper treatments were a water-based, latex-paint solution containing 0, 25 or 50 g/L $CuCO_3xCu(OH)_2$

Assay	Seedlings	Cuttings[#]
Copper treatment	3	3
Blocks	2	4
Plants/block	63	63
Total number of plants	378	756[#]

[#] Plus 150 cuttings corresponding to 12.5 g/L treatment, used only in the field assay.

When plants were seven months-old (25-35 cm height), twenty plants per treatment and plant type were harvested and the following parameters were measured: height (H); stem diameter (D); leaves, stem and root dry weights after 72 h of oven drying at 75ºC (LDW, StDW and RDW, respectively). Root/shoot dry weight ratio (RSDW) and total dry weight (TDW) were calculated from the weights assessed. Foliar copper concentrations were evaluated by external services with a flame atomic absorption spectrometer (AA-6200, Shimadzu®, Japan). Root morphology and root coiling were assessed by means of the following three parameters:

X = number of main roots (tap-roots for seedlings and primary roots for rooted cuttings (SASSE and SANDS, 1997)).

Y = percentage of the container volume occupied by lateral roots. It was assessed by weighing to the nearest 0.01 g while the roots were submerged in distilled water.

Z = percentage of the external surface of the root core covered by lateral roots. It was assessed visually; each plant assigned one of the following five categories, *viz*. (1) 0-20%, (2) 21-40%, (3) 41-60%, (4) 61-80%, (5) 81-100%.

Three months later, in January 2003, 140 cuttings and 105 seedlings per treatment were planted in two field trials: Aroche and Rociana (Huelva, Spain). Rociana is characterised as a flat site (< 2% slope) and sandy soil, while Aroche has a muddy-sandy soil with 30 % slope. The climate is a typical Mediterranean semiarid one, with mean daily maximum and minimum temperatures of 30.2 - 19.0ºC (Aroche) and 31.6 - 19.2ºC (Rociana) in July; 13.8 - 3.6ºC (Aroche) and 16.3 - 6.6ºC (Rociana) respectively in January. The mean annual temperatures were 16.2 and 17.5ºC for Aroche and Rociana. The long-term average annual rainfalls for the sites are 835 mm (Aroche) and 555 mm (Rociana), 3.0% of these falling during the dry period (from June to August). The design of the field experiments was a randomised complete block in each site. Each plant type and copper treatment were represented by five plants in each block. Survival, height and stem diameter at breast height were assessed 12, 24 and 39 months after planting.

Data were analysed using SPSS® 11.0. One-way analysis of variance was used to test differences between copper treatments. Seedlings and cuttings were analysed separately. Treatments were considered as fixed effects. All the parameters were checked in advance to ensure they complied with normal distribution and variance equality. No data transformation was carried out. The Tukey test (Tukey HSD - Honest Significant Difference-) was used for a comparison of means when differences were significant ($p \geq 0.05$), but the Dunett test was used when an equality of variances was not assumed. Relationships between parameters were analysed by means of linear correlation.

Results

At the end of the nursery period, there were no significant differences in height due to copper treatment (Figure 1) within each type of plants (p = 0.269 and 0.482 for cuttings and seedlings, respectively). On the other hand, there were significant differences between treatments in seedling stem diameters (p = 0.012). Seedlings produced in copper-treated containers had larger diameters than untreated ones. However, differences in cuttings stem diameters were not significant (p = 0.083), and only a trend of slightly larger diameters were observed in 25 g/L treatment than in 0 g/L and 50 g/L treatments (Figure 2). Differences between treatments in roots, leaves and stem dry weights were not significant. The same was the case for the root to shoot dry weight ratio (Table 2). The block effect was not significant for any parameter.

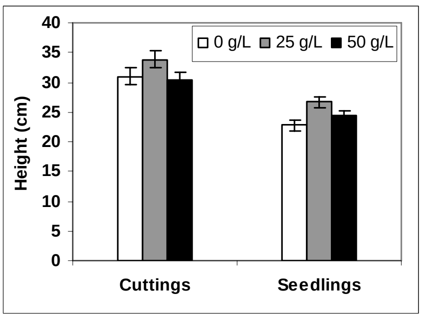

Figure 1 - Mean values (±SE) of plant height per copper treatment and plant type at the end of the nursery period

With regard to root structure parameters (X, Y, Z), differences obtained between treatments were more evident than those obtained for H, D and dry weight (Table 3). Copper untreated plants (both cuttings and seedlings) showed a greater number of main roots (X) and lateral roots appearing on the external surface of the root core (Z) than treated plants. For cuttings, the volume occupied by lateral roots (Y) was significantly greater in the 25 g/L treatment than in 50 g/L treatment.

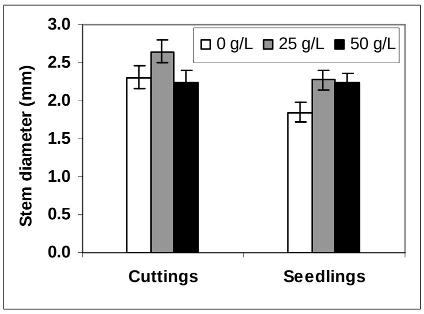

Figure 2 - Mean values (±SE) of plant stem diameter per copper treatment and plant type at the end of the nursery period

Both types of plants showed positive and significant relationships between height (H), stem diameter (D) and dry weight parameters (RDW, LDW, StDW, TDW) at the end of the nursery period. Nevertheless, RSDW correlated significantly only to RDW (and to LDW for seedlings) (Table 4). The X and Z parameters correlated highly and positively between themselves but they showed a weak ($r^2 < 0.18$) or non-significant relationships with H, D and dry weight parameters. With the exception of H for seedlings, there were positive relationships between Y and plant size (H, D, and dry weight). Foliar copper concentrations were significantly different between treatments (Table 5).

Table 2 - Mean values (± SE) of roots, leaves, stems and total dry weights (RDW, LDW, StDW, TDW, respectively) and shoot/root dry weight ratio (RSDW) for each type of plants and copper treatment at the end of the nursery period. *p* = significance level

Plant type	Treatment	RDW (g)	LDW (g)	StDW (g)	RSDW	TDW (g)
Cuttings	0 g/L	0.39 ± 0.04	1.25 ± 0.12	0.98 ± 0.06	0.18 ± 0.01	2.62 ± 0.18
	25 g/L	0.43 ± 0.04	1.50 ± 0.12	1.08 ± 0.06	0.16 ± 0.01	3.01 ± 0.28
	50 g/L	0.36 ± 0.04	1.19 ± 0.12	1.02 ± 0.06	0.16 ± 0.01	2.62 ± 0.18
	p	0.480	0.235	0.676	0.252	0.267
Seedlings	0 g/L	0.25 ± 0.03	0.47 ± 0.05	0.23 ± 0.03	0.34 ± 0.05	0.96 ± 0.06
	25 g/L	0.25 ± 0.03	0.68 ± 0.05	0.38 ± 0.03	0.25 ± 0.05	1.31 ± 0.14
	50 g/L	0.27 ± 0.03	0.58 ± 0.05	0.32 ± 0.03	0.30 ± 0.03	1.09 ± 0.06
	p	0.867	0.191	0.224	0.239	0.242

Table 3 - Mean values (±SE) of root structure parameters at the end of the nursery period

Plant type	Treatment	*X*	*Y* (%)	*Z*
Cuttings	0 g/L	3.77±0.29 a	23 ± 16 ab	2.5 ± 0.08 a
	25 g/L	1.90±0.029 b	41 ± 17 a	1.1 ± 0.08 b
	50 g/L	1.62±0.29 b	11 ± 16 b	1.0 ± 0.09 b
	p	0.001	0.001	<0.001
Seedlings	0 g/L	2.60±0.19 a	22 ± 18 a	3.6 ± 0.10 a
	25 g/L	1.35±0.19 b	37 ± 19 a	1.1 ± 0.08 b
	50 g/L	1.35±0.19 b	22 ± 18 a	1.1 ± 0.10 b
	p	0.030	0.141	0.005

X = Number of main roots. *Y* = Percentage of container volume occupied by lateral roots. *Z* = percentage of the external surface of the root core covered by lateral roots (1: 0-20 %; 2: 21-40 %; 3: 41-60 %; 4: 61-80 %; 5: 81-100 %). *p* = significance level. The different letters in the same column indicate significant differences.

Table 4 - Correlation matrix for the measured parameters at the end of the nursery period (n = 60)

	H	D	RDW	LDW	StDW	RSDW	TDW	*X*	*Y*	*Z*
H	1.000	0.546***	0.405***	0.537***	0.576***	-0.155	0.549***	0.165	0.491***	0.017
D	0.632***	1.000	0.566***	0.647***	0.660***	-0.040	0.663***	0.098	0.657***	-0.082
RDW	0.392**	0.538***	1.000	0.836***	0.820***	0.484***	0.891***	0.338**	0.554***	0.031
LDW	0.643***	0.673***	0.556***	1.000	0.916***	-0.006	0.984***	0.190	0.622***	-0.029
StDW	0.762***	0.857***	0.654***	0.769***	1.000	-0.029	0.964***	0.179	0.598***	-0.123
RSDW	-0.308	-0.226	0.490***	-0.376**	-0.178	1.000	0.069	0.367**	0.078	0.147
TDW	0.696***	0.778***	0.763***	0.940***	0.906***	-0.147	1.000	0.219	0.627***	-0.051
X	-0.071	-0.293*	0.184	-0.131	-0.204	0.420***	-0.092	1.000	0.119	0.609***
Y	0.251	0.353**	0.484***	0.403***	0.438***	0.149	0.483***	-0.062	1.000	0.030
Z	-0.207	-0.359**	0.121	-0.162	-0.280*	0.407***	-0.148	0.936***	-0.085	1.000

The data under the diagonal correspond to seedlings and those over the diagonal to cuttings.
*** significant at $p \leq 0.001$. ** significant at $p \leq 0.01$. * significant at $p \leq 0.05$.

Table 5 - Foliar copper concentrations (ppm) of copper-treated and untreated *Eucalyptus globulus* seedlings and cuttings. *p* = significance level (n = 6)

Treatment	Stecklings	Seedlings
0 g/L	6.50 c	5.16 b
12.5 g/L	6.72 c	---
25 g/L	7.50 b	6.00 a
50 g/L	8.65 a	6.60 a
p	0.035	0.042

The different letters in the same column indicate significant differences.

Pooled survival of the two plant types (cuttings and seedlings) was not significantly different ($p > 0.100$) between copper treatments after 39 months in the field; the rates of survival were over 80 % for both types of plants and the two field trials. Also, seedlings height development at 12, 24 and 39 months was not affected by copper treatments ($p > 0.200$). Seedlings height at 12, 24 and 39 months for all the treatments as a whole were 191±6, 536±10 and 878±11 cm at Aroche trial, and 203±8, 574±19 and 931±27 cm at Rociana trial, respectively. Differences between treatments in stem diameter, for both types of plants, were not significant ($p > 0.146$) as well. Nevertheless, the height of cuttings was significantly different between treatments at 12 and 24 months in the field, but not at 39 months (Figures 3 and 4). In terms of cuttings, after 12 and 24 months, plants were taller when less copper was added to the containers. After 39 months in the field, height (H_{39}) and stem diameter at breast height (D_{39}) were positively correlated ($H_{39} = 10.5914\ D_{39}$, $r^2 = 0.80$, $p < 0.001$ for cuttings; $H_{39} = 10.141\ D_{39}$, $r^2 = 0.78$, $p < 0.001$ for seedlings).

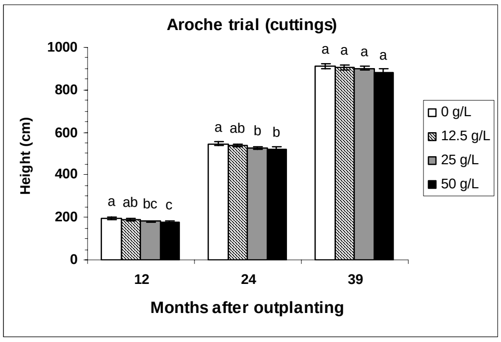

Figure 3 - Mean values (±SE) of plant height after 12, 24 and 39 months in the field (Aroche trial) for cuttings. The different letters indicate significant differences between copper treatments within each measurement date

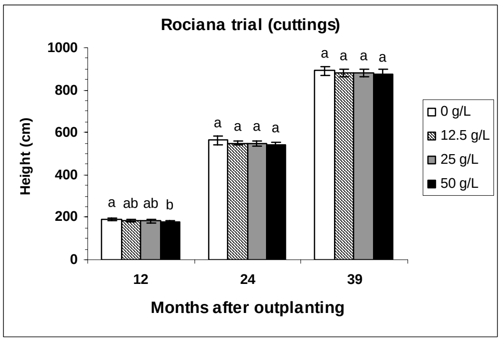

Figure 4 - Mean values (±SE) of plant height after 12, 24 and 39 months in the field (Rociana trial) for cuttings. The different letters indicate significant differences between copper treatments within each measurement date

Discussion

Similar to our results, copper treatment had no effect on nursery growth of several *Eucalyptus* (SMITH and McCUBBIN, 1992; HUTH *et al.*, 1996; SCHUCH and PITTENGER, 1996) and conifer (RUEHLE, 1985; BURDETT and

MARTIN, 1982; McDONALD *et al*., 1984) species and, in some instances, average seedling height and/or root-collar diameter were improved by copper (HUTH *et al*., 1996; FERNÁNDEZ *et al*., 2001; BARAJAS *et al*., 2004; TSAKALDIMI and GANATSAS, 2006). In contrast, DUNN *et al.* (1997) found a small inhibition of growth in the nursery due to the chemical root-pruning (50 g/L $CuC0_3$) of five native Australian tree species. NEVES *et al.* (2004b) also found that copper concentration over 77 g/L cause a significant growth reduction in the nursery.

The low *Y* values (i.e.: low volume occupied by lateral roots) obtained for cuttings in 50 g/L-treated containers respect to 25 g/L treatment might suggest that the first copper concentration was middly toxic. However, the shoots showed no sign of copper toxicity (necrotic stems, leaf chlorosis or senescent leaves). Furthermore, although foliar concentrations of Cu were significantly higher in treated plants than in untreated plants, all levels were within the normal range for *Eucalyptus* (JUDD *et al*., 1996) and well below what is considered toxic (>20 ppm dry weight) in plant tissues (MARSCHNER, 1995). For seedlings, there were no significant differences in *Y* due to copper treatment.

Without chemical pruning the architecture of the root system showed the main and lateral roots well developed and directed vertically. When copper was used, however, the lateral roots were pruning, which resulted in a more fibrous root system, with fewer roots on the external surface of the root cores and at the bottoms of the containers. These results coincide with those obtained by many other authors (SMITH and McCUBBIN, 1992; SCHUCH and PITTENGER, 1996; PEZZUTTI and SCHUMACHER, 2000; FERNÁNDEZ *et al*., 2001; NEVES *et al*., 2004a). Growth performance in the nursery and root structure suggest that 50 g/L may be an excessive concentration of copper in the water based paint for *E. globulus*, particularly for cuttings; 25 g/L are preferable for this purpose. Although 12.5 g/L treatment was not evaluated at the end of the nursery period, the morphology and root structure were apparently similar to those of the 25 g/L treatment. On the other hand, the positive relationships between *Y* and plant size, and since plant size was not affected by *X* and *Z* parameters, a more branched root system (i.e.: higher *Y* values) contributed to produce larger plants.

We need to bear in mind that, although chemical root pruning could have some advantages, copper-treated containers could lead to higher costs (greater use of seeds, growing media and containers during sowing) to compensate for seedlings which germinate at the edge of the cavity wall and are thus exposed to excess copper concentrations, resulting in mortality. Similarly, we noticed in these assays that it is necessary to keep leaves away from the edge of the containers during rooting of cuttings because copper can kill some cells, thereby making plants more vulnerable to some diseases such as *Botrytis cinerea*.

The slight differences in growth between treatments during the nursery period or during the first two years in the field, although in some cases significant, did not lead to a different response in the field after 39 months. This lack of differences between treatments in field survival and growth coincide with the results showed by

DUNN *et al.* (1997), who found that there was no effect of copper treatment on height and basal diameter development of *Eucalyptus argophloia* and *E. camaldulensis* seedlings after 24 months of planting on a semiarid site. HUTH *et al.* (1996) found that after twelve months in the field, the effect of copper significantly increased height and stem diameter of six *Eucalyptus* species, had no effect on six other *Eucalyptus* species, and root growth in the upper portion of the root core increased significantly. BURDETT *et al.* (1983), BARAJAS *et al.* (2004) and TSAKALDIMI and GANATSAS (2006) meanwhile found no difference in the growth of chemically pruned and unpruned pine seedlings two to three years after planting out. Sometimes growth improvement by copper coating containers may be noted three to four years after planting in the field (WENNY, 1988; BURDETT *et al.*, 1983), but this was not the case in our experiment.

Previous assays showed us that copper treatment increased the number of lateral roots growing from the upper portion of the plug in the field, while untreated plugs characteristically produced their new roots from the lower portion (FERNÁNDEZ *et al.*, 2001). This coincides with the results obtained by other authors (WENNY, 1988; SMITH and McCUBBIN, 1992; DUNN *et al.*, 1997; BARNETT and McGILVRAY, 2002; NEVES *et al.*, 2004b). These differences later persisted in the field (DUNN *et al.*, 1997; FERNÁNDEZ *et al.*, 2001; RIVERA, 2003; NEVES *et al.*, 2004a). Thus, the root structure achieved by chemical root pruning could produce root systems near the soil surface more similar to those in natural settings than untreated plants. It would also improve anchoring, stability, mineral nutrition and the sites for mycorrhizal infection. Recent studies, however, show that root quality (architecture and root mass) can also be improved by the use of mini-cuttings for plant propagation (BURGUEÑO *et al.*, 2004a,b), without the necessity of chemical root pruning, although this latter method needs to be improved, as well.

In summary, the results suggest that chemical root pruning could be an important nursery practice when growing cuttings or seedlings for arid and semiarid land plantations with variable and unpredictable rainfall. Plants raised specifically for these environments are often kept in the nursery until the soil moisture in the field is adequate. It seems that cuttings are more sensitive to copper coated containers than seedlings. 25 g/L is a suitable copper concentration for chemical root pruning in both types of plants, but seedlings can tolerate a little more concentration and cuttings prefer a little less. However, although dramatic differences in root morphology were observed in the nursery, copper-treated plants performed no better than untreated plants in the 39 months after planting.

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