

**THE DEVELOPMENT OF A WALL-LESS PLUG FOR
PLANTING STOCK OF FOREST TREES**

By

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of the requirement for the degree of**

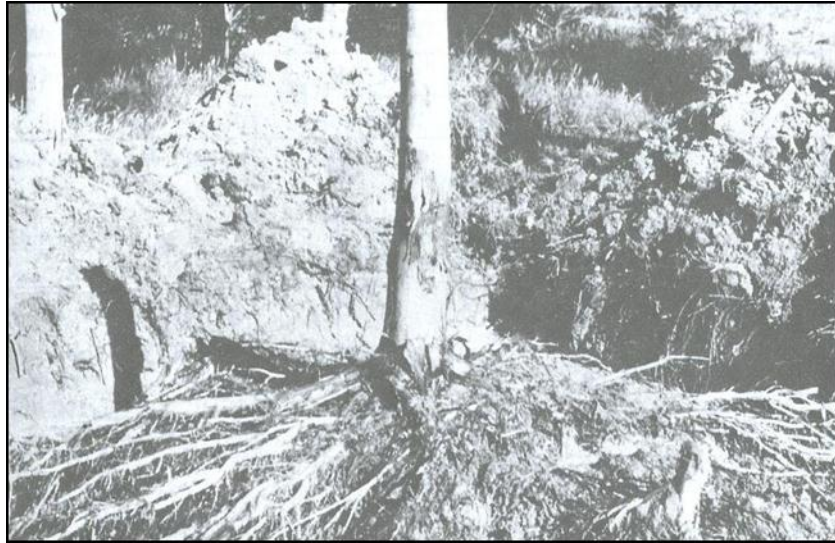
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FRONTICEPIECE



A rare photograph of an excavated root system of a *Eucalyptus grandis* tree at Mildura in Victoria, North Western Australia. Clearly visible is the complexity of the root system [Baldwin and Stewart (1986) Distribution, length and weight of roots in young plantations of *Eucalyptus grandis* W. Hill ex Maiden irrigated with recycled water. *Plant and Soil*. 97: 243-252]



A paper-maché wall-less plug with a *Eucalyptus dunnii* seedling grown from seed ten weeks after germination

ABSTRACT

High output commercial nurseries that cater for the forestry industry are continuously challenged to efficiently and cost effectively produce good quality planting stock to establish large plantations. Currently, South African commercial nurseries produce planting stock in solid compartmentalized trays. One of the major drawbacks of these containers is the susceptibility of trees to root deformation following outplanting, combined with the need for the return of empty containers to the nursery. A potential solution to these challenges is the introduction of wall-less plugs for the production of planting stock. Wall-less plugs are volumes of growing medium, usually cylindrical in shape, devoid of an impenetrable wall in which a plant can grow and establish itself. Such plugs may enable the production of planting stock with improved root systems, without the need for the return of empty containers after outplanting. In this research four prototypes of wall-less plugs were developed, produced on a small scale and tested. These were: 1) Paper-maché plugs made using the original WRIBLOK protocol whereby composted pine bark was bound together with repulped newspaper, 2) Sponge blocks, 3) Hessian bags and 4) Covetan bags. The performance of these prototype wall-less plugs was compared with the performance of four tray types used commercially by the forestry industry: 1) Poly 128 shallow, 2) Poly 98 deep, 3) Unigro 128 and 4) Sappi 49. These are polystyrene and polypropylene-based containers. Of these containers the Unigro 128 and Sappi 49 containers were of similar performance. Performance in terms of height and root collar diameter increase over ten weeks from the time of sowing of the paper-maché plugs was similar to that of the Unigro and Sappi 49 containers. The sponge block, Hessian bags and Covetan bags produced inferior quality planting stock compared to the other treatments tested.

Although little progress was made in the ability to describe how one root system differs from another in terms of their branching patterns, a technique was developed to determine root surface area by image analysis software that is freely available. This method may prove useful for further research and for determining seedling quality in commercial nurseries.

DECLARATION

I hereby declare that the work reported in this dissertation is the result of my own investigation, except where acknowledged.

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I certify that the above statement is correct.

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LIST OF ABBREVIATIONS

AGR	absolute growth rate
ANOVA	analysis of variance
CEC	cation exchange capacity
CERU	Controlled Environment Unit
$(\text{CH}_2\text{O})_n$	carbohydrate empirical formula
$^{\circ}\text{C}$	degrees Celsius
cm	centimeter
cm^2	square centimeter
cm^3	cubic centimeter
CO_2	carbon dioxide
DNA	deoxyribonucleic acid
dpi	dots per inch
g	gram
g/dm^3	grams per cubic decimeter
ITTO	International Tropical Timber Organization
Kg	kilogram
L	litre
Ln	natural log
LSD	least significant difference
m.e./L	milliequivalents per litre
mL	millilitre
mm	millimeter
O_2	oxygen
$P < 0.05$	probability at 95% level of significance
RBI	root-bound index
RCBD	randomised complete block design
RCD	root collar diameter
RGP	root growth potential
RGR	relative growth rate
RNA	ribonucleic acid

rpm	revolutions per minute
RSA	root surface area
RV	root volume
SD	standard deviation
SE	standard error
SFRA	stream flow reduction activity
SRF	slow-release fertilizers
TNC	total nonstructural carbohydrate
TPS	Toyota Production System
TSI	timber stand improvement
W	watt
WRIBLOKS	Wattle Research Institute bloks

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CHAPTER 1 PREFACE

High output commercial nurseries of forest tree planting stock are continuously challenged to efficiently and cost effectively produce good quality planting stock to establish large plantations. In South Africa, seedling production of forest trees is in the region of 240 million plants per year (Zwolinski and Bayley, 2001) to restock approximately 1.5 million hectares of land dedicated to plantation forestry (Owen and van der Zel, 2000). The nurseries produce plants of several species of *Eucalyptus*, *Pinus* and *Acacia* genera. The need for generating such high volumes of transplants is the driving force for further research into improving the strategies presently implemented in nurseries. A small improvement in the system could significantly increase overall profit margins and shareholder dividends.

Currently, South African commercial nurseries produce planting stock in compartmentalized trays that have a number of drawbacks associated with them; (i) Trays with impenetrable solid walls prevent optimal root development after outplanting in the field (Burdett *et al.*, 1983). (ii) The empty containers which represent a substantial capital outlay must be returned to the nursery. This operational procedure must be planned for, provided for financially and carried out in conjunction with other phases of the regeneration process. (iii) Trays manufactured from expandable polystyrene are sometimes dipped in a solution of copper hydroxide to promote disease control and induce root pruning thereby countering the problem of tree instability after transplanting, eliminating root caging, improving nursery labour productivity by making seedlings easier to extract from their containers and also thereby reducing the incidence of damping-off (Nelson, 1992). However this practice can become prohibited in the future if it is deemed to pollute run-off water from nurseries in a way that is environmentally hazardous.

These are considerable limitations to the current method of producing planting stock if one considers the fundamental importance of roots in tree growth and development, the need to reduce labour costs and to make operational procedures as streamlined as possible and lastly the need for nursery operations to comply with environmentally acceptable practices that are audited not only internally by nursery managers, but also by externally-controlled and recognized entities.

Jiffy Products® has developed a soft-walled container that allows the development of a natural lateral root system by making use of air pruning to facilitate this process. In the late 1970s, members of the South African Wattle Research Institute, subsequently renamed the Institute for Commercial Forestry Research (ICFR), developed a kind of soft wall container known as the WRIBLOK (Zeijlemaker and de Laborde, 1977). That system made use of the paper-maché technique whereby growing medium was glued together in the form of a cylindrical block.

The paper-maché technique of producing a wall-less plug was the first prototype of wall-less plug produced and tested in this study. Other prototypes were obtained from cutting up a block of sponge and lastly, by sowing and filling pockets with growing medium. These pockets were made from Hessian and from Covetan, the latter being a material used for the protection of horticultural crops against frost damage. Both Hessian and Covetan allow the roots to pass through the spaces between the threads making up the material. The ability of roots to move through the boundary of the plug into the surrounding air in each of these prototype container systems is critical for efficient air pruning of developing root systems and the subsequent promotion of a highly branched root system that cannot become root-bound.

The principle aim of this investigation was to examine innovative methods to achieve forest regeneration at a reduced cost through research into container types and nursery practices. More specifically, this study aims to investigate the production of a biodegradable wall-less container and furthermore, to compare the quality of planting stock produced in these biodegradable wall-less containers to that produced in containers currently used in the industry. Within the large topic of planting stock quality emphasis has been placed on the issue of root system quality. The problems associated with container-produced planting stock and the research methodology adopted in this research is outlined in Figure 1.1.

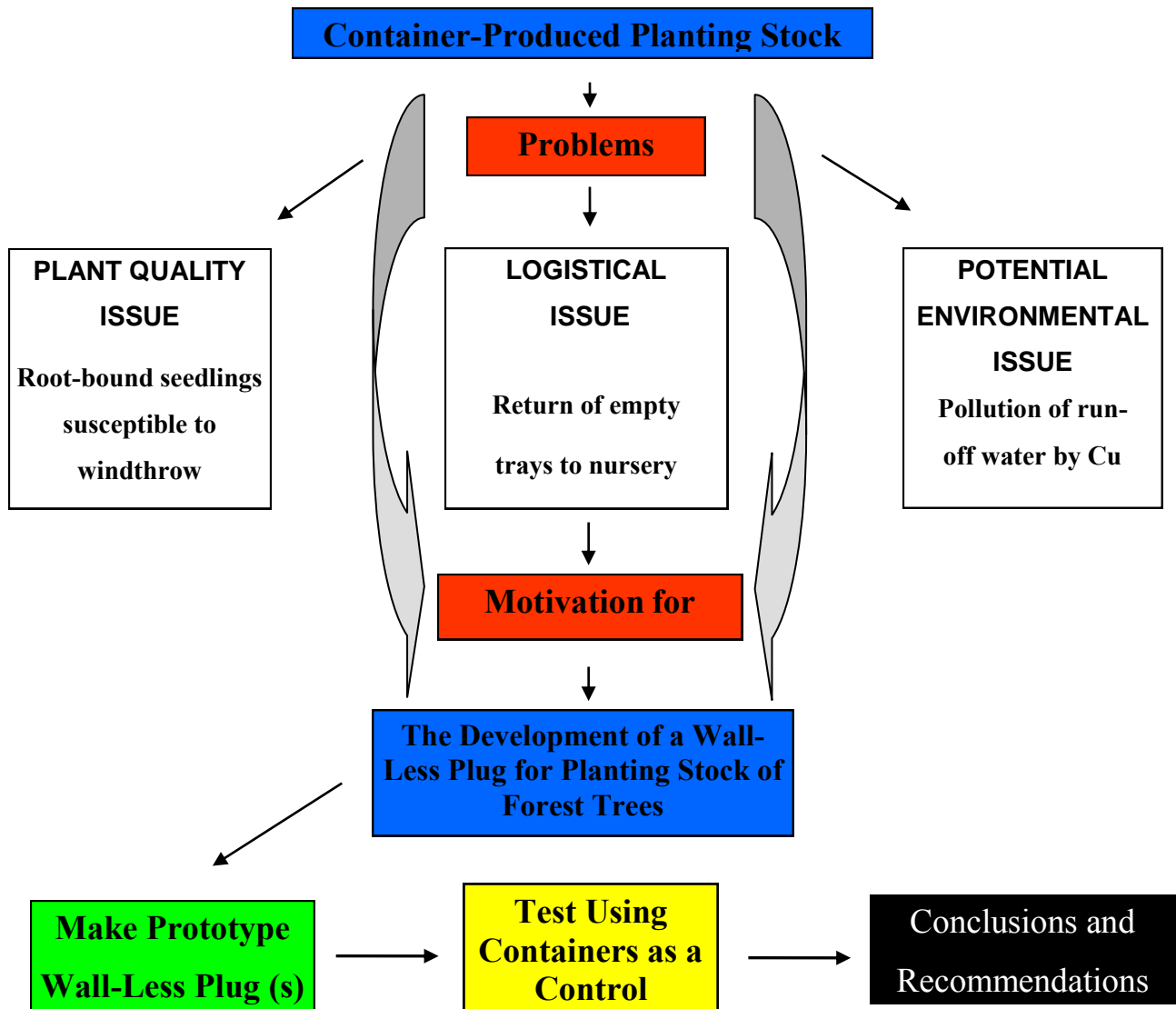


Figure 1.1: Schematic representation of the study drivers and research methodology

It will be useful to commence this discussion by establishing a good definition of seedling quality, as well as the criteria that planting stock must comply with, in order to produce desirable trees with subsequent minimal blanking. In turn such criteria can then be used by nursery managers as a comparative standard to evaluate systems for the production of planting stock in commercial forestry nurseries.

CHAPTER 2 LITERATURE REVIEW

2.1 Introduction

Commercial timber trees are grown for harvest and a time inevitably comes for them to be replaced. One of the most important objectives of intensive forest management is to achieve full expression of the productive potential of a site. This objective can only be achieved through successful forest regeneration. The two most obvious measures of regeneration success are: 1) the post-planting survival, and 2) the rate of early growth of trees. Unsuccessful tree planting increases the direct regeneration cost and causes losses in timber value and volume yield due to delays in growth (Albert *et al.*, 1980).

Treeless areas are common and reforestation is also needed to remedy the effects of previous misuse or natural accidents. One of the defining events of the past century was the astonishingly rapid decimation of tropical forests. An estimated 350×10^6 hectares have been deforested, and another 500×10^6 hectares of secondary and primary tropical forests have been degraded (ITTO, 2002). One response to this process of forest degradation has been to undertake some form of reforestation. This process should also help restore degraded landscapes. Improved methods of regrowth management and reforestation are thus critical to speed up and to facilitate this process (Lamb *et al.*, 2005).

Young vegetation is so sensitive that what is done during the period of regeneration or establishment determines most of the future development of trees and stands. The events or treatments carried out during the first few weeks or months can govern the future characteristics more than even the most strenuous subsequent tending (Smith *et al.*, 1997).

2.1.1 Eucalyptus (genus *Eucalyptus*)

It is likely that the exact number of eucalypt species is not known since different literature sources do not agree on the known number of species identified so far. However, it is probable that the genus *Eucalyptus* comprises of some 500 species of evergreen trees and shrubs, found chiefly in Australia, with a few extending to New Guinea, eastern Indonesia and Mindanao. None are native to New Zealand. They range in height from less than 1 m to over 100 m in *Eucalyptus regnans*, known in Victoria as the Mountain Ash (the tallest of flowering plants, although surpassed by the coniferous *Sequoia sempervirens*, the Californian Redwood) (Johnson, 1981). The name “Eucalyptus” has its roots in two Greek words: *eu* (well) and *kalypto* (to cover), referring to the calyx which forms a lid over the flowers in bud (Coombes, 1985).

2.1.2 Silvicultural treatments and regeneration systems

The term silviculture is derived from the Latin words *silva*, meaning “forest” and *culture*, meaning “culture”, with the latter word referring to the rearing or development of, in this case, a forest (Webster’s New Encyclopedic Dictionary, 1994).

Silvicultural treatments can be divided into two categories each with its own terminology. (1) Methods of reproduction refer to treatments of stand and site during the period of regeneration or establishment while (2) tending or intermediate pruning refers to treatments at other times during the rotation. The regeneration period begins when preparatory measures start and it ends when young trees, free to grow, are dependably established in acceptable numbers. The rotation is the period during which a single crop or generation is allowed to grow. The act of replacing old trees, either naturally or artificially is called regeneration or reproduction and these two words, which are synonymous in this usage, also refer to the new growth that develops (Smith *et al.*, 1997).

Thus, silvicultural treatments go beyond those necessary for securing regeneration. Other treatments are applied after the stand is established and also during the long period that elapses while the crop grows through various stages until it is ready for replacement. Various intermediate cuttings or tending operations are all conducted to improve the existing stand, regulate its growth, and provide for early financial returns, without any effort directed at regeneration. These treatments are

sometimes referred to as “stand improvement operations” or “timber stand improvement” (TSI), when they yield no products (Smith *et al.*, 1997).

Silvicultural interventions will impact on stand composition and growth. This impact however is most prominent during forest regeneration. The most commonly used regeneration systems are (i) natural and (ii) artificial regeneration.

Natural regeneration methods rely on establishing and releasing propagules of both seed and of vegetative origin from sources within or adjacent to the stand being regenerated. In these methods, forest canopy manipulations are carried out to favor or discourage certain kinds and amounts of propagules. These manipulations give an indication of the type and size of a natural disturbance that achieves the same results (Smith *et al.*, 1997).

Artificial regeneration methods rely on sowing seeds or more frequently on planting seedlings or cuttings. Seedlings are young trees established from seeds in nurseries and then transplanted to the field. Planted trees are not always grown from seed. Cuttings are produced by rooting sections of stems, branches, leaves or roots. Important advantages can accrue from bypassing sexual reproduction. In the first place, asexual regeneration by vegetative propagation is the only sure way of perpetuating trees with the same genetic qualities as the parent. Secondly, if cuttings form roots readily, it may be much easier, and give quicker results, to plant these rather than to grow seedlings. Even if it is difficult to produce rooted cuttings, it can still be advantageous to do so in order to multiply a few individuals with highly desirable genetic characteristics.

With some species, such as the cottonwood poplars, one can merely plant long pieces of thin shoots and depend on most of them to form roots and become established (Mc-Knight, 1970). However, most tree species are not propagated from cuttings as simply or easily. In fact, for most species it is so difficult that it is done mainly for multiplying valuable ornamentals, fruit trees, and trees for seed orchards. Such work is usually done in greenhouses with the use of rooting hormones such as indoleacetic acid (Smith *et al.*, 1997).

In micropropagation, rooting of microcuttings is often problematic. Losses at this stage have vast economic consequences. In conventional propagation by cuttings, many genotypes are also recalcitrant to root (Hartmann *et al.*, 1990). Thus, research on adventitious root formation is highly important from a practical point of view. At the same time, adventitious root formation is a fascinating scientific subject matter (De Klerk, 2002).

Plantation forestry in South Africa comprises of the cultivation a number of species within three genera of trees, namely, *Eucalyptus*, *Pinus* and *Acacia* (Wattle). Planting stock of all three genera is produced from seed and from rooted cuttings though rooted cuttings of *Acacia* species in an almost negligible amount (Table 2.1).

Table 2.1: Estimated numbers of seedlings and cuttings produced annually by major nurseries in South Africa (Zwolinski and Bayley, 2001)

Plantation Type	Seedlings (millions)	Cuttings (millions)
Eucalypts	110	16
Pines	105	2
Wattle	10	-

2.1.3 Current principles of plantation regeneration in South Africa

Unlike the silviculture of indigenous forests in South Africa, which is designed to preserve forest cover and enhance natural processes, the silviculture in plantation forestry is designed for profit making. Therefore, it is not surprising that much of the regeneration decision-making takes place before the nursery stage and, for all practical reasons, product-based tree selection pre-determines the breeding strategy and the silvicultural systems which follow (Denison and Kietzka, 1993). Efforts are made to integrate breeding and nursery technology with site selection, protection against pests and diseases and enhanced tolerance of trees to frost and drought to optimize product output.

For these reasons, a successful regeneration system can no longer be determined in the traditional way, such as ground cover by young trees, or survival of trees planted, or even volume of timber produced. Instead, it has to be determined with measures such as yield or value of product, e.g. pulp (Clarke, 1999), or a cost/benefit ratio of water used at the end of a production cycle (Zwolinski and Bayley, 2001). The concept of a designer tree includes aspects of genetics, planting stock type and planting stock quality (Menzies, 1994).

2.2 Factors that influence planting stock quality

2.2.1 Introduction

Zwolinski and co-workers (1994) stated that one of the most important objectives of intensive forest management is to achieve full expression of the site productive potential in the quickest possible way. This objective can only be achieved through successful forest regeneration. The two most obvious measures of regeneration success are: 1) the post-planting survival, and 2) the rate of early growth. Unsuccessful tree planting increases the direct regeneration cost and causes losses in value and volume yield due to delays. Regeneration success depends largely on successful matching of planting stock with planting site environment. Adaptation of planting site environment is often neither economically attractive nor a practical alternative (Albert *et al.*, 1980). Therefore an alteration of seedling suitability for planting remains the best option and systems that grade plants to eliminate unsuitable seedlings before planting are often put in place. The function of grading is to classify planting stock in categories within which seedlings should perform similarly in the field under specific planting conditions.

According to Sutton (1979), seedling quality can be defined as “fitness for purpose” or more specifically in terms of survival, growth rate, ability to compete, and to counteract stresses, pests and diseases. The purpose of quality measurement is the achievement of management objectives.

McCubbin (1992), states that seedling quality is evaluated in order to:

1. determine the quality of nursery stock and thus assess its regrowth potential;
2. cull seedlings with low performance potential, thus avoiding costly replanting and infilling;
3. match seedlings to suitable planting sites;
4. aid in research and improve nursery cultural and outplanting practices.

Initially grading systems were based only on morphological attributes. Already a century ago, height was suggested as a single factor for seedling classification (Schmidt-Vogt, 1981). In 1948, Wakeley (1949) proposed physiological grades as being superior to morphological grades. Today, quality standards are represented by multiple factors defined by means of multiple criteria. Ritchie (1984) grouped attributes of seedling quality into two categories: 1) material attributes, both physiological and morphological, and 2) performance attributes, i.e. measures of performance of the seedlings when subjected to specified test conditions (Figure 2.1).

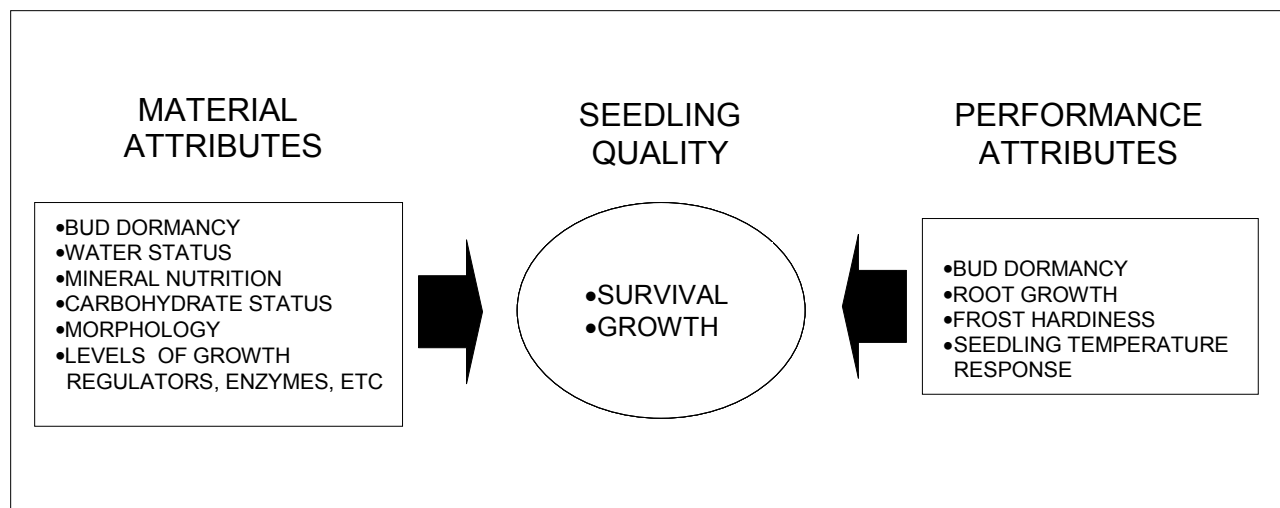


Figure 2.1: Attributes of seedling quality (Ritchie, 1984)

However, no physiological grades have been developed so far that can predict the potential for growth better than morphological characteristics (South, 1987). Classification of seedlings in European countries is based mainly on morphological characteristics, and international standards

have been defined and legislation adopted by different countries to meet them (Schmidt-Vogt, 1981). Morphologically-based grading results from an assumption that physiological quality is best expressed by morphological attributes (Parviainen, 1986). This assumption might be of practical importance where sufficient time allows physiological stresses to develop into visible symptoms before grading. But even then an interpretation of the symptoms remains vague without specific physiological tests. As the physiological state of seedlings cannot be easily defined by direct measures, some countries adopted highly sophisticated cultural and handling procedures aimed at seedling uniformity (Sutton, 1979).

In conclusion it is important to note that though a reliable means of grading seedlings would prove useful to the industry, experimental results generated from the outplanting success of graded seedlings have, in the past, not always given rise to the anticipated results. Contradictory conclusions of this kind may be due to a lack of knowledge about the interaction between the seedlings and their planting site environment. The ideal seedling grading criteria for a batch of seedlings should thus be determined using climatic, soil and topography data of the site that is to be replanted as well as morphological and physiological seedling characteristics that are known to promote outplanting success (Zwolinski *et al.*, 1994).

2.2.2 Material attributes

2.2.2.1 Physical characteristics

2.2.2.1.1 Seedling height

Seedling height is very easy to observe. Seedling height is also one of the more variable morphological measures (Rietveld, 1989) and is not consistently correlated with survival since it may even be negatively correlated in some cases (Thompson, 1985). Seedlings can be culled both for being too tall and too short.

Very tall seedlings are inconvenient to plant, have poor root:shoot ratio, and are susceptible to wind damage. However, tall seedlings may be desirable as they are often genetically superior (McCubbin, 1992). They also tend to maintain their size advantage over time as compared to smaller container

seedlings (Sutherland and Day, 1988; Simpson, 1994; Kope *et al.*, 1996). Furthermore, large seedlings can be topmowed, which makes them plantable, and as improves uniformity in the tray or seedbed (McCubbin, 1992).

Studies on the relationship between seedling height and growth increment after outplanting have also yielded interesting information with significant silvicultural implications. While the absolute growth of larger stock may only be one year ahead of smaller stock (Simpson, 1994), the competitive advantage may result in the need for less vegetation and browse control measures. In addition, survival of large stock can be higher relative to smaller stock (Sutherland and Day, 1988).

A positive correlation between height increase and initial height is frequently observed (Thompson, 1985), but is not a consistent phenomenon. Sutton (1983) noted that *Pinus banksiana* Lamb. (jack pine) and *Picea glauca* (white spruce) seedling size at outplanting was poorly correlated with subsequent first and second year height.

2.2.2.1.2 Sturdiness ratio

This is generally used as an indicator of a seedling's ability to withstand physical damage and it is calculated as follows: $Height (cm) / Collar Diameter \times 10$. It can be particularly important for container grown seedlings, where the sturdiness quotient can get very high on spindly stock (Thompson, 1985).

2.2.2.1.3 Root volume

Measurement of seedling root volume became popular in the mid-1980s as a means of evaluating seedling root system size. Seedling root volume can be assessed non-destructively using the water displacement method (Burdett, 1979). A drawback associated with this method is that it does not differentiate between fine and coarse roots, and therefore has limited capacity to characterize root system architecture (Thompson, 1985). Several studies (Rose *et al.*, 1991a, 1991b, 1997; Jacobs *et al.*, 2005) found positive relationships between seedling root volume and field survival and/or performance. However, it must be noted that other studies have yielded different results. With high moisture stress seedlings with larger root systems may not necessarily have greater capacity to

alleviate water stress following transplanting. Seedlings with larger root volumes could initially be at a disadvantage following transplant because of corresponding greater leaf area, which acts to increase water loss due to higher transpiration demand. This suggests the importance of coordinating seedling root system specifications with the intended outplanting site characteristics (Davis and Jacobs, 2005), and this is especially true for a country such as South Africa which frequently experiences extended periods of low rainfall.

2.2.2.1.4 Root system fibrosity

A fibrous root system has a high water and nutrient absorption area and a large number of active root tips, which benefits seedling establishment (Thompson, 1985; Deans *et al.*, 1990). However, root system fibrosity is a poorly defined term at present, and therefore it has been difficult to develop a standardized assessment of this characteristic.

Tanaka, *et al.* (1976) described fibrosity as the percentage of root dry mass represented by lateral roots. Using another strategy, Kainer and Duryea (1990) counted total lateral root length for all lateral roots ≥ 2 cm in length, sampled 10% of these roots, and then counted the number of root tips. Deans and co-workers (1990) described fibrosity in terms of the number of higher-order lateral roots per seedling.

While these studies all found that greater root system fibrosity resulted in better field performance, the lack of a standardized approach makes fibrosity a difficult test to apply. Furthermore, the tedious and time-consuming nature within the range of present-day fibrosity assessment techniques provides little potential for transfer to operation. Thus, development of a more rapid and standardized assessment of fibrosity would allow for better and more frequent application of this method. Such a system, based on inputs including species, anticipated site conditions, and above-ground morphology could help provide an accurate characterization of root system quality (Davis and Jacobs, 2005).

2.2.2.2 Physiological characteristics

2.2.2.2.1 Structural and non-structural carbohydrates

According to Kramer and Kozłowski (1979), carbohydrates (direct products of photosynthesis) constitute the primary energy storage compounds for seedlings, provide the basic carbon skeleton for the synthesis of essentially all other organic compounds, and constitute up to 75% of their total dry mass.

In discussions of seedling quality, however, the term “carbohydrate” is often used imprecisely. By strict definition, a carbohydrate is any compound that has the empirical formula $(\text{CH}_2\text{O})_n$, however a compound may contain additional elements such as nitrogen or phosphorus and still be referred to as a carbohydrate provided the principal make-up of the compound is the $(\text{CH}_2\text{O})_n$ backbone. Carbohydrates, when referred to in this chapter, are synonymous with sucrose and starch. Sucrose and starch are the carbohydrates most important to seedling quality and are what most people imply by the term carbohydrate. In more recent literature, the term total nonstructural carbohydrate (TNC) has been used to refer to ethanol-soluble sugars, primarily sucrose, plus starch which is enzymatically degraded into sucrose for colorimetric assay (Kramer and Kozłowski, 1979).

Sucrose is the primary form in which carbohydrates are translocated throughout the plant; up to 95% of translocated dry mass is sucrose. In contrast, starch is the primary form in which carbohydrates are stored and can be found in virtually all seedling tissue. Sucrose and starch are enzymatically interconvertible, and conversion appears to be controlled by sucrose concentration; high sucrose concentration favours the synthesis of starch, whereas low concentration favours starch breakdown. Noland and co-workers (2001) found that the length of new roots of jack pine (*Pinus banksiana* Lamb.) seedlings was negatively correlated with root starch content, suggesting that carbohydrate reserves were depleted to support root initiation and extension. Were this enzymatic interconversion of starch into sucrose not possible, growth would come to a standstill due to an inability to synthesize sufficient sucrose for inclusion into cellulose and other structural components enabling the required growth. Such substrate control helps ensure adequate sucrose levels for both maintenance (cellular respiration) and growth metabolism (Johnson *et al.*, 1991).

In a discussion of structural and non-structural carbohydrates within a context of seedling quality it is important to note that it is thought that root carbohydrate content, measured as root total non-structural carbohydrates (TNC), may be an indicator of seedling growth potential (Davis and Jacobs, 2005). Insufficient carbohydrate reserves during the period between lifting and the resumption of production of photosynthate can result in a loss of vigour and, in extreme circumstances, mortality (Marshall, 1985). Root carbohydrate content may serve to predict seedling survival and growth in conditions where the ability to photosynthesize is limited immediately following outplanting (Davis and Jacobs, 2005). In a study with naturally regenerated seedlings of four hardwood species, Canham and co-workers (1999) found a positive relationship between seedling carbohydrate reserves and survival, with roots representing the dominant TNC storage site for all species (Davis and Jacobs, 2005). Seedling root carbohydrate content could be a useful indicator of seedling internal reserves and the determination of a minimum root carbohydrate content (as TNC) for species prone to cycles of dieback and resprouting (i.e., *Quercus* spp.) could be especially useful in facilitating survival of these species (Davis and Jacobs, 2005).

2.2.2.2.2 Macro and micro nutrients

In addition to light, plants require water and certain chemical elements for metabolism and growth. The concentration of specific elements in plants is known to vary over a wide range. On the basis of the usual concentrations in plants, the essential inorganic nutrients can be divided into macronutrients and micronutrients. In general, macronutrients are elements that are required in large amounts, and micronutrients (“trace elements”) are those required in very small, or trace, amounts. Elements from both categories of inorganic nutrients are involved in fundamental processes that ensure not only plant growth and development but also resistance to stress and disease (Sutton, 1990; Rook, 1991). The most important of these processes are (Raven *et al.*, 1992):

1. participation of Nitrogen in catalysis of the enzymatic reactions of cells;
2. electron transport by Iron in cytochromes in the oxidation-reduction reactions in both the photosynthetic and respiratory pathways;
3. provision of physical structure at both the cellular (Silicon in cell walls) and molecular level (Phosphorus in the sugar-phosphate backbone of DNA, RNA and phospholipids and Sulphur in amino acids);

4. the movement of water through cell walls by osmosis regulated by the uptake of ions into cells essential for cellular growth and the maintenance of turgor regulated by Chlorine and Potassium;
5. maintenance of cell membrane integrity through regulation of cell permeability by Calcium.

Chiefly through fertigation, nursery managers can influence the nutritional status of seedlings which is a fundamental characteristic of planting stock (Landis, 1990). In spite of the abundant literature concerning plant nutrition (see. e.g. Epstein, 1972; Morrison, 1974; Van den Driessche, 1980b; Bigg and Schalau, 1990; Timmer *et al.*, 1991), studies correlating nutrient status with field performance are not very extensive and often contradictory (Mullin and Bowdery, 1978; Duryea and McClain, 1984; Van den Driessche, 1991). Plant mineral nutrient analysis as an indicator of seedling quality has mainly focused on the nutrients nitrogen (N), phosphorus (P), and potassium (K) (Hellmers, 1962; Hocking and Nyland, 1971; Ingestad, 1979; Puttonen, 1980; Sandvik, 1980; Van den Driessche 1980b; Venn, 1980).

In *Picea mariana* (black spruce) seedlings, net re-translocation of N, P, and K from older plant tissues to new growth was greatly improved by nutrient loading during nursery culture, indicating that translocation is driven by the magnitude of plant nutrient reserves (Salifu and Timmer, 2001).

The literature indicates that foliar nitrogen concentration shows some potential as a predictor of growth after outplanting (Mattsson, 1996). As an example of these studies, Switzer and Nelson (1963) presented results from linear regressions between foliar nitrogen content at lifting, and the height of loblolly pine seedlings three years after outplanting. They found a correlation coefficient of 0.84. In other studies with *Pseudotsuga menziesii* (Douglas-fir) and *Pinus contorta* (lodgepole pine), van den Driessche (1980b; 1984) reported poor correlation coefficients of 0.18 and 0.02, respectively, between foliar nitrogen concentration and height growth in the field. Perhaps it is because the effects of mineral nutrition on seedling physiology are complex and interacting that no consistent relationship has yet been demonstrated between seedling nutrient content and seedling field performance. There are also many other parameters besides nutrition that can affect seedling performance in the field.

In conclusion, “further research regarding the significance of root carbohydrate and nutrient reserves can help identify nursery cultural practices which tailor seedling reserves to a particular outplanting condition, resulting in improved root initiation and establishment” (Davis and Jacobs, 2005).

2.2.3 Performance attributes

2.2.3.1 Root growth potential

Root growth potential (RGP) is defined as “the quantified ability of a tree seedling to initiate and elongate new roots within a prescribed period of time in a standard environment optimized to promote root growth” (Simpson and Ritchie, 1997). This test is performed by counting the number and length of new roots or by measuring the change in root volume of a representative sample of a seedling crop over a fixed amount of time.

Root growth potential (RGP) has become a standard component of seedling quality assessment in many operational nurseries (Dunsworth, 1997) and is the most common physiological measurement performed on seedlings in North America (Simpson and Ritchie, 1997). Thorough reviews by Ritchie and Dunlap (1980) and Simpson and Ritchie (1997) cover this topic in detail. While RGP is not a measure of actual physiological status, it is a representation of the expression of multiple physiological parameters, including dormancy status, carbohydrate content, nutrient status, and moisture content, under a given set of environmental conditions.

Despite its widely accepted use in seedling quality assessment, RGP is not considered to be an ideal test for predicting outplanting success. Many of the questions regarding the acceptability of RGP as a measurement are based on the difference between the optimal conditions of the test and the actual outplanting conditions (Binder *et al.*, 1988; Simpson and Ritchie, 1997). Root growth potential describes seedling performance potential, rather than performance, which is an important distinction, as field conditions are rarely optimal for seedling root growth. Folk and Grossnickle (1997) propose the use of limiting environmental conditions in RGP testing as a means to overcome this problem, whereby actual field conditions are simulated. In addition to concerns over the applicability and accuracy of this method for evaluating seedling physiological status, RGP testing does not allow for

rapid decision making by the grower since seedlings must be placed in the testing environment for at least 7 days (Sutton, 1990; Sampson *et al.*, 1997).

2.2.3.2 A root-bound index

Root-bound planting stock produced from both seed and cuttings have been a concern of nursery managers for more than four decades. A root-bound root system typically is one which has the appearance of being clearly too large for its container and has as a consequence become matted and tangled. This situation usually arises if the plant is left to grow for too long in the same container. South and Mitchell (2006) define it as: a plant grown with roots too large for its container resulting in a reduction in field performance or RGP. Therefore, age *per se* might not be the critical factor that affects the quality of container-grown stock. Instead, it might be a root mass/container size relationship that influences performance.

When container-grown seedlings are root-bound (or potbound), survival, growth or stability might be reduced after outplanting (van Eerden and Kinghorn, 1978; Hulten, 1982; Lindström and Rune, 1999). It is thought that this is largely due to the inability of root-bound root plugs to readily produce new roots soon after outplanting.

A root-bound index (RBI) can be determined subjectively by scoring the appearance of the root-plug or objectively by measuring a parameter such as root mass. In addition, the RBI can be determined with destructive and with nondestructive measures. A RBI that is both objective and non-destructive is the most desirable and it is the RCD that is usually determined as the measured seedling attribute since this is often related to root mass. When using RCD two kinds of RBI can be determined:

- (i) $RBI_{\text{diameter}} = (\text{RCD in mm} \div \text{Cavity diameter in mm}) \times 100$
- (ii) $RBI_{\text{volume}} = (\text{RCD in mm} \div \text{Cavity volume in cc}) \times 100$

The RBI_{diameter} value is expressed as a percentage but, since the units in the denominator and numerator are not the same, the RBI_{volume} index is expressed as a whole number.

There is evidence that a RBI can be useful in the evaluation of stock quality prior to outplanting by nursery managers and regeneration foresters. However, though research has been conducted using pine species, additional research needs to be conducted to determine if a RBI threshold is useful for evaluating stock quality of non-pine genera.

2.3 Tree roots

2.3.1 Root systems and their development: Root morphology and root deformation

The importance of differences in root system morphology in the ability of forest trees to withstand environmental stress is not well understood. Natural root systems themselves vary widely in depth, lateral spread, straightness and other factors. Furthermore, planted seedlings have different root system morphologies from natural ones (Harrington *et al.*, 1989).

The first step in determining the significance of root system morphology is to quantify it. The parameters of interest concerning lateral roots are their number, spatial distribution, size, kinkiness, and degree that they coil around the root stock, or primary root. The size and orientation of the root stock itself are of interest as well. These parameters have potential biological significance: Hay and Woods (1968, 1975) speculated that kinks in roots, which provide physical resistance to carbohydrate flow in the cambium and result in carbohydrate accumulation will increase lateral root initiation above the kink. Hultén and Jansson (1977) demonstrated that root systems with a higher percentage of crooked laterals provided less stability than those with more straight roots. Derr and Enghardt (1957) found that trees that had blown over during Hurricane Audrey had root systems lacking laterals on the windward side of the tree. Finally, Ball (1976) observed that coiling lateral roots resulted from planting stock becoming root-bound during the production phase in the nursery and that stems of such container stock snapped off above these coiling lateral roots.

2.3.2 Methods for analyzing root systems in terms of shape and size

Jourdan and Rey (1997) stated that “root architecture is a fundamental aspect of plant productivity through its functional importance in the efficient acquisition of soil resources. It is therefore a

subject of considerable interest in agriculture and ecology. Root system architecture is directly linked to the various functions ensured by the roots.”

The architecture of a branched system can be perceived through architectural analysis (Jourdan and Rey, 1997). This analysis was first conducted on the aerial system of tropical trees, and it was based on studying how the meristems of each axis function and how hierarchical relations are established between these axes. It also enables one to carry out a complete diagnosis of the different root types making up the branched system, and show the growth, branching, differentiation and mortality processes, which are characterized for each category of axes identified. This analysis led on to the concept of the root architectural unit (Atger, 1992). Architectural analysis is therefore an appropriate tool for measuring the root architecture of plants. With practical applications in mind, it can also serve as a back-up for modeling and quantification of the various processes identified (Jourdan and Rey, 1997).

2.4 Root pruning

2.4.1 Definition

It is a commonly held view that trees of the same species will have the same root form. This is not found in practice. Even without nursery and transplanting effects, the root form within a single species can vary tremendously depending on the physical environment encountered by the roots (Wagg, 1967).

While root form is species and site dependent, various typical deformities are known to occur. Kinked or “J” roots are generally caused by poor transplanting technique (Donald, 1986). This may occur in the nursery when very young seedlings are transplanted into empty cells, known as “gap filling” or “pricking out”. Excessive firming-in and the use of long, thin containers often lead to field-induced deformities (Herbert, 1981; Persson, 1981).

Root pruning of transplants is an established practice in horticulture (Poincelot, 1980) and silviculture (Evans, 1982; Geisler and Ferree, 1984). In open ground or bare-root stock, root pruning

forces the development of a fibrous root system. A higher proportion of roots in the root ball makes for faster and more successful transplanting compared with unpruned stock (Watson, 1986). In container stock where roots have become coiled, pruning of these roots at the time of transplanting will correct the root form (Bell, 1978; Stone and Norberg, 1978; Watson, 1986). Modern nursery systems usually incorporate root pruning to prevent the formation of circling roots.

2.4.2 Mechanical pruning

Undercutting is standard practice in many forest nurseries throughout the world as a method of conditioning planting stock to withstand transplanting (Goor and Barney, 1968). It is done while seedlings are growing in seedbeds, or while lining out beds, by a tractor-mounted sharp blade being drawn beneath the bed to undercut the roots of seedlings at a predetermined depth, usually 7.5 – 10 cm below ground level. Apart from the disease risk caused by wounding, this option requires another regular nursery operation which may or may not be acceptable (Nelson, 1989). Physical cutting of roots in two directions using scissors or a sharp blade has been suggested and implemented when using non-compartmentalised containers (Burdett, 1981; Örlander, 1981) such as polypot containers (Marsh, 1978).

2.4.3 Chemical pruning

Most early work on evaluating the effect of copper compounds was carried out in the U.S.A and Canada, and was primarily concerned with coniferous forestry species such as pines and spruce (Beeson and Newton, 1992). Early problems focused on how to apply the compounds to nursery containers. The most common method of application now in use is to incorporate the copper compound into latex or acrylic paint, and to paint or dip the compound onto the inside wall of the nursery container (Gordon and Hayes, 1994).

2.4.4 Air pruning

Air pruning, though a well-established concept and commonly observed phenomenon, is not a term for which a definition is readily available in the literature. That is so for the literature on production

of planting stock for the forestry industry as well as for the literature published on the production of crops in horticulture and floriculture.

Walker (2005) states that air pruning happens naturally when roots are exposed to air in the absence of high humidity. Roots are effectively “burned” off, causing the plant to constantly produce new and healthy branching roots. Roots that are not exposed to air continue to grow around the container in a constricted pattern resulting in root deformations observed in the absence of other strategies available to counter root deformations of containerised planting stock. Air pruning promotes the development of a healthier, more highly branched root structure which ultimately allows the plant to more efficiently take up water and nutrients while increasing growth and overall plant health (Walker, 2005).

2.4.5 Benefits of root pruning practices

Root pruning of transplants is an established practice in horticulture and silviculture (Geisler and Ferree, 1984; Evans, 1982). According to Nelson (1989), a nursery system which incorporates adequate root pruning minimizes the risk of root malformation caused by poor handling by inexperienced field staff. He further states that “root pruning of containerised forest stock should be viewed as a means of minimizing the risk of failure”.

2.5 Growing media

2.5.1 Introduction

Good plant development depends largely on the growing medium used. If a plant develops a good root system in a well-balanced substrate, this does not mean that the plant is pampered and will not adapt to the harsh life outside a nursery. In fact, the opposite applies. To survive in the harsh environment of a field, often without additional watering and fertilizing, a plant needs a well-developed and strong root system. The development of a healthy root system depends not only on the genetic properties of the plant but to a large extent on the physical and chemical properties of the substrate used (Jaenicke, 1999). Growing media serve four functions: 1) to provide water, 2) supply nutrients, 3) permit gas exchange to and from the roots and 4) provide support for the plants (Reed, 1996).

“The general suitability of plug media for water and mineral absorption is determined by the nature of the open spaces, or pores, between the solid particles of the media. It is in the soil matrix that the plant’s roots grow and where water and air are stored and move. Both total pore volume (porosity) and pore size distribution are important. Porosity, or pore volume, determines the potential water-holding capacity, and pore size distribution determines the actual water retention and air space after irrigation and drainage. Suitable growing media must maintain a balance between water retention and aeration. Excess water retention means plants will suffer from poor soil aeration, and excess aeration means plants are likely to experience water deficit. Under conditions of rapid plant water use (low humidity, high light, relatively large size), less air porosity is generally required than under conditions of low water use (high humidity, low light, small size)” (Styer and Koranski, 1997). In addition to providing a balance between water retention and aeration, a root medium also serves as a reservoir for plant nutrients and finally it must provide anchorage or support for the plant.

2.5.2 Types of growing media

Most media today are blends of two or more components. The chemical and physical properties of the resulting medium are not always equal to the sum of its parts. When greenhouse media are blended, the chemical and physical properties of the components are “married” to each other to form new properties that are different from the individual components (Reed, 1996). According to Hanan

(1998), one of the greater difficulties in handling modified soils and artificial substrates is predicting the outcome. In fact, Johnson (1980) states that there are nearly as many methods for assessing composts as there are researchers.

2.5.3 Growing media characteristics

2.5.3.1 Water holding capacity

Container capacity, according to Reed (1996) is the percent volume of media or media component that is filled with water after saturating media and allowing it to drain. It is the maximum amount of water (or capacity) media can hold. Since drainage is influenced by media height, this property is dependent on container size and shape. The taller the container, the more drainage and the less capacity media will have to hold water.

2.5.3.2 Bulk density

Bulk density can be regarded as the opposite to the total pore space of a soil and can thus also be interpreted as a measure of how 'heavy' the medium is. It is defined as the mass in grams of a dried cubic centimeter (cm^3) or milliliter (mL) of soil. The more pore space there is in that mL the less will be the bulk density. Also, as the density of the actual particles increases, bulk density will also increase (Handreck and Black, 1994).

2.5.3.3 Air filled porosity / Water filled porosity

For most plants to grow in soil or a medium, a proportion of the pore space needs to be gas-filled. The gas-filled proportion of the pore space allows the influx of oxygen (O_2) into, and the efflux of carbon dioxide (CO_2) from the soil. The consumption of O_2 in the soil and production of CO_2 is because of the oxidative demands of plant roots, microorganisms, and chemical reactions. The status of a soil in relation to the proportion of gas-filled pores and the concentration of O_2 and CO_2 in the soil is often referred to as the aeration status of the soil (Cook and Knight, 2003).

At a physiological level, the minimum aeration status occurs when the flux of O₂ to the root surface, especially the root tip, is just able to meet the O₂ demand of the root tissues. The transport of O₂ to a plant root occurs first through diffusion from the atmosphere via the gas-filled porosity of the soil to the depth of the root, and then through a boundary layer surrounding the root (Cook and Knight, 2003).

2.5.3.4 Cation exchange capacity

The total amount of cations held by a material is called its cation exchange capacity (CEC). A common way of giving the amounts of exchangeable cations held in a growing medium is in milliequivalents, abbreviated to either m.e. or meq. The cation exchange capacities of potting media are usually given on a volume basis – for example, 65 m.e./L (Handreck and Black, 1994).

There are seven main cations involved in plant growth – H⁺, Al³⁺, Ca²⁺, Mg²⁺, K⁺, NH₄⁺ and Na⁺ - together with many others present in tiny amounts (Handreck and Black, 1994).

Cations, being positively charged, are attracted to the negatively charged surface of colloid particles, where they are held loosely. Often they exchange places with other cations in the soil water nearby. The capacity of the particles to hold cations is limited to the number of negative charges on their surfaces. An increase in the concentration of a cation in the water in a medium will allow that cation to displace other cations from colloid surfaces (Handreck and Black, 1994).

2.5.4 Other factors that influence plant growth

2.5.4.1 Pathogens and deficiencies of management

There are numerous pathogenic diseases of greenhouse crops. They come under four general categories: the presence of either viruses, bacteria, fungi or nematodes. It is important to know the characteristics of each as well as the life cycles of specific pathogens in order to determine how to control them. Some require free water on the plant foliage in order to develop. Others require a very wet root medium. Each of these requirements suggests ways of controlling the pathogen (Nelson, 1985).

2.5.4.2 Growth-stimulating additives

“Nursery growers and field foresters aim to achieve ‘free-to-grow’ status in plantations while being mindful of increasing restrictions on silvicultural tools such as chemical vegetation control and controlled burning for site preparation” (Rose and Morgan, 2000). Haase and co-workers (2006) state that the use of slow-release fertilizers (SRF) has gained recognition as an important tool to meet reforestation objectives. Incorporating SRF directly into the growing medium is a relatively new approach to seedling nutrition in container forest nurseries. They further state that, until recently, there was concern that placement of fertilizer in direct proximity to the seedling root system would result in toxicity but that with improved SRF technology many nurseries are now routinely incorporating SRF into the growing medium.

Seedling field establishment and growth can be enhanced by SRF fertilization at the time of outplanting (Carlson, 1981; Carlson and Preisig, 1981; Arnott and Burdett, 1988; van den Driessche, 1988) or by nutrient loading in the nursery prior to lifting (Malik and Timmer, 1996; Irwin *et al.*, 1998). Slow release fertilizers incorporated into the nursery growing medium has the potential to accomplish both of these by providing a means for nutrient loading in the nursery as well as continued fertilization after outplanting (Haase *et al.*, 2006).

2.5.5 Use of industrial and agricultural by-products as potential growing media

Sawdust of some tree species makes an acceptable component of container growth media. However, a great deal of variation exists and therefore caution is the watchword. Sawdust of pine, spruce and most hardwood tree species (oaks, maples, sycamore, cottonwood, ash and others) must be composted prior to use because of the high carbon to nitrogen ratio. The finer the sawdust, the more rapid the decomposition and the greater the quantity of nitrogen fertilizer required (Whitcomb, 1984).

2.6 Nursery containers

2.6.1 Historical perspective

In forest nursery practice a container plant is one raised and planted as a separately grown unit with a root system that has always been isolated from that of any other plant. The container may be single or multiple, made of rigid or flexible materials which can include clay, wood, paper, metal and plastics. Containers may have biodegradable walls that enclose a growing medium (as in the traditional flower pot) or be wall-less and composed entirely of the growing medium as in WRIBLOKS (Figure 2.2). WRIBLOKS are blocks made of composted pine bark that is held together with re-pulped newspaper (paper-maché) acting as the binding agent according to a protocol developed in the Wattle Research Institute in the 1970's (Zeijlemaker and de Laborde, 1977).



Figure 2.2: A WRIBLOK after successful establishment of a *Eucalyptus grandis* seedling in the block (Left), a waterproofed egg tray that acts as a support for 30 WRIBLOKS (Right)

Plant containers thus can be purchased in a large variety of configurations, shapes and sizes, and manufactured of various materials each with its associated advantages and disadvantages.

For many years the most common container used for plant production in the U.S.A was the #10 food can, generally referred to as a one-gallon container. These were generally cleaned somewhat of food or other debris, holes were made in the bottom or sides, and then dipped in paint to improve the overall appearance and reduce the rate of rusting stimulated by the plentiful moisture and fertilizer salts. In addition, since the container was not tapered, it required cutting down the sides before the

plant's roots could be removed. Early plastic containers were quickly weakened by ultra-violet light and became brittle and nearly as unattractive as the rusty metal cans. Fortunately, many advances have been made in the quality of plastic containers. Today, they generally remain attractive and functional throughout the production and retail cycle and add to the sales appeal of packaging of the plant and in many cases can be re-used after the plant is removed and planted in the landscape (Whitcomb, 1984).

The cost of the container adds significantly to the cost of plant production. In many cases, the cost of the container is second only to labour in terms of the largest proportion of container plant production cost. However, the neatness, ease of handling and shipping and retention of all roots, thus reducing or eliminating plant shock loss when transplanting, helps justify the cost.

2.6.2 Container types

According to Barnett and Brisette (1986), there exist essentially three types of containers: tubes, plugs and blocks. However, not all containers used for the production of planting stock of forest trees fit neatly within one of these three categories. A good example of this is conventional polyethylene pots. A more comprehensive system of categorizing containers would need to include the following container characteristics:

1. Re-usability;
2. Biodegradability;
3. Filling requirement;
4. Penetrability of the walls of the container by the roots.

For the purpose of this review Barnett and Brisette's categorization of containers into the three categories stated above will be adopted since this is the most commonly used method of placing different containers within broadly-defined types of containers.

2.6.2.1 Tubes

These containers (Figure 2.3) have an exterior wall, require filling with a growing medium, and the seedlings remain in the container for outplanting (Barnett and Brissette, 1986). The primary advantage of tubes is wall rigidity, providing both ease of handling as well as sufficient impermeability to prevent desiccation when planted in dry soil (Day and Cary, 1974). The major disadvantage of tubes is slow egress of roots into the soil because initial contact with the soil is made primarily through the bottom of the container (Barnett and Brissette, 1986). This system is ecologically unfriendly in that the tubes remain in the soil after outplanting and are essentially non-biodegradable. Biodegradable plastic is now commercially available but the use of such plastics for the production of plants has not yet been implemented on a large scale and its use for this purpose has potential for further investigation.



Figure 2.3: Seedlings produced in plastic tubes and supported in polystyrene trays

2.6.2.2 Plugs

A plug is a containerized transplant (Styer and Koranski, 1997). Seed is mechanically sown, or cuttings are placed, into individual cells in a plug tray (Figure 2.4).



Figure 2.4: Two plastic trays used extensively by large forestry companies in South Africa, the Unigro 128 (Black) and the Sappi 49 (White) tray. The Unigro 128 tray carries 128 square but tapered cells that are held in place individually by an outer carrier tray allowing consolidation of the cells to ensure the dispatchment of full trays to the field. The Sappi 49 tray is white in colour and consists of 49 conical-shaped cells

After germination the seedling grows in its own miniature container until ready to transplant. Each root system is totally self-enclosed, resulting in optimum root growth with plenty of root hairs. When transplanted the plug is simply pushed or pulled out of the cell and placed into a larger container or the field. Little if any damage occurs to the root system, so losses from rot are greatly reduced and growth is very even (Styer and Koranski, 1997).

The production of plugs in solid-wall containers is the system adopted in the large majority of South African nurseries, not only for the production of forest planting stock but also for the production of all other types of nursery-grown vegetables and ornamental plants. The containers in which the plugs are produced are typically manufactured from either plastic or polystyrene.

2.6.2.3 Blocks or Wall-less plugs

Block designs incorporate advantages of both tubes and plugs. The block itself is both the container and the growing medium. Seeds are sown, or cuttings placed, in the block and the entire package is transplanted into the soil. Blocks are usually rigid enough for mechanized planting but still allow rapid root egress upon outplanting. The advantages of the use of blocks include: (1) simplified greenhouse operations because no filling is required, (2) no root manipulation into undesirable patterns or constraint after outplanting, and (3) adaptability to mechanized planting equipment. After outplanting, root egress occurs from the entire block surface and no unusual patterns of root development are evident that might cause future problems in seedling growth or stability. It is the ability of roots to egress from the entire block surface that provides this container system with its wall-less nature. A disadvantage of the system is that plants are subject to some root cross-over from one block to the next if seedlings are held for long periods (Barnett and Brissette, 1986). Examples of true blocks of this type are the original WRIBLOK developed by the Wattle Research Institute in the 1970's and the wall-less plugs produced and tested in this dissertation using the same protocol as the one used for the production of the WRIBLOK.

Modifications of the above type of true blocks are, usually cylindrical, blocks of growing medium that are held together by a supporting sleeve that is penetrable by the plant roots. The best established block-type of containers of this kind, that are commercially available, are different size cylindrical blocks of sphagnum peat extracted from specially selected bogs and manufactured, since 1959, by the Danish company Jiffy Products (Figure 2.5). The blocks are marketed under the term "pellets" and are bound by a thin biodegradable net which provides added structural integrity to the block or pellet. The Jiffy pellets expand to several times their original size when imbibed with water. This reduces their volume for the purpose of cost-effective transport from the manufacturing site to the customer.

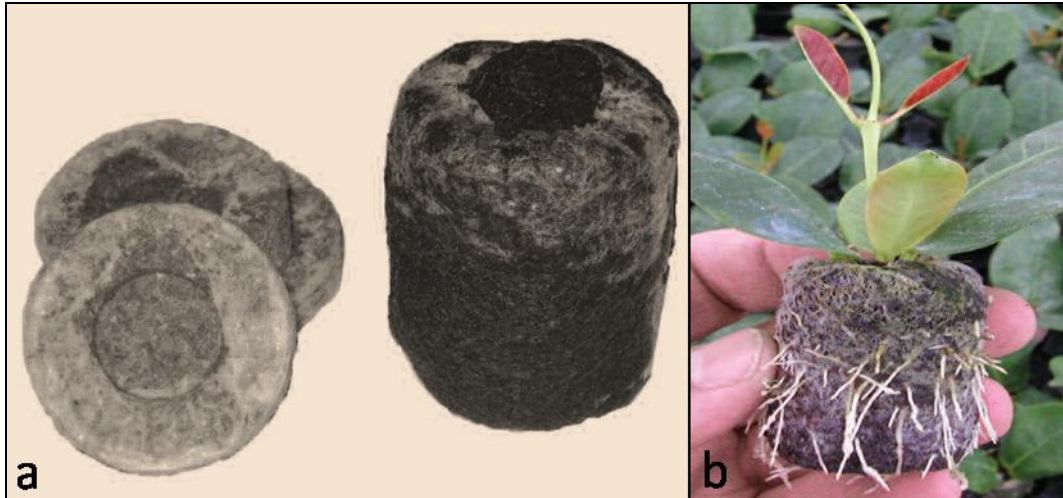


Figure 2.5: (a) Jiffy pellets, dry (left) and after imbibition with water (right) and (b) a Jiffy plug with a plant. The wall-less nature of the plug is clearly visible by the roots that are able to grow through the fine net into the surrounding air

Similar to the Jiffy pellets are blocks produced by a complex machine that can be purchased from another Danish company that markets its products under the trade name “Ellegaard”. The Ellepot blocks, produced by the Ellegaard machine, are made of a degradable paper filled with growing media (Figure 2.6). The paper keeps the roots air-pruned during the propagation cycles and facilitates root penetration after planting in the field. “The world patented system is unique, because it handles the media so gently that the airy and fragile structure is maintained throughout the production cycle” (Ellegaard, 2010). An advantage that the Ellegaard system has over the Jiffy pellets is that only the paper needs purchasing and importation from the mother company as opposed to the ready-made Jiffy pellets requiring purchasing and importation. In addition to this the Ellegaard system is flexible in terms of the growing medium that it can accommodate: “Ellepot can be produced with many types of growing media: peat, coco peat, perlite, pine bark, vermiculite and rock wool” (Ellegaard, 2010).



Figure 2.6: Two Ellepots supported by a tray (Left). The Ellepots are plugs mechanically filled by a machine that fills a paper sleeve with growing medium and cuts up the filled sleeve into plugs of equal length (Right)

The Dutch company Heto Agrotechnics produces a machine (Figure 2.7) identical in principle to the machine manufactured by Ellegaard for the large-scale production of biodegradable, wall-less, sleeved blocks.



Figure 2.7: The Heto Agrotechnics machine that produces blocks similar to Ellepots

It is likely that the Ellegaard machine would be in operation in South Africa today were it not for the cost of the specialized paper that provides the exterior support of the plugs (M Kruger pers.comm., Top Crop Nursery, 2010)*, though it has successfully been implemented in Forestry companies in Africa. It is for the same reason that Forestry companies do not import and use Jiffy pellets for the production of planting stock. The excessive cost of the pellets and their transport makes it impossible for companies to use this product. Improved root architecture but above all, a more streamlined operation in that the return of empty containers is no longer necessary after the introduction of blocks as the type of container used in nurseries, are the two considerable advantages of this container type over other container systems.

2.6.3 Container properties

2.6.3.1 Shape and size

On the Stuewe & Sons, Inc. website (a very comprehensive website depicting the vast range of plant containers available including containers for the production of planting stock for the forestry industry), the largest container available has a cavity volume of 195 cubic inches (3195 cm³) and the container with the smallest cavity has a volume of 0.3 cubic inches (4.9 cm³).

Generally, larger container volumes have greater water and nutrients availability, along with more space for root development. Consequently, larger containers generally result in better seedling growth (McConnughay and Bazzar, 1991; Hsu *et al.*, 1996) and survival (Ward *et al.*, 1981; Matthes-Sears and Larson, 1999) postplanting. The shape and size of containers commonly used for the production of planting stock of tree plantations in South Africa are listed in Table 2.2.

*Kruger M 2010 Pers. Comm., Top Crop Nursery, South Africa

Table 2.2: Dimensions of containers commonly used in South Africa for the production of planting stock of forest trees

Container	Depths (mm)	Volume (cm ³)	Cross-sectional shape
Unigro 128	98	60	Square
Unigro 98	98	90	Square
Sappi 49	80	80	Round
Poly 98 Deep	100	60	Round
Poly 98 Deep	94	78	Square
Poly 128 Shallow	61	36	Square

2.6.3.2 Characterisation of the hydrophysical and mechanical properties of pressed blocks

Good physical conditions of pressed blocks are essential since young roots must have sufficient water and air, and this must be achieved while maintaining the block's stability (Klapwijk and Mostert, 1992). Wever and Eymar (1999) undertook a study of existing methods for the characterization of physical properties of peat blocks. Raising plants in peat blocks is an established method of transplant production. In the Netherlands alone, 750 000 m³ of peat is used annually for this purpose (Van Schie, 1998). To obtain information about the stability of pressed blocks, it is important to consider also the cohesion between the substrate particles. Researchers have achieved this by considering techniques from the food industry where quality parameters like firmness, brittleness, chewiness, gumminess and stickiness are relevant (Wever and Eymar, 1999).

2.6.3 Design features to control root growth

One of the most serious problems in container tree seedling culture is the tendency of tree seedling roots to spiral around the inside of the container. Seedling roots grow geotropically, but if they do not meet any physical obstruction, they may tend to grow laterally around the side of the container (Landis, 1990).

Root spiraling will not adversely affect growth while the seedling remains in the nursery, but it can seriously reduce seedling quality after outplanting. Spiraling roots prevent the seedling from becoming properly established in the surrounding soil, which can result in frost-heaving, toppling, or

even strangulation (Burdett, 1979). The problem of root spiraling has been at least partially solved by designing containers with vertically oriented ridges, ribs, or grooves that protrude into the growing medium and present an obstacle to spiral root growth. Kinghorn (1974) recommended ribs about 2 mm (0.08 inch) high on the inner cavity wall. These ribs intercept spiraling roots and force the developing roots to grow downward to the drainage hole where they stop growing because of the low humidity and become airpruned. Most types of containers used in forest tree nurseries have some sort of anti-spiraling rib design, and one container manufacturer has even incorporated this feature into its brand name, the Spencer-Lemaire Rootrainer[®] (Figure 2.8). Root spiraling occurs in most tree species but has been most serious in pines. Girouard (1982) found that all four species of conifers grown in Quebec tubes exhibited some degree of root spiraling but that this problem was more severe still in the pine species. Even with pines, however, there is variation. Barnett and Brissette (1986) report that *Pinus palustris* (longleaf pine) is more prone to root spiraling than are *Pinus taeda* (loblolly pine) and *Pinus elliotii* (slash pine). Root spiraling and other types of abnormal root growth become more serious the longer the seedlings remain in the container and this tendency was found to be particularly significant for *Pinus banksiana* (jack pine) (Barnett and Brissette 1986).



Figure 2.8: Vertical grooves in the sidewall of the Spencer-Lemaire Rootrainer[®] container depicted above prevents root spiraling by guiding root growth downward (Landis, 1990)

2.7 Summary and problem statement

The Forestry industry in South Africa and in many other countries adopts the method of clearcutting as a means of harvesting its plantations. This leaves vast tracts of land that need to be replanted for the next rotation of crop which will again be harvested. The restocking of newly harvested plantations is achieved by the outplanting of large quantities of seedlings that are produced in large intensive commercial nurseries. In South Africa, seedlings in such nurseries are raised in plastic or polystyrene containers with individual cavities, the number of which ranges from 49 to 128 depending on the type of container used. These containers, and thus each cavity, is filled with a specially selected growing medium and then either sown with seed or planted with a cutting.

The forestry industry in South Africa faces a number of challenges as a result of the implementation of planting stock production by means of containerized nurseries. These challenges are logistical and plant quality related.

Containers by their very nature do not allow for the free development of roots. This limitation can lead to root systems within plugs becoming root-bound. This in turn can result in plants not establishing themselves readily following outplanting or it can result in trees that become susceptible to wind-throw.

Dispatch of the seedlings to the field or to holding nurseries when the seedlings are sufficiently large and hardened off is achieved by means of tight packing of the containers in which the seedlings were raised into trucks or trailers. Seedlings are removed from the container in which they were raised at the time of outplanting and placed into carefully prepared pits. Damage to the root plug is sometimes incurred when the seedling is removed from the container resulting in delays in the establishment of the seedling or in some cases death of the seedling. When all the seedlings in a batch of containers have been outplanted the grouping together of trays on the edge of the field and subsequently their transport back to the nursery must be ensured requiring substantial logistical management and adequate follow-up. The empty containers are purchased at a considerable cost and their return to the nursery is critical to ensure the cost of forest regeneration remains within the allocated budget.

It is in the above limitations imposed on the forestry industry by the currently used container system that the opportunities for improvement of this system can be found. This may be achieved from the successful development of a wall-less plug for planting stock of forest trees. Such a wall-less plug can be described as being a volume of organic or synthetic growing medium, usually rectangular or cylindrical in shape, devoid of an impenetrable wall and free-standing in which a small plant can establish itself either from seed or from a cutting.

Wall-less plugs of this kind could prove to overcome the problems outlined in the following ways:

Problem 1 – Root-bound plugs:

Wall-less plugs are not bound by an impenetrable wall and roots are thus able to grow out of the plug into the surrounding air. At that stage, the roots theoretically become air-pruned and stop growing giving rise to the development of new lateral roots from the taproot or lower order existing lateral roots as the point of origin for the development of these new roots. Wall-less plugs thus produce seedlings with root plugs that are never root-bound, are more fibrous and have a larger number of higher order lateral roots than seedlings produced in containers.

Problem 2 – Damage to the root plug upon removal from the container:

Planting stock produced in wall-less plugs are transplanted into the field without the required step of removing the plant from its container.

Problem 3 – Return of empty containers:

In the case where planting stock is transported to the field in non-degradable boxes or carry cases, these would still need to be returned to the nursery. However, since the wall-less plugs have some inherent strength it can be envisaged that plants could be left bare on their side or freestanding upright on the ground without a support of any form such that any empty carry cases used in the transport of the seedlings can be returned immediately upon delivery of the planting stock to the site of outplanting. In addition, the number of such carry cases could be less than the number of trays that currently need returning since a larger number of seedlings could probably be packed into a single carry case.

The aims of the following dissertation were to:

1. Develop prototype wall-less plugs;
2. Raise *Eucalyptus spp.* seedlings in these prototype wall-less plugs;
3. Compare the performance of the planting stock produced in the prototype wall-less plugs with plants raised in containers currently used by the industry;
4. Devise methods to qualitatively and quantitatively measure differences in *Eucalyptus spp.* seedling root systems;
5. Establish how container type impacts the shape and size of *Eucalyptus spp.* seedling root systems.

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CHAPTER 3 A MANUFACTURING PROTOCOL AND INITIAL TESTING OF A WALL-LESS PLUG FOR PLANTING STOCK OF FOREST TREES

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3.1 Abstract

Wall-less rooting plugs equal in volume and shape to one cell of a Sappi 49 container were produced using the WRIBLOK protocol. Equipment was made for the compression of a paper-maché composted pine bark mix into a mould. The equipment and protocol were improved upon to increase the rate of production of the plugs. Performance of the plugs was tested by comparing the growth of *Eucalyptus dunnii* seedlings in term of height and root collar diameter (RCD) with seedlings raised in Unigro, Sappi 49 and Poly 128 shallow containers. Data was collected on two dates four months apart and analysed by means of ANOVA. Initial seedling establishment and growth in wall-less plugs was the lowest at the start of the trial. However, growth of surviving plants in these plugs was equal to that of plants raised in Poly 128 Shallow trays by week 10 after which growth of plants in wall-less plugs surpassed performance of plants raised in Poly 128 Shallow trays. Performance of plants produced in Sappi 49 and Unigro trays was similar and significantly superior to plants raised in the wall-less paper-maché plugs and in the Poly 128 Shallow containers. Wall-less paper-maché plugs may enable production of good quality planting with improved root form without the need for large capital outlay for purchase of expensive plastic containers and the need to return such containers to the nursery after outplanting.

keywords: root plug, wall-less plug, container type, seedling height, root collar diameter

3.2 Introduction

The Forestry industry in South Africa and in many other countries practices the method of clearcutting as a means of plantation harvest. This leaves vast tracts of land that need to be replanted or restocked with appropriate tree seedlings for the next rotation of crop which will again be harvested. The restocking of newly harvested plantations is achieved by outplanting of large quantities of seedlings that are produced in large commercial nurseries. In South Africa, seedlings in such nurseries are raised in plastic or polystyrene containers with individual cavities, the number of which ranges from 47 to 128 depending on the type of container used. These containers, and thus each cavity, is filled with a specially selected growing medium and then either sown with seed or supplied with a cutting.

The production of planting stock of forest trees in containers is associated with a number of problems. These include logistical and practical problems in addition to problems related to plant quality.

- **Problem 1:** Containers by their very nature do not allow for free development of roots and can lead to root-bound plugs resulting in plants that do not establish themselves readily following outplanting. They can also result in trees that are susceptible to wind-throw even following successful outplanting.

Dispatch of seedlings to the field or to holding nurseries when seedlings are sufficiently large and hardened off is done by means of tight packing of containers in which the seedlings were raised into trucks or trailers. Seedlings are subsequently removed from the container in which they were raised at the time of outplanting and placed into carefully prepared pits.

- **Problem 2:** Damage to the root plug is sometimes incurred when the seedling is removed from the container resulting in delays in seedling establishment or in some cases mortality.
- **Problem 3:** When all seedlings in a batch of containers have been outplanted the grouping together of trays on the edge of the field and their transport back to the nursery must be ensured requiring substantial logistical management and adequate follow-up.

A possible solution to these problems is the production of planting stock in wall-less containers. Such containers are volumes of organic or synthetic growing medium, usually rectangular or cylindrical in shape, devoid of an impenetrable wall and free-standing in which a small plant can establish itself either from seed or from a cutting.

Wall-less plugs can provide solutions to Problems 1, 2 and 3 in the following ways:

Problem 1 – Root-bound plugs:

Wall-less plugs are not bounded by an impenetrable wall and roots are thus able to grow out of the plug into the surrounding air. At this point, in theory, they become air-pruned and stop growing giving rise to the development of new lateral roots with the taproot or lower order existing lateral roots as the point of origin for the development of these new roots.

Wall-less plugs thus produce seedlings with root plugs that are never root-bound, are more fibrous and have a larger number of roots than seedlings produced in containers.

Problem 2 – Damage to the root plug upon removal from the container:

Planting stock produced in wall-less plugs are transplanted as they are into the field.

Problem 3 – Return of empty containers:

1. Since wall-less plugs have some inherent strength it can be envisaged that plants could be left bare on their side or freestanding upright on the ground without support of any form such that any empty carry cases used in the transport of the seedlings can be returned immediately to the nursery upon delivery of the planting stock to the site of outplanting.
2. The number of such carry cases could be less than the number of trays that currently need returning since a larger number of seedlings could probably be packed into a single carry case.

The objective of Trial 1 was firstly to produce a prototype wall-less plug for growing eucalypt seedlings and secondly to compare their performance with planting stock raised in standard containers currently used in South African nurseries. The development of such a plug comprises sourcing potential materials, making equipment and experimenting with ratios and ingredients.

Preliminary results produced from this trial can then serve as an indicator of the potential of wall-less plugs as a possible replacement of the current container system.

3.3 Materials and Methods

3.3.1 Paper-maché mix protocol

Original WRIBLOK protocols (Zeijlemaker and de Laborde, 1977) for making a paper-maché mix were used. Newspaper was hand shredded and repulped in cold tap water in a plastic bucket using a Ryobi 550W electric drill (Model number: HID 550VR) fitted with a paint mixer at approximately 600 rpm (Figure 3.1). Composted pine bark, charcoal dust and the repulped newspaper-water mixture were blended into a thick dough using the electric drill and paint mixer according to ratios in Table 3.1.

Table 3.1: Ingredients and ratios used for the paper-maché mixture

Mass (g)			Vol (L)
Newspaper	Pine bark	Crushed charcoal	Water
300	2400	600	7

3.3.2 Wall-less plug manufacturing

The first attempt at making a wall-less plug was made using improvised tools. These consisted of a modified silicon glue hand gun, the type found in hardware stores for home improvement. Eight, 7 mm holes were drilled around the nozzle of the glue gun to allow for extra draining of excess water during the compression of the paper-maché mix. Inside the empty silicon container mould a metal gauze with 1.5 mm pores was inserted. This gauze prevented the mix from being pushed out during the compression stage. Compression was achieved with the hand-held lever and piston. Once all excess water was expressed from the system the plastic silicon cylinder was removed from the holder. The metal gauze and end stopper with the compressed paper-maché mix were all pushed

back and out of the cylinder using metal rods that were pushed through the holes around the nozzle. With careful manipulation, cylinders of paper-maché plugs were individually produced.

A number of problems were encountered with the above system of making plugs:

The process was time consuming. Loading the 'gun', compressing the mixture to expel excess water and finally carefully removing the wet plug from the gun took between five and six minutes. This equates to a manufacturing rate of 12 plugs per hour. Considering that removal of wet plugs from the 'gun' resulted in some breakages, a more realistic figure for the rate of production was 9 to 10 plugs per hour. That excludes time required for making the paper-maché mix and time required for drying of wet plugs. The high pressures required for extraction of excess water put the metal gun under such stress that it required re-enforcement.

In an attempt to increase the production rate of the paper-maché plugs, a jig was made which had the potential of producing 49 plugs at a time. The jig consisted of a Sappi 49 seedling tray which served as a mould and 49 pistons to compress the paper-maché mixture to express the excess water. The mould consisted of two parts. The first part was an unaltered Sappi 49 tray. The second part consisted of the upper section of a Sappi 49 tray which was transversally sawn off using a handheld crosscut saw. This smaller upper section was attached to the unaltered tray using plastic cable ties and allowed for the production of plugs equal in size to the Sappi 49 cavity. The 49 pistons making up the second half of the jig were bolted onto a square wooden plank. At the end of each piston was a washer held in place with a nut on either side of the washer. The washers were just slightly smaller in diameter to the internal diameter of the individual cavities of the Sappi 49 tray. Using the above Sappi 49 jig system 49 paper-maché plugs could potentially be produced at one time. This system, however, was not successful due to the excessively high pressure required for the expulsion of excess water from the plug mixture.

The third and most successful attempt at making a tool for the production of wall-less plugs using the paper-maché mix was achieved by fitting a single piston to the head of a manual drill press and compressing the paper-maché mix into a single cavity of the extended Sappi 49 tray. The entire jig could be filled with paper-maché mix and by shifting it along the base of the drill press the medium in each cavity could easily and efficiently be compressed. The wet plugs were carefully pushed out

of the Sappi 49 mould and allowed to dry. Then they were placed on commercial egg trays to keep them upright. The egg trays could hold 30 plugs each, and were waterproofed on top and at the bottom using Gripflex[®], (a waterproofing product used for the sealing of roofs of buildings). Two small holes per cavity allowed drainage of excess water during irrigation and fertigation (Figure 3.1).



Figure 3.1: Equipment used for making paper-maché plugs. Shown here is the base of a half-dozen egg-tray supporting six plugs (bottom left), plastic bucket, electric drill with paint mixer, Sappi 49 mould, drill press with piston, container holding water, coal dust, shredded newspaper and composted pine bark

3.3.3 Treatments

The trial comprised of four treatments consisting of four different tray types. These were waterproofed egg trays with wall-less paper-maché plugs, black plastic Unigro 128 trays, white plastic Sappi 49 trays and white polystyrene Poly 128 shallow trays. The Unigro, Sappi 49 and Poly 128 shallow containers were filled with composted pine bark growing medium. The bottom of each

of the cavities of the waterproofed egg trays were perforated in two places with a fine screw driver to ensure drainage of excess water after irrigation and fertigation in the greenhouse.

Table 3.2: Dimensions of containers and plugs used in the study

Treatment	Depths (mm)	Volume (cm ³)	Cross-sectional shape
Paper-maché	80	80	Round
Unigro 128	98	60	Square
Sappi 49	80	80	Round
Poly 128 shallow	61	36	Square

3.3.4 Growing regime

Eight trays of each tray type were double-seeded with *Eucalyptus dunnii* seed and laid out in a randomized complete block design (RCBD). Each experimental unit consisted of 30 cells giving a total of 240 prospective seedlings per treatment. Double-seeding of the trial ensured that all cavities were populated by a seedling at the start of the experiment. After sowing the trays were capped with a fine layer of growing medium and watered. The trays were then stacked on top of one another and covered with a plastic sheet to initiate germination. Germination took place in a plastic covered greenhouse located at the phytotron facility of the Faculty of Agriculture, University of KwaZulu-Natal. Four days after sowing the trays were laid out in a greenhouse. Ten days after germination the entire trial was thinned out to leave a single viable seedling per cavity. The temperatures in the greenhouse ranged between 17 and 37°C. Five weeks after sowing the entire trial was transferred to the Mondi Business Paper, Mountain Home nursery in Hilton where it was housed for the remainder of the test period. Two fertigation regimes were applied during the trial. The first was a manual fertigation regime applied by means of a watering can. This regime was applied from week three after the trial was sown until the end of the fourth week at which time the trial was moved to Hilton. Thereafter plants were fertigated by means of an overhead irrigation system.

3.3.5 Measurements and data analysis

The height (H) and root collar diameter (RCD) of all plants were measured on two dates four months apart. Absolute growth rate (AGR) and relative growth (RGR) rate for the period between the dates that data was collected (t_2-t_1) were determined on the basis of height and root collar diameter measurements according to the following formulae (Clipson *et al.*,1994):

$$AGR_{(H)} = (H_2 - H_1)/(t_2-t_1)$$

$$RGR_{(H)} = \text{Ln } H_2 - \text{Ln } H_1/(t_2-t_1)$$

$$AGR_{(RCD)} = (RCD_2 - RCD_1)/(t_2-t_1)$$

$$RGR_{(RCD)} = \text{Ln } RCD_2 - \text{Ln } RCD_1/(t_2-t_1)$$

Twenty plants were randomly selected from each of the eight blocks, harvested, oven dried for 48 hours and milled to a fine powder for nutrient analysis. The data were subjected to analysis of variance as the growth data were normally distributed. Means were separated using Tukey's test. The data was managed using MS Excel 2007 and analysed with GenStat Release 11.1 (Eleventh Edition).

3.4 Results

The Unigro and Sappi 49 trays produced seedlings that were similar but significantly different in height and RCD (Figure 3.2 and Table 3.2). Significant differences were no longer detected when analyzing height difference at the end of the four-month growing period. The Poly 128 shallow container produced the smallest plants. The Paper-maché plugs produced seedlings that were smaller in height and RCD at first measurement than those produced in the other three containers. Height and RCD however exceeded that of seedlings raised in the Poly 128 shallow containers after four months.

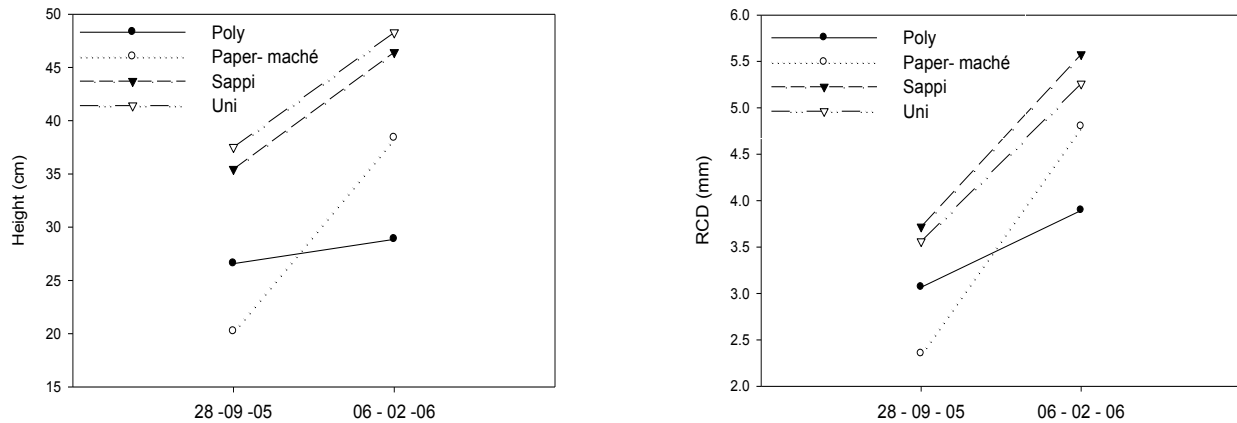


Figure 3.2: Height (Left) and RCD (Right) of *Eucalyptus dunnii* seedlings measured at two dates over a four month period

Table 3.3: The effect of four different tray types on height and RCD of *Eucalyptus dunnii* seedlings at two dates (four months apart)

Treatment	Height (cm)		RCD (mm)	
	28/9/2005	6/2/2006	28/9/2005	6/2/2006
Poly	23.92 ± 0.13 ^d	28.79 ± 0.11 ^c	3.056 ± 0.011 ^c	3.902 ± 0.016 ^d
Paper-maché	19.77 ± 0.40 ^c	37.39 ± 0.57 ^b	2.382 ± 0.037 ^d	4.71 ± 0.064 ^c
Unigro	37.78 ± 0.40 ^a	48.58 ± 0.44 ^a	3.59 ± 0.033 ^b	5.268 ± 0.039 ^b
Sappi	35.80 ± 0.43 ^b	46.98 ± 0.49 ^a	3.747 ± 0.038 ^a	5.566 ± 0.042 ^a

Mean values (±SE) of each column with different letters are significantly different according to Tukey's test ($P < 0.05$).

The AGR and RGR for both height and root collar diameter followed equal trends (Table 3.4). The plants raised in paper-maché plugs had the highest AGR and RGR for both height and RCD followed by plants raised in Sappi 49 and Unigro trays which performed similarly with no significant difference between the two. The AGR and RGR of plants produced in the Poly 128 shallow container were significantly lower than that plants produced in the other tray types (Figure 3.2 and Table 3.4).

Table 3.4: The effect of four different tray types on absolute growth rate (AGR) and relative growth rate (RGR) of *Eucalyptus dunnii* seedlings determined on the basis of height and RCD between two dates four months apart

Treatment	Height (cm month ⁻¹)		RCD (mm month ⁻¹)	
	AGR	RGR	AGR	RGR
Poly	1.107 ± 0.633 ^c	0.043 ± 0.025 ^c	0.192 ± 0.058 ^c	0.055 ± 0.015 ^c
Paper-maché	4.004 ± 2.608 ^a	0.150 ± 0.120 ^a	0.529 ± .0.159 ^a	0.157 ± 0.050 ^a
Unigro	2.453 ± 2.367 ^b	0.058 ± 0.060 ^{bc}	0.381 ± 0.167 ^b	0.088 ± 0.045 ^b
Sappi	2.541 ± 2.499 ^b	0.063 ± 0.065 ^b	0.413 ± 0.187 ^b	0.092 ± 0.046 ^b

Mean values (±SE) of each column with different letters are significantly different according to Tukey's test ($P < 0.05$)

Macro and micro nutrient and non-structural carbohydrate reserve concentrations across the treatments, with the exception of % Carbon, was variable with no easily recognizable patterns. Analysis of means of Carbon, Sulphur, Iron and Boron concentrations yielded results that indicated no significant differences across treatments. Significant differences were observed for all other nutrients and non-structural carbohydrates (Figure 3.3).

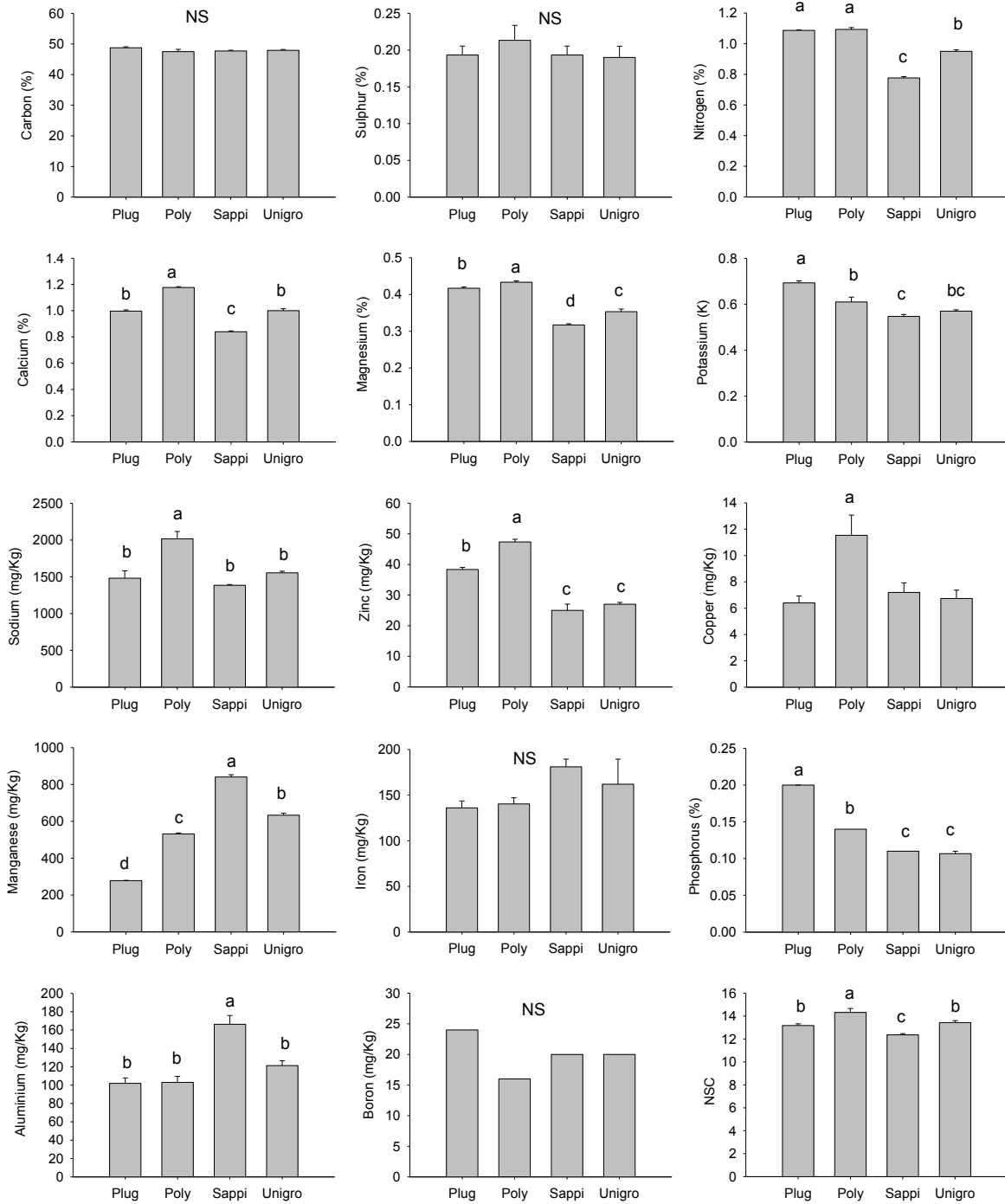


Figure 3.3: Effect of container type on nutrient concentrations of four-month-old *Eucalyptus dunnii* seedlings. Bars (\pm SE) of each nutrient with different letters are significantly different according to Tukey's test ($P < 0.05$). (Plug is equivalent to the Paper-maché plugs)

3.5 Discussion and conclusions

The ratios of ingredients used for preparing the paper-maché mixture (Table 3.1) were derived after some initial tests and can undoubtedly be improved upon following further experimentation with binding agents, growing media and growth promoting substances. Binding agents other than common household flour from the Zeijlemaker and de Laborde (1977) protocol can be natural plant and animal extracts or ecologically friendly synthetic products. Growing media other than composted pine bark that can potentially improve the performance of paper-maché plugs include coconut coir, sawdust, woodchips and shredded wattlebark. Growth promoting substances that can potentially be added are individual nutrients in solution, mixtures of nutrients, slow release fertilizers and biological agents such as mycorrhizae and other fungal, bacterial and plant extracts.

The production efficiency of the paper-maché plugs according to the protocol of Zeijlemaker and de Laborde (1977) was improved following successive modifications to the compression equipment developed for the compression of the paper-maché mixture in the mould. The force of compression required to achieve an adequate expulsion of excess water was larger than expected. This influenced the design of the equipment.

Initial establishment of seedlings in paper-maché plugs was low, both in terms of initial germination (i.e. the number of non-germinants was high) and in terms of seedling growth. Seedlings that did emerge and establish themselves successfully in the plugs, however, rapidly increased in height and RCD (Figure 3.2 and Table 3.3). These outcomes may be attributed to the high density of the plugs. It is likely that the young radicles experienced difficulty penetrating the medium and thereafter for these developing root systems to further establish themselves in the plug.

An in-depth analysis of the nutrient and non-structural carbohydrate reserve concentrations of the results obtained is arduous since samples were not collected and analysed at multiple and regular intervals. The evolution of these concentrations can thus not be followed over time for the duration of the trial and can only be compared to results obtained by other researchers in trials conducted under different experimental trial conditions. Due to the limited amount of published data on eucalypt seedling nutrient concentrations similar data from other tree genera can be consulted but to what extent such results can be relied upon to guide further research or to make inferences regarding

the results obtained is not known. The anticipated finding was to observe similar results for concentrations of nutrients of all plants raised in containers filled with composted pine bark (plants raised in the Unigro, Poly 128 Shallow and Sappi containers) and results that were either similar or different from these in the plants raised in the paper-maché plugs. However, that anticipated result was not obtained. It is thus likely that container properties play a larger role than expected on plant performance. The underlying mechanisms of how container properties in terms of their shape and size and materials from which they were produced impact plant micro- and macro-nutrients and non-structural carbohydrate status requires further investigation.

The effect of tray colour and its effect on the temperature of the growing medium has been investigated by a number of researchers. Research on this container property and its effect on nutrient concentrations and seedling performance as a result of medium temperature may provide some insight into the observed fluctuations in nutrient concentrations.

The dimensions of containers and their effect on drainage properties within the medium may impact not only on water availability but also on nutrient availability and subsequently nutrient concentrations. The effect of dimensions of containers drainage of water in a volume of medium is well documented but how the drainage of water in a medium affects availability of different nutrients to a plant is not well documented. This is probably because the degree of nutrient availability for uptake by plant roots is highly dependent on the physico-chemical properties of the medium. It is thus very difficult to elucidate the mechanisms that determine nutrient availability to plants as a result of the drainage properties of the medium (as a result of container dimensions) irrespective of the physico-chemical properties of the medium.

It is interesting to note that the concentration of copper in seedlings raised in the Poly 128 Shallow containers was found to be significantly higher than concentrations of this macro-nutrient in seedlings raised in the other tray types. This result is not surprising considering that the Poly 128 Shallow trays were dipped in a copper solution to ensure root pruning.

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CHAPTER 4 POTENTIAL USE OF PAPER-MACHÉ PLUGS FOR IMPROVED EUCALYPT SEEDLING PRODUCTION IN SOUTH AFRICA

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4.1 Abstract

Freestanding rooting plugs were made by (1) mixing, pressurizing and gluing paper-maché plugs, (2) cutting a sponge block and (3) filling Hessian bags with standard pinebark-based growing medium. These treatments were also compared with three types of containers used in commercial nurseries: (4) Unigro trays, (5) Sappi 49 trays, and (6) Poly 98 deep trays. *Eucalyptus grandis* and *E. dunnii* plants were raised from seed over a period of 70 days. For each species and treatment 60 plants were outplanted in a field trial. Survival and growth of the outplanted trees was determined after nine weeks. The nursery results showed that paper-maché plugs yielded the tallest plants for *E. grandis*, and Sappi 49 trays produced the tallest *E. dunnii* seedlings. The sponge blocks produced significantly smaller trees in both species. After outplanting, the survival of trees was high for each treatment. There is a need to test performance attributes of the various types of planting stock under more stressful site conditions to determine long-term benefits, if any, of producing planting stock in paper-maché plugs. If these studies are concluded successfully, paper-maché plugs could replace peat in commercial forest nurseries, preventing environmental degradation of peat-mined wetlands.

keywords: root plug, wall-less plug, container type, containerized nurseries

4.2 Introduction

The degree of out-planting performance of planting stock determines the success of any forestry establishment operation. Post-planting survival and rate of early growth are the two most obvious measures of regeneration success. Losses in value and volume yield together with increased regeneration costs should be minimized through efficient plantation establishment (Albert *et al.* 1980). This in turn depends on seedling morphological and physiological characteristics that meet targets associated with favorable performance under an anticipated range of site conditions (Davis and Jacobs, 2005).

Over the last couple of decades, containerized nurseries replaced their bare-root counterparts in many parts of the world, especially where plantation forestry operations depend on effective establishment and fast growth of trees. Containerized nurseries helped also to accelerate propagation techniques, especially vegetative propagation, and to reduce prices of planting material of select genotypes of trees. Despite the fact that these objectives have been largely achieved, many planting operations fail due to early tree dieback or vulnerability to wind in older forest stands. Both problems result from root deformations and stem strangulation of containerized planting stock with roots constrained by hard container walls (Chapman *et al.*, 2003; Rune, 2003). In many studies world-wide root collar diameter of seedlings has been found to correlate strongly with field performance after planting (Thompson, 1985; Bayley and Kietzka, 1997; Jacobs *et al.*, 2005). Diameter of the stem at root collar, however, depends on the amount of space allocated to roots during the nursery stage. The older the seedlings the greater the root collar, but inferior planting stock may result from prolonged cultivation in combination with insufficient rooting space. McCubbin and Smith (1991) showed the importance of recognizing the “planting windows” for *E. grandis* seedlings grown in different containers. They concluded that delaying planting will have a worse effect on post-establishment growth than planting younger seedlings before their optimum quality is reached. This was confirmed by other studies which showed no benefit from planting large diameter containerised seedlings (Morris, 1994). A possible explanation can be that the overgrown seedlings not necessarily facilitate a well branched and functioning root system. Well performing planting material must be characterized by multiple and actively growing root tips for improved soil colonization and root-soil contact (Thompson, 1985; Deans *et al.*, 1990). Whatever findings emerge

from various studies it has been agreed that root architecture derived from nursery cultural practices cannot be effectively corrected and has a life-long impact on tree performance (Moore, 2003).

A well designed container should promote branching of the tap root when it reaches the sides of a container. For drainage, most containers are equipped with an opening at their lowest point and therefore the tap root is usually air-pruned and branched. The lateral roots, however, may be deflected upon reaching the container walls and then continue growing by following the container shape. Such root deformations are called the “caging effect”. After out-planting root circling and continued growth around tree stems may result in stem strangulation and breakage at the root collar in later stages of tree development. Deformed roots disrupt nutrient and water transport in affected trees (Hay and Woods, 1975). The design of containers involves changing their shapes and incorporating grooves, slits or ridges into container walls to create a mechanical barrier or to stimulate air-pruning of the lateral roots. Most of these designs, however, still incorporate smooth container walls which result in undesirable root deformations. The introduction of copper-based chemicals, coated onto the container walls for root pruning (Bayley and Snell, 1997), can be effective but environmentally unfriendly.

Other than the two valuable works on the subject by Barnett and Brisette (1971) and Landis (1990) there is an abundance of published research articles describing a number of specific containers in relation to root development, plant quality and plant performance (Harris, 1967; Whitcomb, 1988; Appleton, 1989; 1993 & 1995; Arnold and McDonald, 1999). Several different systems for categorizing containers have been used. A practical system divides containers into two functional groups: those that are planted with the tree seedling and those that are removed before the seedling is planted (Tinus and McDonald, 1979). The first category includes containers which are biodegradable and result in minimum root disturbance at planting. Containers that constitute both the container and the growing medium as a complete unit are generally known as blocks or plugs.

The objective of this study was to develop and test wall-less plugs for growing eucalypt seedlings and to compare their performance with planting stock raised in standard containers used in South African nurseries.

4.3 Materials and Methods

The first attempt at making a wall-less plug prototype was made using improvised tools and the original WRIBLOK protocols for making the paper-maché mix (Zeijlemaker and de Laborde, 1977). A tool was designed consisting of a drill press with piston to extract excess water from the Paper-maché mixture. This compression tool was used together with a Sappi 49 container which served as a mould to produce plugs equal in shape and volume to the Sappi 49 cavity. The Sappi 49 tray consists of 49 bullet-shaped cells, each of dimensions specified in Table 1. A 550W electric drill mounted with a paint mixer was used at approximately 600 rpm to produce the paper-maché from shredded newspaper, composted pine bark, charcoal dust and water mixed in the proportions of 300 g, 2400 g and 600 g and 7 L, respectively. The resulting paper-maché mixture was poured into the Sappi 49 tray and the largest possible quantity of water was removed. The wet plugs were carefully pushed out of the Sappi 49 mould and allowed to dry. They were then placed on commercial egg holders to keep them upright. Other innovative methods included a Hessian bag filled with composted pine bark used for seedling production in the local nurseries and blocks of sponge cut from sponge of density 57.7 g/dm³. Egg holders were also used for holding the Hessian bags upright and the sponge blocks were supported with Unigro carriers. These treatments were compared to commercial containers used by forestry nurseries. Their characteristics are provided in Table 1. All the walled containers and the Hessian bags were filled with standard composted pine bark.

Table 4.1: Dimensions of containers and plugs used in the study

Treatment	Depths (mm)	Volume (cm ³)	Cross-sectional shape
Paper-maché	80	80	Round
Hessian bag	80	80	Round
Sponge block	110	200	Square
Unigro 128	98	60	Square
Sappi 49	80	80	Round
Poly 98 deep	100	60	Round

The treatments were replicated five times in a randomised complete block (RCBD) design. Each experimental unit consisted of 12 cells giving a total of 60 prospective seedlings per treatment and species. Seed of *E. grandis* and *E. dunnii* was sown in each cavity. The sown trays were initially

inserted in black polyethylene bags and placed in a heated greenhouse for the initiation of germination. After 96 hours (4 days) the trays were taken out of the polyethylene bags and placed in a glasshouse on benches with overhead sprinkler fertigation. The third week after seed started emerging, fertilizing began with applications of Agrofert Orange (13.2% N:17.6% P: 11.0% K) at a concentration of 0.6 g L⁻¹ three times a week through the fertigation system. Seedlings were fertigated to saturation. In addition Calmag (13.7% N: 11.7% Ca: 38 g kg⁻¹ Mg: 2000 mg kg⁻¹ B) was applied once a week at 0.4 g L⁻¹ until seedlings were saturated. This fertilizing regime was followed until seedlings were ready for outplanting.

The seedlings remained in the glasshouse for the remainder of the experiment until the time of outplanting into the field (70 days). The temperatures in the glasshouse reached up to 30 °C on very warm days and dropped to 15 °C on cold nights. The plants were cultivated for 9 weeks in the nursery and then planted in the field. During the outplanting period temperatures reached 28 °C in the field on very warm days and dropped to a few degrees above freezing. Freezing temperatures were prevented by overhead irrigation during very cold nights. Every two weeks after planting, tree height and root collar diameter were measured. A total of 60 plants from each treatment were outplanted for each of the tested species. These were divided into three reps of 20 plants in a RCBD design and grown for 9 weeks. After that period surviving trees were counted and carefully excavated. Their heights, root collar diameters and dry mass of shoots and roots were determined. The data were subjected to analysis of variance as the growth data were normally distributed. Means were separated using Tukey's test. The data was managed using MS Excel 2007 and analysed with GenStat Release, 11.1 (Eleventh Edition).

4.4 Results

Paper-maché plugs showed greater seedling height in comparison to the other two types of plugs examined. However, these results were not significantly different from Hessian bags (Figure 4.1 and Table 4.2). The Paper-maché plugs significantly produced larger root collar diameters compared to all the other containers tested. Even though in many cases the results were not significantly different to the other container types, Paper-maché plugs achieved the highest shoot and root dry mass (Table 4.2).

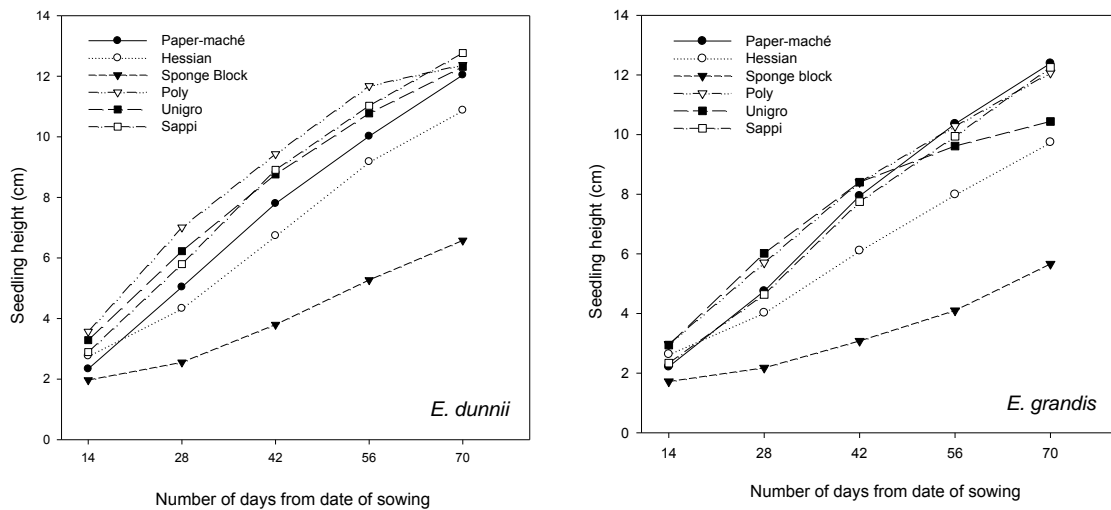


Figure 4.1: Height of *E. dunnii* and *E. grandis* seedlings germinated and raised in various containers

In the outplanting experiments, performance of the six container types was evaluated both in terms of survival and growth. Outplanting survival was close to 100% for all container types with the exception of Sponge blocks which performed significantly poorer. With regards to growth, *E. dunnii* and *E. grandis* performed similarly with the exception of height and root dry mass. The combined results of both species with Paper-maché plugs (seedlings raised and transplanted in the field) showed greater height, root collar diameter and shoot dry mass in comparison to the other five container types evaluated (Table 4.3). This tray type showed significantly greater root dry mass than other trays with the exception of Hessian bags (Table 3). In most cases, the outplanting of *E. dunnii* and *E. grandis* seedlings grown in Paper-maché plugs i.e (Species X Container), recorded higher values for all growth parameters studied. However, there were some exceptions.

Table 4.2: Characteristics of *E. dunnii* and *E. grandis* seedlings by nursery treatment after seventy days from sowing

Treatment	Height (cm)	Root collar diameter (mm)	Shoot dry mass (g)	Root dry mass (g)
Species				
<i>E. dunnii</i>	11.2 ± 4.4 ^a	1.9 ± 0.5 ^a	1.11 ± 0.3 ^a	0.44 ± 0.2 ^a
<i>E. grandis</i>	10.4 ± 2.7 ^a	1.8 ± 0.4 ^a	0.89 ± 0.2 ^b	0.28 ± 0.1 ^b
Container				
Paper-maché	12.2 ± 2.4 ^a	2.2 ± 0.7 ^a	1.25 ± 0.3 ^a	0.45 ± 0.1 ^a
Hessian bag	10.3 ± 5.6 ^a	1.9 ± 0.5 ^b	0.85 ± 0.1 ^b	0.35 ± 0.1 ^{ab}
Sponge block	6.1 ± 3.4 ^b	1.5 ± 0.5 ^c	1.00 ± 0.3 ^{ab}	0.24 ± 0.2 ^b
Unigro 128	12.2 ± 2.3 ^a	1.8 ± 0.3 ^b	1.04 ± 0.1 ^{ab}	0.32 ± 0.1 ^{ab}
Sappi 49	12.5 ± 2.9 ^a	2.0 ± 0.4 ^b	1.00 ± 0.1 ^{ab}	0.44 ± 0.1 ^{ab}
Poly 98 deep	11.4 ± 4.6 ^a	2.0 ± 0.3 ^b	0.82 ± 0.1 ^b	0.37 ± 0.1 ^{ab}
Species X Container				
<i>E. dunnii</i> X Paper-maché	12.0 ± 3.3 ^a	2.0 ± 0.6 ^{ab}	1.42 ± 0.5 ^a	0.58 ± 0.2 ^a
<i>E. dunnii</i> X Hessian bag	10.9 ± 7.3 ^{ab}	2.0 ± 0.6 ^{ab}	0.98 ± 0.2 ^{ab}	0.40 ± 0.1 ^{abcd}
<i>E. dunnii</i> X Sponge block	6.6 ± 5.6 ^b	1.5 ± 0.5 ^{bc}	1.10 ± 0.4 ^{ab}	0.34 ± 0.3 ^{abcd}
<i>E. dunnii</i> X Unigro 128	12.4 ± 2.9 ^a	1.9 ± 0.3 ^{abc}	1.12 ± 0.2 ^{ab}	0.38 ± 0.1 ^{abcd}
<i>E. dunnii</i> X Sappi 49	12.8 ± 3.4 ^a	2.0 ± 0.5 ^{ab}	1.06 ± 0.1 ^{ab}	0.40 ± 0.1 ^{abcd}
<i>E. dunnii</i> X Poly 98 deep	12.3 ± 3.7 ^a	2.0 ± 0.3 ^{ab}	0.98 ± 0.3 ^{ab}	0.55 ± 0.1 ^{ab}
<i>E. grandis</i> X Paper-maché	12.4 ± 1.4 ^a	2.4 ± 0.7 ^a	1.07 ± 0.1 ^{ab}	0.32 ± 0.1 ^{abcd}
<i>E. grandis</i> X Hessian bag	9.8 ± 3.9 ^{ab}	1.7 ± 0.4 ^{bc}	0.75 ± 0.1 ^b	0.31 ± 0.1 ^{abcd}
<i>E. grandis</i> X Sponge block	5.7 ± 1.3 ^b	1.4 ± 0.4 ^c	0.90 ± 0.3 ^{ab}	0.14 ± 0.1 ^d
<i>E. grandis</i> X Unigro 128	10.4 ± 1.6 ^{ab}	1.7 ± 0.2 ^{bc}	0.95 ± 0.1 ^{ab}	0.26 ± 0.1 ^{bcd}
<i>E. grandis</i> X Sappi 49	12.3 ± 2.4 ^a	1.9 ± 0.3 ^{abc}	1.03 ± 0.2 ^{ab}	0.47 ± 0.1 ^{abc}
<i>E. grandis</i> X Poly 98 deep	12.1 ± 5.5 ^a	1.9 ± 0.3 ^{abc}	0.66 ± 0.1 ^b	0.18 ± 0.1 ^{cd}

Mean values (± SD) for species, container and species x container in each column with different letter(s) is significantly different ($p < 0.05$) according to Tukey's test.

Table 4.3: Characteristics of *E. dunnii* and *E. grandis* trees by nursery treatment after nine weeks growth in the field

Treatment	Survival (%)	Height (cm)	Root collar diameter (mm)	Shoot dry mass (g)	Root dry mass (g)
Species					
<i>E. dunnii</i>	96.7 ^a	41.4 ± 5.8 ^a	5.7 ± 0.6 ^a	39.9 ± 13.9 ^a	6.9 ± 2.2 ^b
<i>E. grandis</i>	95.0 ^a	35.2 ± 6.4 ^b	5.9 ± 0.9 ^a	49.8 ± 22.8 ^a	8.1 ± 3.9 ^a
Container					
Paper-maché	97.5 ^a	42.9 ± 5.1 ^a	6.6 ± 0.9 ^a	60.5 ± 24.8 ^a	9.9 ± 4.3 ^a
Hessian bag	96.7 ^a	38.2 ± 3.1 ^a	5.8 ± 0.5 ^{ab}	47.7 ± 15.9 ^{ab}	8.9 ± 4.4 ^{ab}
Sponge block	86.7 ^b	32.7 ± 8.2 ^b	5.0 ± 0.8 ^b	29.0 ± 11.0 ^b	4.9 ± 1.3 ^d
Unigro 128	98.3 ^a	37.6 ± 8.5 ^a	5.7±0.1 ^{ab}	53.5 ± 24.7 ^{ab}	7.9 ± 3.1 ^b
Sappi 49	99.2 ^a	43.9 ± 3.5 ^a	6.4±0.4 ^{ab}	42.0 ± 14.0 ^{ab}	7.1 ± 2.3 ^{bc}
Poly 98 deep	96.7 ^a	34.3 ± 7.9 ^b	5.4±1.1 ^b	36.6 ± 19.9 ^b	6.5 ± 2.9 ^c
Species X Container					
<i>E. dunnii</i> X Paper-maché	98.3 ^a	43.4 ± 2.1 ^{ab}	6.1 ± 0.2 ^{ab}	50.5 ± 19.6 ^{ab}	8.9 ± 4.2 ^a
<i>E. dunnii</i> X Hessian bag	95.0 ^a	40.5 ± 1.3 ^{ab}	5.4 ± 0.4 ^{ab}	31.5 ± 7.7 ^{ab}	5.7 ± 1.2 ^c
<i>E. dunnii</i> X Sponge block	90.0 ^{ab}	32.4 ± 2.9 ^b	4.8 ± 0.7 ^b	21.7 ± 4.1 ^b	3.8 ± 0.2 ^d
<i>E. dunnii</i> X Unigro 128	98.3 ^a	40.5 ± 11.6 ^{ab}	5.6 ± 0.9 ^{ab}	52.4 ± 13.0 ^a	8.6 ± 2.0 ^{ab}
<i>E. dunnii</i> X Sappi 49	98.3 ^a	50.8 ± 5.7 ^a	6.5 ± 0.4 ^{ab}	43.1 ± 11.9 ^{ab}	7.2 ± 2.3 ^b
<i>E. dunnii</i> X Poly 98 deep	100 ^a	40.7 ± 11.0 ^{ab}	5.6 ± 1.0 ^{ab}	40.5 ± 26.8 ^{ab}	7.5 ± 3.3 ^b
<i>E. grandis</i> X Paper-maché	96.7 ^a	42.5 ± 8.1 ^a	7.1 ± 1.5 ^a	70.5 ± 29.9 ^a	10.8 ± 4.3 ^a
<i>E. grandis</i> X Hessian bag	98.3 ^a	35.9 ± 4.9 ^{ab}	6.1 ± 0.5 ^{ab}	63.8 ± 24.0 ^{ab}	12.1 ± 7.6 ^a
<i>E. grandis</i> X Sponge block	83.3 ^b	33.1 ± 13.6 ^{ab}	5.2 ± 0.9 ^b	36.3 ± 17.8 ^b	6.1 ± 2.3 ^{bc}
<i>E. grandis</i> X Unigro 128	98.3 ^a	34.7 ± 5.5 ^{ab}	5.8 ± 1.0 ^{ab}	54.5 ± 36.1 ^{ab}	7.3 ± 4.2 ^b
<i>E. grandis</i> X Sappi 49	100 ^a	36.9 ± 1.2 ^{ab}	6.2 ± 0.3 ^{ab}	40.9 ± 16.1 ^b	7.0 ± 2.2 ^b
<i>E. grandis</i> X Poly 98 deep	93.3 ^a	27.9 ± 4.8 ^b	5.2 ± 1.1 ^b	32.7 ± 13.0 ^b	5.4 ± 2.5 ^c

Mean values (± SD) for species, container and species x container in each column with different letter(s) is significantly different ($p < 0.05$) according to Tukey's test.

4.5 Discussion and conclusions

Plants grown in sponge blocks were significantly smaller than seedlings produced in other container types for both species of eucalypts (Figure 4.1 and Table 4.2). It is possible that relatively large holes prevented sufficient water absorption and storage. In addition to this, sponge blocks do not have an available nutrient source due to their inert nature. Watering and fertigation of these types of plugs was a challenge, as all of them were exposed to the same irrigation system, but it was observed that wall-less plugs dried out fast. This can be attributed to greater evaporation since the rooting material was not protected by solid walls. Despite this, seedling characteristics remained similar among the treatments (Table 4.2). However, the water requirement of wall-less plugs needs further investigations.

The field performance of trees is shown in Table 4.3. *Eucalyptus dunnii* yielded taller trees than *Eucalyptus grandis*. Perhaps this is because they were cultivated during winter and *E. dunnii* performs better under cold conditions than *E. grandis*. *E. grandis* produced more roots than *E. dunnii*. The survival of trees was high and non-conclusive results were obtained regarding tree stability after planting. Paper-maché-treated plants performed better than Sponge blocks and Poly 98 deep trays. Similar trends were recorded within species responses to nursery treatments.

It is likely that for the first time unconventional materials, such as sponge and Paper-maché, have been experimented with to grow eucalypt seedlings. Paper-maché appeared to be an appropriate substrate to grow seedlings when the standard fertigation regime was applied. In the past it was used to grow black wattle (*Acacia mearnsii*) seedlings (Barrett, 1978). It is possible that seedling growth could be improved by adjusting watering and nutrition and further studies are recommended in this regard. It is also uncertain if seedling quality could be enhanced in terms of rooting structure. Seedlings cultivated in Paper-maché plugs were similar to their counterparts prepared in the standard commercial nursery containers. They also performed equally well under field conditions. A range of seedling size and age grades would need to be tested on a range of stressful sites to show whether wall-less plugs offer any advantage compared to standard planting stock. An advantage of Paper-maché plugs is that they are manufactured by recycling waste paper and not by mining natural peatlands with its harmful environmental impact on wetland hydrology. Paper-maché plugs also offer an advantage over composted pine bark-filled containers, used extensively in South Africa.

Despite bark being an excellent rooting medium, its quality can be inferior due to inadequate composting. Commercial nurseries are therefore always looking for new rooting media and paper-maché plugs may provide a good alternative to the current system.

This study shows the potential of wall-less plugs for the production of planting stock of forest trees. Between 120 and 130 million eucalypt seedlings are produced annually in various nurseries throughout South Africa (Zwolinski and Bayley 2001). Paper-maché plugs evaluated in this study showed promising results for *E. dunnii* and *E. grandis*. However, long-term growth studies are required to confirm the benefits of implementing Paper-maché plugs on a large scale. The manufacturing process of these plugs is relatively easy and can likely be improved. However, at this stage, the production cost and overall economics of large-scale implementation is not yet available and needs to be assessed.

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**CHAPTER 5 DIGITAL IMAGE ANALYSIS SOFTWARE TO DETERMINE THE EFFECT
OF SIX CONTAINER TYPES AND TWO GROWING MEDIA ON
EUCALYPTUS GRANDIS SEEDLING QUALITY**

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5.1 Abstract

In this study *Eucalyptus grandis* seedling root systems were measured to determine their size by means of two methods 1) Archimedes' principle of water displacement to determine root volume and 2) Image analysis software to determine two dimensional surface area of roots. Plants were raised in three types of walled containers (Sappi 49, Unigro 128 and Poly 98 Deep) and three prototype wall-less containers (Paper-maché plugs, Covetan and Hessian bags) in two types of media over a 10 week period. Plant performance was measured in terms of height, RCD, root volume and root surface area. Correlation analysis was carried out to determine the possibility of using root surface area measurements for plant quality assessment prior to dispatchment of plants from the nursery. Coconut coir resulted in a reduced number of non-germinants and plants with larger height and RCD measurements compared to planting stock produced in composted pine bark. No significant differences were observed between performance of plants produced in the three types of walled-containers and the Paper-maché plugs in the greenhouse. Growth of plants raised in Covetan and Hessian pockets was significantly inferior. A high correlation between root volume and root surface area indicates that root surface area can be used by nursery managers as a tool to determine readiness of plants for dispatchment into the field.

keywords: root volume, seedling quality, container, composted pine bark, coconut coir

5.2 Introduction

Seedling quality and production costs are the two most important factors when deciding on the implementation of operational procedures in large-scale containerized nurseries.

From the host of measurable parameters available to determine nursery stock quality root volume has been shown to have potential when used in conjunction with other characteristics such as height and root collar diameter (Wahlenberg, 1928; Chapman, 1948; Fowells, 1953; Adams, 1964; Pawsey, 1972; George and Frank, 1973; Blair and Cechy, 1974; Zaerr and Lavender, 1976; Bacon *et al.*, 1977; Carrier, 1984; Rose *et al.*, 1991) and shoot:root ratio (Hermann, 1964; Stonecypher *et al.*, 1966; Lopushinsky and Beebe, 1976; Glerum and Boufford, 1979). Archimedes's principle of water displacement is the conventional and widely used method for root volume determination.

Computer software has been developed to determine physical parameters of root systems (Berntson, 1992). This has been done in research institutes and by laboratory instrument manufacturers. Although such software has been useful for studies on radical emergence and development following seed germination, it has been less successful when analyzing highly complex root systems. Most of these methods make use of software that analyzes scanned images of carefully washed root systems. These images are generated with the aid of conventional or specially modified flat-bed scanners onto which the washed root systems are placed and scanned, either in a shallow basin of water to enhance the spreading of the roots, or dry, directly on the glass top of the scanner.

Software has been developed to determine physical parameters of roots including total root length and branching patterns of roots, i.e. the number of different order lateral roots. Surface area is another parameter that is accurately measured by freely and easily available image analysis software. Examples of surface areas that can successfully be measured are leaf surface areas, areas of organelles within cells and differentially pigmented areas on plant and animal structures. With the aid of such software surface area of root systems can be determined regardless of the complexity of the root system provided the size of the root systems under analysis does not exceed the size of the scanner window.

According to our knowledge no studies have been conducted in which root volume measurements and root surface area measurements have been compared by means of correlation analysis. Such a study would determine the possible use of root surface area measurements in place of or as an added measurement of root volume in decision-making processes regarding the quality of planting stock.

Furthermore, containers in which planting stock are raised are known to impact root volume and seedling quality. Many different types of containers are available to the nurseryman and the selection or development of containers that provide optimal outplanting survival, early growth and tree stability is critical for large-scale operations.

The objectives of the present study were to determine (i) the degree of correlation between root volume and root surface area of *Eucalyptus grandis* seedlings, and (ii) the effect of a) six different seedling tray types (three types of walled containers used in the industry and three prototype wall-less containers) and b) two different growing media on greenhouse performance of seedlings over a 10-week period.

5.3 Materials and Methods

Eucalyptus grandis seedlings were produced in a glasshouse at the University of KwaZulu-Natal Controlled Environment Unit (CERU) over a period of 10 weeks (70 days) in six different tray types (Table 5.1) using two types of growing media. The media were 1) composted pine bark and 2) a mixture of coconut coir and perlite. The plants were raised from seed obtained from the Mondi Business Paper Mountain Home nursery (Batch number: M9627).

Table 5.1: Dimensions of containers and plugs used in the study

Treatment	Depths (mm)	Volume (cm ³)	Cross-sectional shape
Paper-maché	80	80	Round
Hessian bag	80	80	Round
Covetan bag	80	80	Round
Unigrow 128	98	60	Square
Sappi 49	80	80	Round
Poly 98 deep	100	60	Round

The Unigro 128, Sappi 49 and Poly 98 deep trays were obtained from their respective mother companies where they are currently used for commercial seedling production whereas the paper-maché plugs and the Hessian and Covetan bags were wall-less experimental containers. The paper-maché plugs were manufactured using the revised protocol described in Chapter 3. The Hessian and Covetan bags were produced using a similar though not identical protocol. Strips of both materials were placed on a table and cut using conventional scissors. These were sewn with an Elna sewing machine to make bags that could be filled with growing medium. Both bags were made such that the volume of medium they could hold was equal to the volume of medium that could be held in each one of the cavities of the Sappi 49 tray (Table 1).



Figure 5.1: Photographs depicting the first stage of the Hessian bag production (Left) and the initial stage and the intermediate stages of the production of Covetan bags (Right)

Egg trays (6 x 5 trays) were waterproofed and used as support for the three types of experimental wall-less plugs. The bottom of each of the thirty cavities of the waterproofed egg trays were perforated in two places with a fine screw driver to ensure drainage of excess water after irrigation and fertigation in the greenhouse.

The treatments were replicated four times in a randomised complete block (RCBD) design. Each experimental unit consisted of 12 cells giving a total of 48 prospective seedlings per treatment for both types of growing media.

Each cavity of the three tray types and each plug and bag with growing media was double-seeded to ensure that the experiment could be started with a complete set of viable seedlings. The seeded trays were initially inserted in black polyethylene bags and placed in a heated greenhouse for germination initiation. After 96 hours (4 days) the trays were taken out of the polyethylene bags and placed on benches in a glasshouse. Those cavities, plugs and bags in which both seeds had germinated were thinned out to a single seedling two weeks after germination. This was carried out by removing the smallest and largest of all the seedlings in order to obtain a crop that was fairly uniform. The number of non-germinants for each tray type were counted and recorded. Irrigation was achieved by means of an overhead sprinkler. The third week after seedlings started emerging, fertilisation began with applications of Agrofert Orange (13.2% N:17.6% P: 11.0% K) at a concentration of 0.6 g L⁻¹ three times a week through the fertigation system. Seedlings were fertigated to saturation. In addition Calmag (13.7% N: 11.7% Ca: 38 g kg⁻¹ Mg: 2000 mg kg⁻¹ B) was applied once a week at 0.4 g L⁻¹ until seedlings were saturated. This fertilisation regime was followed for the duration of the 10-week experiment. The temperatures in the glasshouse reached 30 °C on very warm days and dropped to 15 °C on cold nights. In addition to a single set of RCD measurements taken and recorded at the end of the 10-week period, five sets of biweekly seedling height measurements were acquired.

On week ten of the seedling growing period ten seedlings from each treatment and each growing medium type were removed from their respective trays. The root systems of the seedlings were laid bare by carefully removing the growing medium with the aid of water in a large open container. After the initial bulk of growing medium was removed, all the remaining medium was further carefully removed with clean water in a second large open container. A total of 120 seedling root

systems (10 seedlings x 6 tray types x 2 medium types) were washed with each washing taking approximately 5 minutes per root system. Root volume (RV) and root surface area (RSA) of the seedlings were determined.

Root volume measurements were acquired by means of the well-established water displacement technique based on Archimedes's principle. The seedling root systems were immersed in a container of water placed on a balance. The displaced water (measured in grams) is equal to the volume (measured in cubic centimeters) of the root system in that 1 g of water equals the displacement of a volume of 1 cm³ of water at room temperature (Burdett 1979).

The RSA of seedlings was determined with the aid of a desktop computer, a conventional flat-bed scanner and the freely available digital image analysis software programme, "Image J" using the following protocol:

Scanner Settings:

Scan images on a flatbed optical scanner at a resolution of 200 dpi.

Image Analysis:

1. Download Image J:
<http://rsbweb.nih.gov/ij/download.html>
2. Open root image via:
File → Open → Open desired scanned image
3. Convert scanned color image of the root to grayscale:
Image → Type → 8-bit
4. Set measurement scale:
Draw a line over section of the ruler. Then click **Analyze → Set Scale**
In *Set Scale* window enter the distance in mm selected on the ruler into the 'Known Distance' box and change the 'Unit of Measurement' box to mm, check 'Global'
5. Threshold new image of the root using manual settings:
Image → Adjust → Threshold and adjust the sliders to include all of the root in red and click 'Apply'. (The manual threshold setting includes all of the root area.)
6. Calculate area of the root:
Enclose the root with the freehand selection tool
Analyze → Analyze Particles
Use previous window settings and click 'OK'
7. The result is displayed in the "Results window" and the next image can be analysed.
Following analysis of a new image the newly acquired results are automatically added into the table in the "Results window".

The data were subjected to analysis of variance as the growth data were normally distributed. Means were separated using Tukey's test. The data were managed using MS Excel 2007 and analysed with GenStat Release 11.1 (Eleventh Edition).

5.4 Results

Coconut coir and perlite promoted germination percentage when compared to seedlings sown in composted pine bark seedling mix. Wall-less type containers also promoted germination percentage when compared to seed sown in walled containers. The Covetan and Hessian pockets filled with coconut coir produced the best germination combination for the six tray and two medium type combinations tested (Fig 5.2).

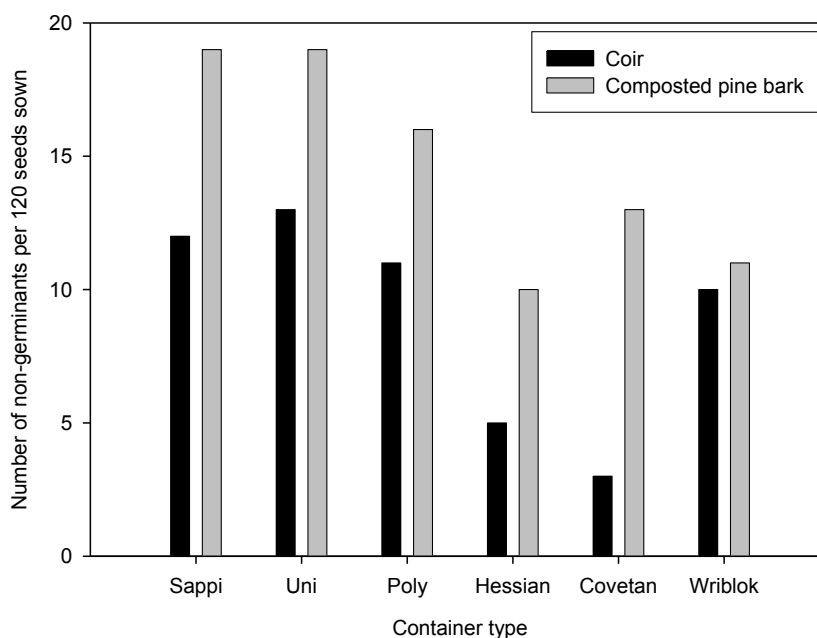


Figure 5.2: Number of non-germinants determined two weeks after sowing in six tray types and in two medium types. Figures were determined from a total of 120 seed per tray type and medium type

Greenhouse performance in terms of height is depicted in Figure 5.3. The overall best performing container type in terms of seedling height 10 weeks after sowing was the paper-maché plug. This was closely followed by the other five container types that performed similarly. All the plants grown in coconut coir performed better than plants produced in composted pine bark seedling mix.

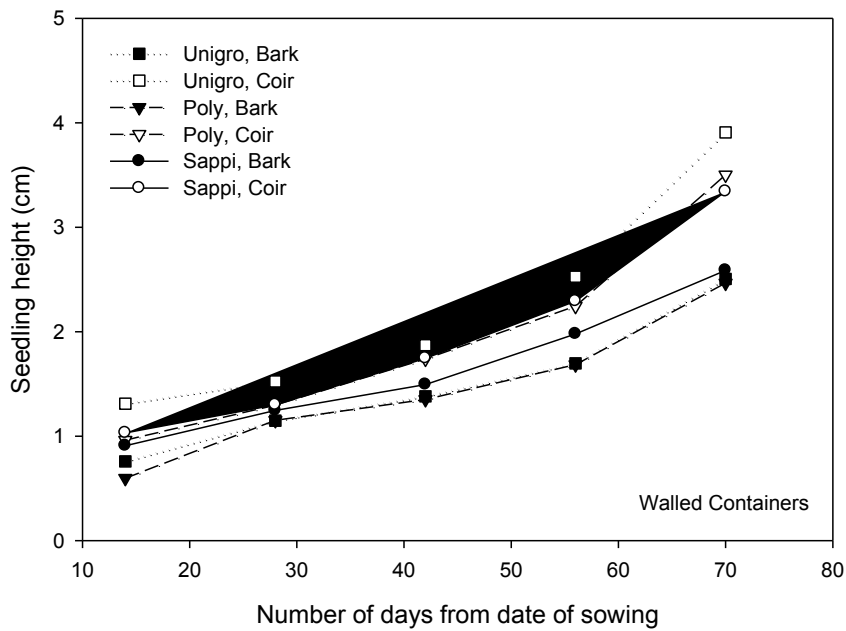
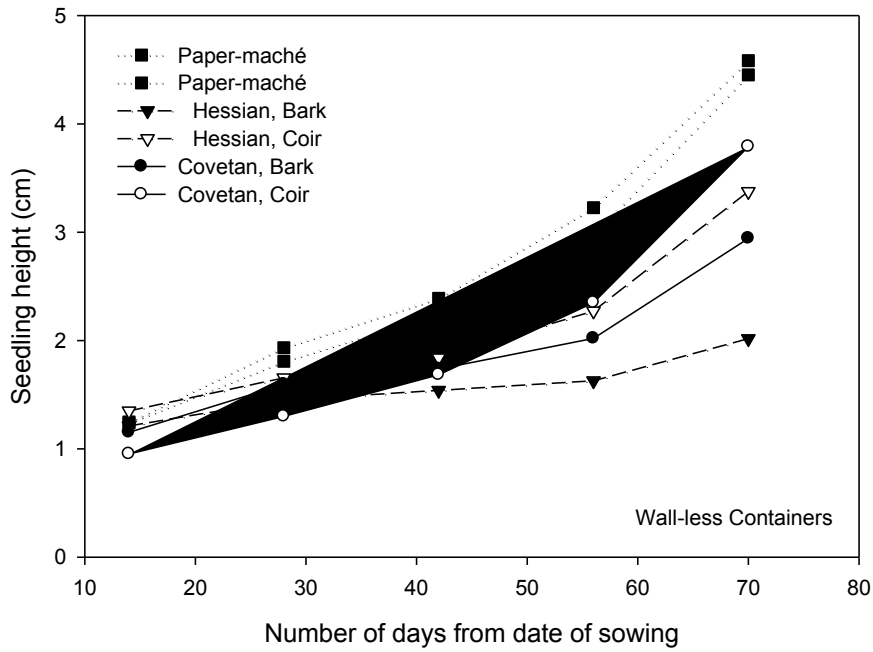
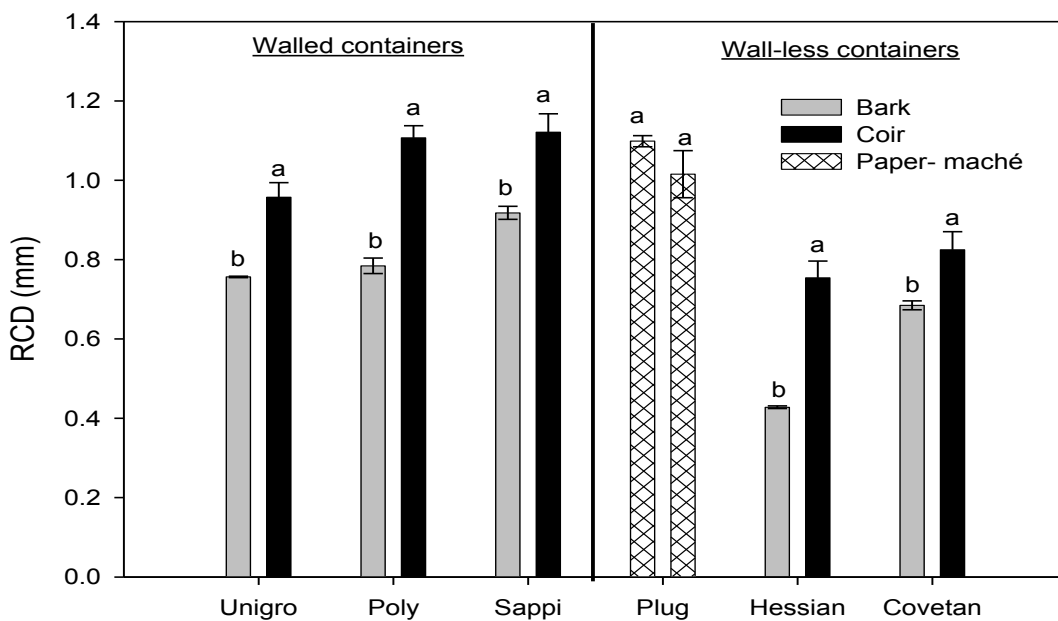


Figure 5.3: Growth of *E. grandis* seedlings exposed to various nursery treatments grown in composted pine bark and coir over a 10 week period

From the bar graphs in Figure 5.4 it can be seen that RCD measurements obtained 10 weeks after sowing produce results that follow a similar trend to seedling height results depicted in Figure 5.3. The overall best performing container type were the paper-maché plugs closely followed by the Sappi 49 and Poly Deep trays filled with coconut coir. The Covetan and Hessian pockets produced the most inferior plants in terms of RCD. Of the three walled containers tested the Sappi 49 produced the best results though closely followed by the Unigro and Poly 98 deep. With regards to



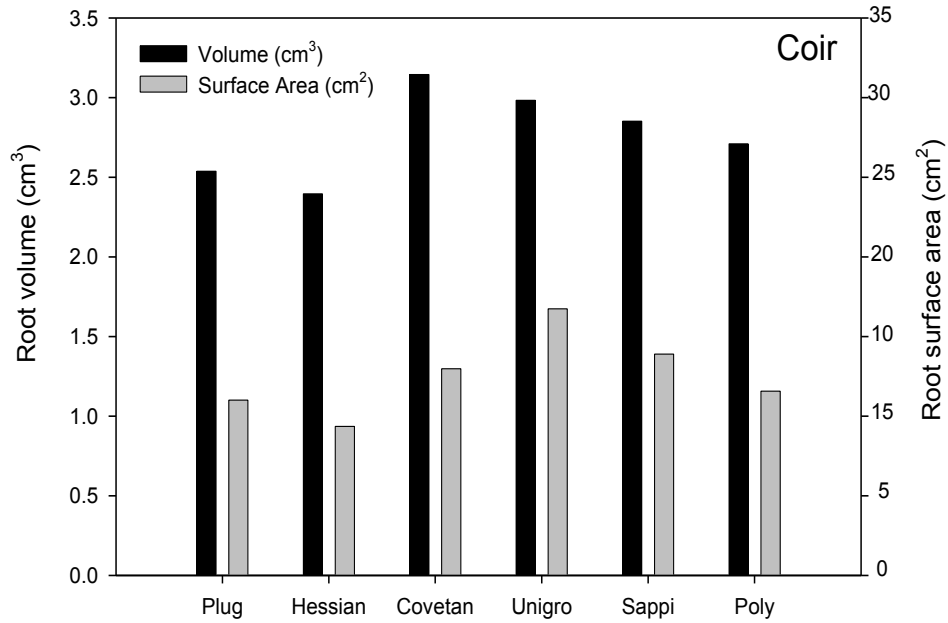
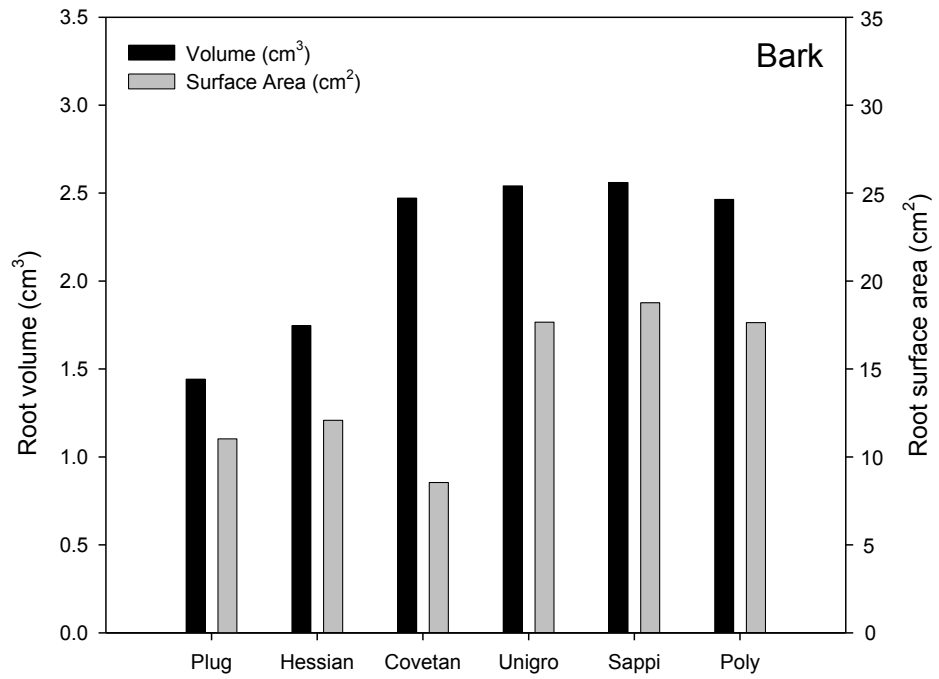


Figure 5.5: A comparison of surface area (cm²) and volume (cm³) of *E. grandis* seedling roots raised in six tray types and in two different media

The trend for root volume and root surface area measurements followed one-another closely with the exception of measurements taken on root systems of plants produced in Covetan pockets (Fig 5.5). From the graphs it can visually be seen that the two measurements correlate closely. An increase in root volume was always followed by an increase in root surface area. This result was obtained across the six tray types and for both medium types tested with the exception of the Covetan pockets. For plants produced in Covetan pockets root surface areas were lower than the expected when compared to the trend observed for plants produced in the other five container types for both medium types. Correlations carried out on the relationship between root volume and root surface area are reported in Table 5.2.

Table 5.2: Correlations (ρ) between seedling root Surface Area (mm^2) and Volume (cm^3) of 10 measurements of roots raised in all six tray types with or without the Covetan treatment (Correlations of the data depicted in Figure 5 5)

	Bark	Coir
All treatments	0.54	0.77
All treatments but no Covetan	0.99	0.96

5.5 Discussion and conclusions

The observed improved germination and early establishment in coconut coir (Figure 5.2) is most likely attributable to improved physical properties of this medium such as increased water-holding capacity without a reduction in air-filled porosity. With regards to container type, improved germination in wall-less containers may be attributable to an increased air-filled porosity compared to this characteristic of the same media in walled containers. The relatively high number of non-germinants observed in the paper-maché plugs is probably due to the resistance exerted on the young radical upon its attempts to penetrate the relatively dense and tightly held together medium of this plug.

The greater height and RCD of plants grown in coconut coir can probably also be attributed to improved physical properties, especially the greater water holding capacity of this medium. The poor performance of plants raised in the Hessian and Covetan pockets is likely due to the increased evaporation of water from these plugs due to the large surface area from which moisture can be lost. This however remains to be further investigated since plants raised in the paper-maché plugs performed well and were subjected to the same potential limiting factor. The better performance of plants established in the paper-maché plugs warrants further investigation since the other two types of wall-less plugs filled with composted pine bark produced significantly inferior results. An investigation of the air filled porosity and the water holding capacity of these various media may prove useful in this regard.

Volume and root surface area measurements correlated well for all tray types with the exception of Covetan pockets. The likely explanation for this is that a large number of fibrous roots were lost during the removal of the Covetan material compared with the amount lost from Hessian and Paper-maché plugs. This is possibly due to the nature of the Covetan material in that the material is tightly woven and does not readily break down unlike the Hessian material, which is more loosely woven and breaks down at a substantially faster rate. Since the paper-maché plugs have no fabric pocket keeping them together less fibrous roots are lost when carefully removing the paper-maché plugs from the roots in preparation for the root analysis. These fibrous roots scored highly in the surface area measurements using image analysis as they were in proportionally greater numbers than the larger diameter roots that remain of the root system after the removal of the Covetan material. Hence a single large root will produce a large volume measurement for a relatively small surface area measurement and a fibrous root system will produce a large surface area measurement for a relatively small volume measurement. However, for root systems with both large and fibrous roots this effect is minimized and the two measurements correlate well.

This method of quantifying the size of seedling root systems may prove useful for nursery managers if in addition to size determination a benchmark figure is available to the nursery manager to which he can compare the seedling's root system under investigation. The Image J software is freely available from the internet and a standard computer and a flat-bed scanner are the only other hardware components required to successfully implement this method. This technique can thus be

adopted with relative ease to help nursery managers decide on the fitness of seedlings for dispatch to the field with the aim of minimizing the need for blanking and maximizing early growth and subsequent stand development.

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CHAPTER 6 CONCLUSIONS AND RECOMMENDATIONS

Commercial plantation forests play a significant role in South Africa's economy with just over 1.3 million ha currently cultivated (DWAF, 2005). Forestry products and related services contributed 1.54% to the country's GDP in 2003 (R12.2 billion), employing over 170 000 people in areas where little alternative employment exists (Chamberlain *et al.*, 2005). Products derived from the timber industry include pulp, paper and solid wood products, earning R270 million in exports in 2003 (DWAF, 2005). Over and above the contribution made to the economy, both natural and commercial forests provide invaluable environmental services. For example, through photosynthesis, forests act as an atmospheric filter replacing carbon dioxide with oxygen (Beedlow *et al.*, 2004). Carbon is stored in both above and below ground biomass as well as dead wood and litter. It is reported that over 900 million metric tons of carbon is stored in forests within the borders of South Africa (FAO, 2005) with forestry activities offsetting approximately 3.8% of carbon dioxide emissions each year (Christie and Scholes, 1995).

The sector has come under increasing pressure since the early 1970's and especially so after the first democratic elections in 1994 regarding the environmental impacts of timber plantations even though the benefits of the forestry industry in terms of economics and job creation are well known (Tewari, 2001). Several impacts are now recognised, including a loss of biological diversity (Heydenrych, 1995; Richardson, 1998), land degradation and associated soil compaction (Brink, 1990), as well as stream flow reduction (Schonau and Grey, 1987; Scott and Lesch, 1997). The environmental concerns culminated in the replacing of the old forestry permit scheme with a licensing system called the 'stream flow reduction activity' (SFRA) regulated under the National Water Act (No 36 of 1998). Currently afforestation is the only agricultural activity declared an SFRA and is seen as a limiting factor in terms of expansion within the forestry industry in the Eastern Cape and KwaZulu-Natal (Chamberlain *et al.* 2005). Due to fast growth and high yields the industry has an international competitive advantage. In spite of this, cheap imports from emerging growers in Asia and South America threaten to flood the international market and significantly reduce profit margins. Within the context of sustainable and environmentally friendly development the timber industry therefore needs to reduce operating costs while satisfying an ever vigilant environmental lobby striving to highlight any mismanagement on the part of commercial growers. To achieve this, the industry should explore the potential benefits of using new and emerging technologies to streamline the

forestry processing chain and minimise costs where necessary. One such area is that of forest regeneration which may be achieved more efficiently and in a more cost effective manner through the successful implementation of biodegradable wall-less containers for the production and subsequent outplanting of planting stock. It is interesting to note that most of the motivating drivers for the development of such a novel and innovative container system fall within the realms of lean manufacturing, lean enterprise, lean production, or often simply as, "Lean" (Jones and Womack, 2003).

The Toyota Production System (TPS) is an integrated socio-technical system, developed by Toyota, that comprises its management philosophy and practices (Jones and Womack, 2003). The TPS organizes manufacturing and logistics for the automobile manufacturer, including interaction with suppliers and customers. The system is a major precursor of the more generic "Lean manufacturing." Taiichi Ohno, Shigeo Shingo and Eiji Toyoda developed the system between 1948 and 1975. Essentially, lean is centered on preserving value with less work. It considers the expenditure of resources for any goal other than the creation of value for the end customer to be wasteful, and thus a target for elimination. Working from the perspective of the customer who consumes a product or service, "value" is defined as any action or process that a customer would be willing to pay for. Lean manufacturing is a variation on the theme of efficiency based on optimizing flow; it is a present-day instance of the recurring theme in human history toward increasing efficiency, decreasing waste, and using empirical methods to decide what matters, rather than uncritically accepting pre-existing ideas (Jones and Womack, 2003).

A best practice, in addition to the notions of lean manufacturing, is a method or technique that has consistently shown results superior to those achieved with other means, and that is used as a benchmark (Carter *et al.* 2001). Best practices are generally-accepted, informally-standardized techniques, methods or processes that have proven themselves over time to accomplish given tasks. A "best" practice can evolve to become better as improvements are discovered. "Best practice" is considered by some as a business buzzword, used to describe the process of developing and following a standard way of doing things that multiple organizations can use. Best practices are used to maintain quality as an alternative to mandatory legislated standards and can be based on self-

assessment or benchmarking. Best practice is also a feature of accredited management standards such as ISO 9000 and ISO 14001 (Carter *et al.* 2001).

The development of a wall-less plug for planting stock of forest trees has the potential of enabling the production of planting stock with the following advantages over the production of planting stock in containers:

1. reducing the need for blanking, improved early growth and the loss of timber from windthrow due to the production of better quality planting stock;
2. no need for the purchase of containers that are re-usable but do not have an unlimited lifespan and thus require subsequent replacing, at a substantial initial capital outlay;
3. no need for ensuring the transport of empty containers back to the nursery after outplanting in the field provided the following can be successfully developed and implemented:
 - a. a support for the wall-less plugs with their developing seedlings while still in the nursery;
 - b. a transport system for seedlings from their place of growth in the nursery to the point of outplanting in the field.

The above potential improvements as a result of the successful development and implementation of such a wall-less plug, following careful consideration of the principles of lean manufacturing, may well lead to a new method of planting stock production. It may also ultimately be adopted as the method of best practice by the forestry community in South Africa.

The best known commercially available wall-less plugs for the production of planting stock of forest trees are the Jiffy plugs manufactured by Jiggy Plugs®. These plugs have been manufactured by the Swedish company since 1948. Their operations have subsequently expanded to factories ensuring production in the USA, Canada and Europe. In South Africa, the use of wall-less plugs for forest regeneration has thus far never been implemented by timber producers though several investigations have been made. Jiffy Plugs, Ellepots and WRIBLOKS are three types of wall-less plugs. Their use for forest regeneration in South Africa over the past three decades has been seriously considered by forestry companies, private nurseries and research institutes. For different reasons those past

investigations have indicated that the gains achieved following their implementation would not outweigh the costs thereof.

Forest regeneration is currently successfully achieved by means of planting stock that is produced in large scale containerized nurseries. This may be achieved more efficiently and at a reduced cost through the development and implementation of a wall-less plug. However, it is critical that replacing the current system by the production of planting stock using new technology is decided upon only after identifying and quantifying every component of each system. Only that can ensure that, overall, the advantages in monetary terms of any new system outweigh the costs of the current way of achieving forest regeneration.

In this study three experiments were conducted in the form of greenhouse trials in which the performance of different container types was evaluated in terms of their ability to produce *Eucalyptus* seedlings. Two fundamentally different container categories were evaluated and within each several types of containers were tested. The first container type tested were walled containers that have in the past or are still currently being used by both private nurseries and nurseries of commercial forestry companies. The second tray type tested was wall-less containers. The distinction between these two types of trays types is the ability of the seedling roots to penetrate through the wall of the container. Roots are only able to penetrate wall-less containers.

In the first trial, the performance of three different walled containers was compared with the performance of one type of wall-less container. In the second and third trials, the performance of three walled containers was compared with that of three types of wall-less containers. In addition to testing the greenhouse performance of the different tray types, trials two and three each had an additional research component associated with them. In trial two the outplanting performance of the seedlings was evaluated and in trial three differences in the shape and size of the seedling root systems were evaluated using image analysis software and scanned images of root systems.

The results of the three trials show that seedlings can be raised in wall-less plugs that are locally produced. The greenhouse performance (growth in terms of height and RCD) of such wall-less plugs varies depending on the kind of materials used for their manufacture. The performance of the paper-

maché plugs, produced according to the original WRIBLOK protocol as devised by Zeijlemaker and de Laborde (1977) was the most successful of the four prototypes produced and tested in the three trials described in this dissertation. Their performance was similar to that of plants raised in the Unigro 128, Sappi 49 and Poly 98 deep containers currently used by timber companies for the production of their planting stock. This research has shown that developing a wall-less plug that performs well can be achieved.

The results of the outplanting stage of trial two yielded no significant differences in outplanting survival due to differences in the container types used for raising the seedlings. This indicates that in order for the influences of the container characteristics on the ability of a seedling to survive outplanting to be observable, more stressful site conditions may be required.

In Trial Three, with the exception of the paper-maché plugs the remaining five container types were filled with both composted pine bark and coconut coir. Of the two media types, coconut coir was consistently the better performing medium in terms of both number of non-germinants and growth of the seedlings in the greenhouse.

Of the methods available to quantify the size and characterize the shape of seedling root systems some have been more successful than others. Volume and mass measurements remain the two most widely used methods for quantifying the size of seedling root systems whereas no readily available and practical method is currently available to characterize the root architecture or branching patterns of root systems as complex as those of eucalypt seedlings. In this research Archimedes' principle of water displacement to determine the size of root systems was compared with another elegant method that makes use of freely available computer software and scanned images of seedling root systems. Though this method may have some merit, a precise measurement of the size of the root system in this way is not possible due to the measurement of the two-dimensional surface area of the root system as an indicator of its size rather than its actual size. The merit of this method thus stems from the very strong correlations obtained between root surface area determined by image analysis software and root volume measurements obtained using Archimedes' principle of water displacement.

In this research no progress was made in the ability to describe how one root system differs from another in terms of the branching patterns of root systems. Thus the hypothesis that wall-less plugs produce root systems that are more highly branched with a larger number of fibrous roots, could not be tested.

Listed in Table 6.1 are a number of important aspects of forest regeneration that will need to be quantified to ensure the implementation of a wall-less plug with the certainty that the anticipated gains of the new system are actually achieved. Quantifying some of the costs in Table 6.1 is difficult and others not yet possible due to the early stage of development of this technology. Estimating the cost of technology that will ensure the handling of planting stock in the nursery and its transport from the nursery to the point of outplanting can at this stage not be done. Challenges associated with determining the loss of timber due to windthrow as a result of poor seedling root architecture is twofold; 1) trees become affected by windthrow typically only several years into their growing cycle and thus obtaining such data is a decidedly long-term exercise and; 2) though poor seedling root architecture is often the cause for the loss of timber that is affected by windthrow this is not always the case. Windthrow can result from excessively strong winds that dislodge trees produced from planting stock devoid of inferior root system quality. Identifying the cause for windthrow correctly is important when considering data of this nature for the purpose of deciding on the implementation of one regeneration system over another.

The large-scale production of a wall-less plug that has a greenhouse performance equal or possibly superior to the greenhouse performance of seedlings produced in the Sappi 49 or Unigro 128 containers is almost certainly possible. Further research into the use of various new ingredients in combination with an exterior support in the form of a material through which roots can grow and the development of specialized equipment to ensure their efficient manufacture will almost certainly make this possible.

Table 6.1: Important considerations associated with regeneration by means of planting stock produced in containers and in wall-less plugs

<u>Aspects common to both systems</u>
<i>Placing the cutting or seed in a wall-less plug or container cavity</i>
<i>Watering</i>
<i>Fertigation</i>
<i>Pest management</i>
<i>Transport to the field</i>
<i>Pit preparation</i>
<i>Planting</i>
<i>Blanking</i>
Timber loss from windthrow due to poor seedling quality
<u>Aspects specific to planting stock produced in containers</u>
<i>Return of empty containers back to the nursery</i>
<i>Washing empty containers</i>
<i>Copper dipping of polystyrene containers</i>
<i>Filling the container with growing medium</i>
Container
Growing medium
<u>Aspects specific to planting stock produced in wall-less plugs</u>
<i>Root pruning to prevent cross-over of roots from one plug to adjacent plugs</i>
Wall-less plug
Technology that enables handling of wall-less plugs in nursery
Technology that enables transport of planting stock to the point of outplanting

Italicised items are operational procedures that require execution. Some operational procedures require materials that need purchasing. Such materials are not listed in the table but their cost should be included into the operational cost. Non-italicised items are loss of product to the grower or materials that need purchasing

Extensive further research however will also be required for the development of technology that will enable:

- 1) the wall-less plugs to be supported in the nursery;
- 2) transport of the wall-less plugs to the field;
- 3) transport of the wall-less plugs from their point of delivery in the field to the pit into which they will be planted.

No attempt has been made in this study, to develop the above technology with the exception of the waterproofed egg trays that provided support for the Paper-maché plugs, the Hessian and Covetan plugs and the Unigro carriers for the Sponge blocks. The successful development thereof is however critical to achieve the implementation of a system that negates the need for the return of empty containers to the nursery. The support and handling of planting stock in the nursery can be achieved by means of plastic carriers. Amongst other solutions, for transport to the field, the use of biodegradable cardboard boxes that could be left to decompose in the field can be investigated. Alternatively, if a method of delivery of planting stock in the field can be devised that allows the immediate return to the nursery of any carriers that are used, then the carriers used for handling the planting stock in the nursery can also be used for transport of the plants in trucks or trailers to the field. Subsequent transfer of plants from their point of delivery in the field to their place of outplanting may then be achieved by means of specially designed backpacks.

Provided the performance of planting stock can be determined on the basis of root volume, seedling height, root collar diameter and shoot, stem and root dry and fresh weight at various stages of growth in the greenhouse and on outplanting survival without the need to quantify root system architecture, the development of a wall-less plug that performs equally well or possibly better than the Unigro and Sappi 49 containers is probably achievable. If the performance of planting stock produced in different types of containers must also be evaluated on the basis of the container's ability to reduce the need for blanking and the number of trees affected by windthrow due to improved root system architecture then more resources are first required for the development of techniques that will enable that kind of evaluation to be done.

In conclusion, the development of a wall-less plug that performs equally well or possibly better than the Unigro and Sappi 49 containers can be taken forward at this point provided the performance of planting stock can be assessed on the basis of standard, currently available, methods of seedling evaluation and data on outplanting survival and early growth of planting stock. However, if the performance of planting stock produced in different types of containers must also be evaluated on the basis of the container's ability to reduce the need for blanking and the number of trees affected by windthrow due to improved root system architecture, then more resources are first required for the development of techniques that will enable that kind of evaluation to be done. Such research is

not only a decidedly long-term exercise but also one that requires the development of techniques that are able to accurately make the link between seedling root architecture and outplanting survival, early growth and resistance to windthrow. Currently no easy to use and well-established techniques are available for any of these steps. Furthermore, such research is wholly dependent, not only on the development of such techniques, but also on the availability of accurate data of variables including container type, species, age of planting stock, type of planting stock (plants produced from seed or from cuttings), soil type, early growth, number of plants lost following outplanting, and number of trees affected by windthrow. This means, for such a research initiative to be successful, the input of reliable data from all parties involved in the regeneration process would be required.

Forest regeneration research is fascinating and through innovation allows the adoption of continuously improving high-tech techniques to suit our specific needs. Such techniques are at times developed for applications in engineering fields unrelated to Biology. Ultimately further research and the adoption of such techniques will deepen our understanding of the principles that underline plant growth regulation and development. Why plants develop into the shapes that they ultimately adopt is a fascinating question, with no simple answer but with benefits to both our understanding of plant development biology and to a wide range of plant related industries. A better understanding of plant developmental biology will enable us to better manipulate plant growth to suite our needs.

Finally, noteworthy also, is the interest shown in this research by forestry companies and private nursery growers alike. That interest in this research provides evidence for the availability of an opportunity to develop a product and a system that will allow a more streamlined production of good quality, and possibly superior quality, planting stock of forest trees in a more efficient and cost effective manner than that currently produced in reusable solid-walled containerized nurseries. The future successful implementation of a biodegradable wall-less plug for the production of planting stock of forest trees is ultimately likely to be adopted as the method of best practice in South Africa.

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APPENDIX
PAPER(S) IN PRESS

ORIGINAL ARTICLE

Potential use of papier mâché plugs for eucalypt seedling production in South Africa

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Abstract

Rooting plugs were made by (1) mixing, pressurizing and gluing papier mâché (plugs), (2) cutting a sponge block, and (3) filling Hessian bags with standard growing medium. These treatments were also compared with three types of container used in commercial nurseries, giving three additional treatments: (4) Unigrow, (5) Sappi 49, and (6) Poly 98 deep. *Eucalyptus dunnii* and *Eucalyptus grandis* plants were raised from seed over 70 days. In each species and each treatment 60 plants were outplanted to determine survival and growth of the trees after 9 weeks. The nursery results showed that plugs yielded the tallest plants for *E. grandis*, and Sappi 49 resulted in the tallest *E. dunnii*. The sponge blocks produced significantly smaller trees in both species. After outplanting, the survival of trees was high for each treatment. There is a need to test performance attributes of the various types of planting stock under stressful site conditions to determine long-term benefits, if any, of producing planting stock in papier mâché plugs. If these studies are concluded successfully, papier mâché plugs could replace peat in commercial forest nurseries, preventing environmental degradation of peat-mined wetlands.

Keywords: container type, containerized nurseries, root plug, wall-less plug.

Introduction

The degree of outplanting performance of planting stock determines the success of any establishment operation. Postplanting survival and rate of early growth are the two most obvious measures of regeneration success. Losses in value and volume yield together with increased regeneration costs can be minimized through efficient plantation establishment (Albert et al., 1980). This in turn depends on seedling morphological and physiological characteristics that meet targets associated with favourable performance under an anticipated range of site conditions (Davis & Jacobs, 2005).

Over the past couple of decades, containerized nurseries replaced their bareroot counterparts in many parts of the world, especially where plantation forestry operations depend on effective establishment and the fast growth of trees. Containerized nurseries helped also to accelerate propagation

techniques, especially vegetative propagation, and to reduce prices of the planting material of select genotypes of trees. Despite the fact that these objectives had been largely achieved, many planting operations failed owing to early tree dieback or vulnerability to wind in older forest stands, both resulting from root deformations and stem strangulation of the containerized planting stock with roots constrained by hard container walls (Chapman et al., 2003; Rune, 2003). In many studies worldwide the root collar diameter (RCD) of seedlings has been found to correlate strongly with field performance after planting (Thompson, 1985; Bayley & Kietzka, 1997; Jacobs et al., 2005). Diameter of the stem at root collar, however, depends on the amount of space allocated to roots during the nursery stage. The older the seedlings the greater the root collar, but inferior planting stock may result from prolonged cultivation in combination with

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insufficient rooting space. McCubbin and Smith (1991) showed the importance of recognizing the “planting windows” for *Eucalyptus grandis* seedlings grown in different containers. They concluded that delaying planting will have a worse effect on postestablishment growth than planting younger seedlings before their optimum quality is reached. This was confirmed by other studies which showed no benefit from planting large-diameter containerized seedlings (Morris, 1994). A possible explanation could be that the overgrown seedlings do not necessarily facilitate a well-branched and functioning root system. Well-performing planting material must be characterized by multiple and actively growing root tips for improved soil colonization and root–soil contact (Thompson, 1985; Deans et al., 1990). Whatever findings emerge from various studies it has been agreed that root architecture derived from nursery cultural practices cannot be effectively corrected and has a lifelong impact on tree performance (Moore, 2003).

A well-designed container should promote branching of the tap root when reaching the sides of a container. For drainage, most containers are equipped with an opening at their lowest point and therefore the tap root is usually air-pruned and branched. The lateral roots, however, may be deflected upon reaching the container walls and then continue growing by following the container shape. In South Africa such root deformations are called the “caging effect”. After outplanting root circling and continued growth around tree stems may result in stem strangulation and breakage at the root collar in later stages of tree development. Deformed roots disrupt nutrient and water transport in the affected trees (Hay & Woods, 1975). The design of containers involves changing their shapes, incorporating grooves, slits or ridges into container walls to create a mechanical barrier or to stimulate air-pruning of the lateral roots. Most of these designs, however, still incorporate smooth container walls which result in undesirable root deformations. The introduction of copper-based chemicals, coated onto the container walls for root pruning (Bayley & Snell, 1997), can be effective but is environmentally unfriendly.

Beyond the two valuable works on the subject by Barnett and Brisette (1971) and Landis (1990) there is an abundance of published research articles describing a number of specific containers in relation to root development, plant quality and their performance (Harris, 1967; Whitcomb, 1988; Appleton, 1989, 1993, 1995; Arnold & McDonald, 1999). Several different systems for categorizing containers have been used. A practical system divides containers into two functional groups: those that are planted with the tree seedling and those that are removed

before the seedling is planted (Tinus & McDonald, 1979). The first category includes containers that are biodegradable and result in minimum root disturbance at planting. Containers that constitute both the container and the growing medium as a complete unit are generally known as blocks or plugs.

The objective of this study was to develop a wall-less plug for growing eucalypt seedlings and to compare the performance of these seedlings with planting stock raised in standard containers used in South African nurseries.

Materials and methods

The first attempt at making a wall-less plug prototype was made using improvised tools and the original WRIBLOK protocols for making the papier mâché mix (Zeijlemaker & de Laborde, 1977). A tool was designed consisting of a drill press with a piston to extract excess water from the papier mâché mixture. This compression tool was used together with a Sappi 49 container which served as a mould to produce plugs equal in shape and volume to the Sappi 49 cavity. The Sappi 49 tray consists of 49 bullet-shaped cells, each of dimensions specified in Table I. A 550 W electric drill mounted with a paint mixer was used at approximately 600 rpm to produce the papier mâché from shredded newspaper, composted bark, charcoal dust and water mixed in the proportions 300 g, 2040 g and 600 g and 7 litres, respectively. The resulting papier mâché mixture was poured into the Sappi 49 tray and the largest possible quantity of water was removed. The wet plugs were carefully pushed out of the Sappi 49 mould and left to dry. Then they were placed on commercial egg holders to keep them upright. The other innovative methods included a hessian bag filled with composted pine bark used for seedling production in the local nurseries and blocks of sponge cut from sponge of density 57.7 g dm^{-3} . Egg holders were also used for holding the hessian bags and the sponge blocks were inserted and thus supported with the Unigro carrier. These treatments were compared with commercial containers used by forestry nurseries. Their characteristics are provided in Table I.

Table I. Dimensions of the containers and plugs used in the study.

Treatment	Depth (mm)	Volume (cm ³)	Cross-sectional shape
Papier mâché	58	80	Round
Hessian bag	58	80	Round
Sponge block	110	200	Square
Unigrow 128	80	60	Square
Sappi 49	58	80	Round
Poly 98 deep	66	63	Round

All the walled containers and the hessian bags were filled with standard composted bark. The treatments were replicated five times in a randomized complete block (RCB) design. Each experimental unit consisted of 12 cells, giving a total of 60 prospective seedlings per treatment and species. Seed of *Eucalyptus grandis* and *Eucalyptus dunnii* were sown in each cavity. The sown trays were initially inserted in black polyethylene bags and placed in a heated greenhouse for the initiation of germination. After 96 h (4 days) the trays were taken out of the polyethylene bags and placed in a glasshouse on benches with overhead sprinkler fertigation. The third week after seed started emerging, fertilizing began with applications of Agrofert Orange (13.2% N/17.6% P/11.0% K) at a concentration of 0.6 g l^{-1} three times a week through the fertigation system. Seedlings were fertigated to saturation. In addition, Calmag (13.7% N/11.7% Ca/38 g kg^{-1} Mg/2000 mg kg^{-1} B) was applied once a week at 0.4 g l^{-1} until seedlings were saturated. This fertilizing regime was followed until seedlings were ready for planting.

The seedlings remained in the glasshouse for the remainder of the experiment until the time of outplanting into the field (70 days). The temperatures in the glasshouse reached up to 30°C on very warm days and dropped to 15°C on cold nights. The plants were cultivated for 10 weeks in the nursery and then planted in the field. During the outplanting period temperatures reached 28°C in the field on very warm days and dropped to a few degrees above freezing. Freezing temperatures were prevented by overhead irrigation during very cold nights. Every 2 weeks after planting tree heights and RCDs were measured. A total of 60 plants from each treatment was outplanted for each of the tested species. These were divided into three lots of 20 plants in an RCB design and grown for 9 weeks. After that period surviving trees were counted and carefully excavated. Their heights, RCDs and dry mass of shoots and roots were determined. The data were subjected to analysis of variance as the growth data were normally distributed. Means were separated using Tukey's test. The data were managed using MS Excel 2007 and analysed with GenStat Release 11.1 (11th ed.).

Results

The papier mâché plugs improved the seedling height in comparison with the other two types of plugs examined. However, these results were not significantly different from hessian bags (Table II, Figure 1). The papier mâché plugs significantly showed the highest RCD compared with all the other containers. Even though in many cases the results were not significantly different from the other

container types, the papier mâché plugs achieved the highest shoot and root dry mass (Table II).

In the outplanting experiments performance of the six container types was evaluated in terms of both survival and growth. Outplanting survival was close to 100% for all container types with the exception of sponge blocks, which performed significantly worse. With regard to growth, *E. dunnii* and *E. grandis* generally both performed similarly, with the exception of height and root dry mass.

The combined results of both species show that the seedlings raised in papier mâché plugs transplanted in the field showed an increase in height, RCD and shoot dry mass in comparison with the other five container types evaluated (Table III). This tray type significantly showed greater root dry mass than other trays, with the exception of hessian bags (Table III). In most cases, the outplanting of *E. dunnii* and *E. grandis* seedlings grown in papier mâché plugs, i.e. Species \times Container, recorded higher values for all growth parameters studied. However, there were a few exceptions.

Discussion

The plants grown in sponge blocks were significantly smaller than seedlings produced in the other container types for both species of eucalypts (Figure 1, Table II). It is possible that relatively large holes prevented sufficient water absorption and storage. In addition to this, sponge blocks do not have an available nutrient source owing to their inert nature. Watering and fertigation of these various types of plugs was a challenge as all of them were exposed to the same irrigation system, but it was observed that wall-less plugs dried out quickly. This can be attributed to a greater evaporation since the rooting material was not protected by solid walls. Despite this, seedling characteristics remained compatible among the treatments (Table II). However, the water requirement of wall-less plugs needs further investigation.

The field performance of trees is shown in Table III. *Eucalyptus dunnii* yielded higher trees than *E. grandis*. Perhaps this is because they were cultivated during winter and *E. dunnii* is a better performing species under cold conditions than *E. grandis*. *Eucalyptus grandis* produced more roots than *E. dunnii*. The survival of trees was high and inconclusive results were obtained regarding tree stability after planting. Papier mâché-treated plants performed better than those in sponge blocks and Poly 98 deep trays. Similar trends were recorded within species responses to nursery treatments.

It is likely that for the first time unconventional materials, such as rubber sponge and papier mâché,

Table II. Characteristics of *Eucalyptus dunnii* and *Eucalyptus grandis* seedlings by nursery treatment 70 days after sowing.

Treatment	Height (cm)	Root collar diameter (mm)	Shoot dry mass (g)	Root dry mass (g)
Species				
<i>E. dunnii</i>	11.2±4.4 ^a	1.9±0.5 ^a	1.11±0.3 ^a	0.44±0.2 ^a
<i>E. grandis</i>	10.4±2.7 ^a	1.8±0.4 ^a	0.89±0.2 ^b	0.28±0.1 ^b
Container				
Papier mâché	12.2±2.4 ^a	2.2±0.7 ^a	1.25±0.3 ^a	0.45±0.1 ^a
Hessian bag	10.3±5.6 ^a	1.9±0.5 ^b	0.85±0.1 ^b	0.35±0.1 ^{ab}
Sponge block	6.1±3.4 ^b	1.5±0.5 ^c	1.00±0.3 ^{ab}	0.24±0.2 ^b
Unigrow 128	12.2±2.3 ^a	1.8±0.3 ^b	1.04±0.1 ^{ab}	0.32±0.1 ^{ab}
Sappi 49	12.5±2.9 ^a	2.0±0.4 ^b	1.00±0.1 ^{ab}	0.44±0.1 ^{ab}
Poly 98 deep	11.4±4.6 ^a	2.0±0.3 ^b	0.82±0.1 ^b	0.37±0.1 ^{ab}
Species × Container				
<i>E. dunnii</i> × Papier mâché	12.0±3.3 ^a	2.0±0.6 ^{ab}	1.42±0.5 ^a	0.58±0.2 ^a
<i>E. dunnii</i> × Hessian bag	10.9±7.3 ^{ab}	2.0±0.6 ^{ab}	0.98±0.2 ^{ab}	0.40±0.1 ^{abcd}
<i>E. dunnii</i> × Sponge block	6.6±5.6 ^b	1.5±0.5 ^{bc}	1.10±0.4 ^{ab}	0.34±0.3 ^{abcd}
<i>E. dunnii</i> × Unigrow 128	12.4±2.9 ^a	1.9±0.3 ^{abc}	1.12±0.2 ^{ab}	0.38±0.1 ^{abcd}
<i>E. dunnii</i> × Sappi 49	12.8±3.4 ^a	2.0±0.5 ^{ab}	1.06±0.1 ^{ab}	0.40±0.1 ^{abcd}
<i>E. dunnii</i> × Poly 98 deep	12.3±3.7 ^a	2.0±0.3 ^{ab}	0.98±0.3 ^{ab}	0.55±0.1 ^{ab}
<i>E. grandis</i> × Papier mâché	12.4±1.4 ^a	2.4±0.7 ^a	1.07±0.1 ^{ab}	0.32±0.1 ^{abcd}
<i>E. grandis</i> × Hessian bag	9.8±3.9 ^{ab}	1.7±0.4 ^{bc}	0.75±0.1 ^b	0.31±0.1 ^{abcd}
<i>E. grandis</i> × Sponge block	5.7±1.3 ^b	1.4±0.4 ^c	0.90±0.3 ^{ab}	0.14±0.1 ^d
<i>E. grandis</i> × Unigrow 128	10.4±1.6 ^{ab}	1.7±0.2 ^{bc}	0.95±0.1 ^{ab}	0.26±0.1 ^{bcd}
<i>E. grandis</i> × Sappi 49	12.3±2.4 ^a	1.9±0.3 ^{abc}	1.03±0.2 ^{ab}	0.47±0.1 ^{abc}
<i>E. grandis</i> × Poly 98 deep	12.1±5.5 ^a	1.9±0.3 ^{abc}	0.66±0.1 ^b	0.18±0.1 ^{cd}

Note: Means values (±SD) for species, container and species × container in each column with different letter(s) are significantly different ($p < 0.05$) according to Tukey's test.

have been tried to grow eucalypt seedlings. Papier mâché was shown to be an appropriate substrate on which to grow seedlings when the standard fertigation regime was applied. In the past it was used to grow black wattle (*Acacia mearnsii*) seedlings (Barrett, 1978). It is possible that seedling growth could be even better by adjusting watering and nutrition, and further studies are recommended in

this regard. It is also uncertain whether seedling quality could be improved in terms of rooting structure. Seedlings cultivated in papier mâché plugs were similar to their counterparts prepared in the standard commercial nursery containers. They also performed equally well under field conditions. A range of seedling size and age grades would need to be tested on a range of stressful sites to show

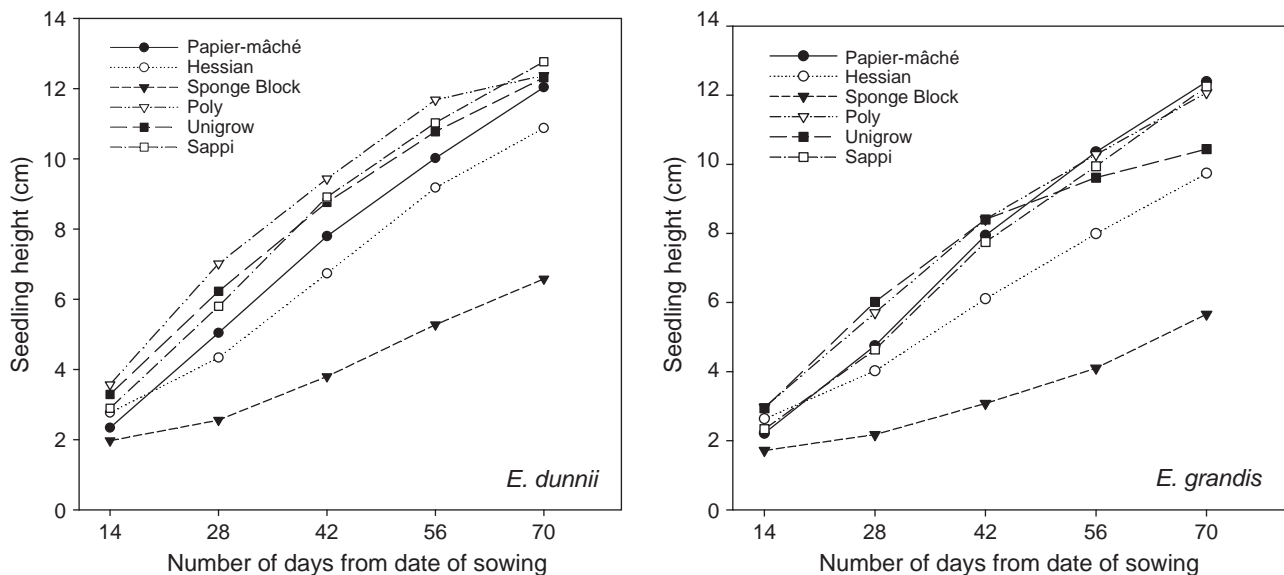


Figure 1. Growth of *Eucalyptus dunnii* and *Eucalyptus grandis* seedlings exposed to various nursery treatments.

Table III. Characteristics of *Eucalyptus dunnii* and *Eucalyptus grandis* trees by nursery treatment after 63 days' (9 weeks') growth in the field.

Treatment	Survival (%)	Height (cm)	Root collar diameter (mm)	Shoot dry mass (g)	Root dry mass (g)
Species					
<i>E. dunnii</i>	96.7 ^a	41.4±5.8 ^a	5.7±0.6 ^a	39.9±13.9 ^a	6.9±2.2 ^b
<i>E. grandis</i>	95.0 ^a	35.2±6.4 ^b	5.9±0.9 ^a	49.8±22.8 ^a	8.1±3.9 ^a
Container					
Papier mâché	97.5 ^a	42.9±5.1 ^a	6.6±0.9 ^a	60.5±24.8 ^a	9.9±4.3 ^a
Hessian bag	96.7 ^a	38.2±3.1 ^a	5.8±0.5 ^{ab}	47.7±15.9 ^{ab}	8.9±4.4 ^{ab}
Sponge block	86.7 ^b	32.7±8.2 ^b	5.0±0.8 ^b	29.0±11.0 ^b	4.9±1.3 ^d
Unigrow 128	98.3 ^a	37.6±8.5 ^a	5.7±0.1 ^{ab}	53.5±24.7 ^{ab}	7.9±3.1 ^b
Sappi 49	99.2 ^a	43.9±3.5 ^a	6.4±0.4 ^{ab}	42.0±14.0 ^{ab}	7.1±2.3 ^{bc}
Poly 98 deep	96.7 ^a	34.3±7.9 ^b	5.4±1.1 ^b	36.6±19.9 ^b	6.5±2.9 ^c
Species × Container					
<i>E. dunnii</i> × Papier mâché	98.3 ^a	43.4±2.1 ^{ab}	6.1±0.2 ^{ab}	50.5±19.6 ^{ab}	8.9±4.2 ^a
<i>E. dunnii</i> × Hessian bag	95.0 ^a	40.5±1.3 ^{ab}	5.4±0.4 ^{ab}	31.5±7.7 ^{ab}	5.7±1.2 ^c
<i>E. dunnii</i> × Sponge block	90.0 ^{ab}	32.4±2.9 ^b	4.8±0.7 ^b	21.7±4.1 ^b	3.8±0.2 ^d
<i>E. dunnii</i> × Unigrow 128	98.3 ^a	40.5±11.6 ^{ab}	5.6±0.9 ^{ab}	52.4±13.0 ^a	8.6±2.0 ^{ab}
<i>E. dunnii</i> × Sappi 49	98.3 ^a	50.8±5.7 ^a	6.5±0.4 ^{ab}	43.1±11.9 ^{ab}	7.2±2.3 ^b
<i>E. dunnii</i> × Poly 98 deep	100 ^a	40.7±11.0 ^{ab}	5.6±1.0 ^{ab}	40.5±26.8 ^{ab}	7.5±3.3 ^b
<i>E. grandis</i> × Papier mâché	96.7 ^a	42.5±8.1 ^a	7.1±1.5 ^a	70.5±29.9 ^a	10.8±4.3 ^a
<i>E. grandis</i> × Hessian bag	98.3 ^a	35.9±4.9 ^{ab}	6.1±0.5 ^{ab}	63.8±24.0 ^{ab}	12.1±7.6 ^a
<i>E. grandis</i> × Sponge block	83.3 ^b	33.1±13.6 ^{ab}	5.2±0.9 ^b	36.3±17.8 ^b	6.1±2.3 ^{bc}
<i>E. grandis</i> × Unigrow 128	98.3 ^a	34.7±5.5 ^{ab}	5.8±1.0 ^{ab}	54.5±36.1 ^{ab}	7.3±4.2 ^b
<i>E. grandis</i> × Sappi 49	100 ^a	36.9±1.2 ^{ab}	6.2±0.3 ^{ab}	40.9±16.1 ^b	7.0±2.2 ^b
<i>E. grandis</i> × Poly 98 deep	93.3 ^a	27.9±4.8 ^b	5.2±1.1 ^b	32.7±13.0 ^b	5.4±2.5 ^c

Note: Means values (±SD) for species, container and species × container in each column with different letter(s) are significantly different ($p < 0.05$) according to Tukey's test.

whether wall-less plugs offer any advantage over the standard planting stock. One advantage of the plugs tested here is that they are manufactured by recycling waste paper and not by mining natural peatlands, which has a harmful environmental impact on wetland hydrology. Papier mâché plugs also offer an advantage over the composted pine bark-filled containers used extensively in South Africa. Despite the bark being an excellent rooting medium, its quality can be inferior owing to inadequate composting. South African commercial nurseries therefore are constantly looking for a new rooting medium and papier mâché plugs may be an option.

In conclusion, this study shows that there are possibilities for using wall-less plugs for the production of planting stock of forest trees. Between 120 and 130 million eucalypt seedlings are produced annually in various nurseries throughout South Africa (Zwolinski & Bayley, 2001). Papier mâché plugs evaluated in this study showed promising results for *E. dunnii* and *E. grandis*. However, long-term growth studies are required to confirm the benefits of implementing papier mâché plugs on a large scale. The manufacturing process of these plugs is relatively easy and can be improved. However, at this stage, the production cost and overall economics of large-scale implementation are not available and need to be assessed.

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