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The effects of Kelpak® *growth regulator on the growth responses' of three selected Fynbos species.*

by

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DECLARATION

I, Trevor Deon Adams, declare that the contents of this dissertation/thesis represent my own unaided work, and that the dissertation/thesis has not previously been submitted for academic examination towards any qualification. Furthermore, it represents my own opinions and not necessarily those of the Cape Peninsula University of Technology.

Signed **Date**

ABSTRACT

The effects of liquid Kelpak® and Kelpak® Plantit® disk growth regulator on the growth responses of three selected fynbos species were evaluated in this study. The experiment was arranged using a block design, consisting of 270 plant samples. The experiment consisted of three fynbos species, *Coleonema album*, *Erica verticillata* and *Leucospermum cordifolium*. Each species was subjected to three treatments, the control with no applications, liquid Kelpak® and Kelpak® Plantit® disks (hereafter referred to as disks). The control consisted of 10 plants samples, for each of the three species, arranged is numerical order 1-10. The liquid Kelpak® consisted of 40 plant samples. These 40 plants were divided into 4 groups, containing 10 plants and each group having a different treatment with group 1; 300 ml, group 2; 200 ml, group 3; 100 ml and group 4; 50 ml. The disks consisted of 40 plant samples. These 40 plants were divided into 4 groups, containing 10 plants each and each group having a different treatment with group 1, 2 disks, group 2, 1 disk, group 3, ½ disk and group 4 ¼ disk. The objectives of this study were to assess the desired application of Kelpak® by analysing the physiological improvements or growth responses on of *Coleonema album, Erica verticillata and Leucospermum cordifolium.*

Prior to planting, pre-trial measurements were recorded of each individual cutting. A standard ruler was used to measure the root length and shoot length of each plant, measurements were taken in millimetres. The weight of each plant was measured with a Radwag AS 220/C/2 analytical scale in grams. Plant growth, in terms of plant height was measured on a weekly basis. Plant height was measured with a standard ruler, from the surface of the medium to the tip of the tallest leaf. Watering during the trial period was conducted, using a hand held hose with a rosehead sprayer twice a week and during the third month of the trial once a week. Each container received an average of 250 ml of water. The final week of the trial final readings of the plants was conducted. Plants were carefully harvested and their roots were rinsed with tap water. A standard ruler was used to measure the root length and shoot length of each plant. The roots and shoots were then separated with a secateurs from each other. The fresh weight of each root and shoot weighed and recorded. The combined total weight in grams was captured. The roots and shoots were placed in a manila brown paper bag and placed in a laboratory oven at 55°C for 48 hours. The plants were then removed from the oven and the dry weight of each root and shoot weighed. The combined total was also recorded.

The results indicated that liquid Kelpak® and Kelpak® Plantit® disk had an effect on the growth of fynbos species at different application rates. Liquid Kelpak® and Kelpak® Plantit® disks had significantly increased the shoot, root growth and total weight of plants in *C. album* grown in 15 cm pots over an 18 week period. The liquid Kelpak® indicated higher growth

rates in the initial growing stages of C*. album* as the liquid was immediately available to the plant. The Kelpak® Plantit® disks had better influence on the growth over a longer period as the disk dissolved at a slower rate which eventually became available to the plant. The liquid Kelpak® and Kelpak® Plantit® disks had significantly increased the dry root weight and post-harvest root length of *E. verticillata* grown in 15 cm pots over a period. The Kelpak® Plantit® disks indicated higher growth rates in the dry root weight of *E. verticillata* but both the liquid and the disk had a positive effect on the post-harvest root length. The results also indicated that the successful rooting of *E. verticillata* was attributed to rooting hormone Seradix 2 under greenhouse heating environment. Liquid Kelpak® and Kelpak® Plantit® disks had significantly increased the wet and dry shoot weights, dry root weights and post-harvest wet and dry total plant weights of *L. cordifolium* grown in 15 cm pots over the growth period. The Kelpak® Plantit® disks indicated higher growth rates in the dry shoot weights of *L. cordifolium* but both the liquid and the disk had a significant effect on the wet shoot weight. The liquid and the disks were also responsible for the improved dry root weight. The liquid application indicated the best post-harvest wet weight but the disks improved the post-harvest dry weights. It can therefore be confirmed that organic seaweed concentrates such as Kelpak® is effective on the growth development of *L. cordifolium.*

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DEDICATION

This thesis is dedicated to my nieces,

Lauren Adams, Kayla Adams and Chloe´ Adams

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GLOSSARY

Terms/Acronyms/Abbreviations	Definition/Explanation
ANOVA	Analysis of variance
CFSF	Cape Flats Sand Fynbos
CFR	Cape Floristic Region
CPUT	Cape Peninsula University of Technology
EW	Extinct in the wild
IUCN	International Union for Conservation of Nature
KNBG	Kirstenbosch National Botanical Garden
SANBI	South African National Biodiversity Institute
SANParks	South African National Parks

Chapter 1 : LITERATURE REVIEW AND INTRODUCTION

The use of seaweed growth regulators in the cultivation of Fynbos species at Kirstenbosch National Botanical Garden nurseries.

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1.1 INTRODUCTION

The southern tip of the Africa is characterised by an exceptionally rich flora. This flora includes both the Cape Floristic Region (CFR) and the Succulent Karoo Region. The CFR is the richer of the two floral regions, which includes 9 000 species in an area of 90 000 km^2 (Dreyer, 2014). It has been described as one of six global floral kingdoms (Dreyer, 2014) and according to SANParks the world's richest natural garden (Barker & Yeld, 2004). The region has a Mediterranean climate and the indigenous vegetation, Fynbos, is short scrubby and sclerophyllous vegetation that is fire adapted and fire prone (Anderson *et al*., 2014). The word is derived from the Dutch word "fijnbosch" which describes the narrow-leaved type of shrubs characteristic of this vegetation (Dorrat-Haaksma & Linder, 2012). Conservation of the rich Cape flora is a great challenge, especially given the increased request for land and resources, and the effects of climate change (Hitchcock et al., 2013). A concern has been expressed over exploitation of Protea and other fynbos species, which are harvested from wild populations (Witkowski, 1990). The planting of fynbos is advantages in conserving and promoting indigenous flora. Kirstenbosch National Botanical Garden has been encouraging and promoting fynbos species and gardening with fynbos plants for many years. A study of the propagation of *Erica verticillata,* P.J. Bergius, by horticulturists at Kirstenbosch National Botanical Garden and the successful transplanting of them to local municipal and private reserves, Hitchcock (2006) has made a significant effort in saving endangered fynbos species. Information from such studies is important and likely to provide a better understanding how to save these species through propagation and cultivation.

1.2 Fynbos as a garden plant

Gardening in South Africa has shifted from the traditional exotic European garden to a more indigenous South African garden (Van Jaarsveld, 1996). Fynbos is not only famous for its diversity and unusual composition of its plants species but also for the sheer beauty of many of its wildflowers (Manning, 2007). While many species have also produced numerous ornamentals that are also prized in gardens throughout the world, species of Protea, Erica and Coleonema remain popular choices for a purely indigenous garden (Joffe, 2001). The planting of fynbos also attracts birds and other wild life to the garden (Privett, 2011). Gardening with fynbos plants generally require less care and remain the most appropriate vegetation to use for any water wise garden (Privett, 2011, Branch & Jennings, 2008). A few of the most attractive and ornamental species are almost impossible to grow in the average home garden, inevitably giving rise to disappointment (Brown & Duncan, 2006). Many ardent fynbos gardeners have been disappointed when plants bought from garden centres die within a few months of planting. Most species have quite specific preferences as to its adaptability to climatic and soil conditions (Privett, 2011).

Fynbos occurs on low-nutrient soils similar to other 'Mediterranean' climate zones with diverse floras (Sinclair, 2012). The soil for most species should be acid (pH 4.4 – 7.0), sandy and derived from weathered quartzitic sandstone (Joffe, 2001). In nature soils are mainly deprived of nutrients especially nitrogen and phosphorus (Brown & Duncan, 2006). There are several adaptations which fynbos display to enhance the uptake of nutrients from the soil. Protea species for example have developed an unusual form of root growth to adapt to nutrient poor soils. Tufts of fine roots sprout from the surface roots of Protea plants after the first winter rains and absorb moisture and minerals from the soil (Manning, 2007). Research on many fynbos species have been conducted for example medicinal research on *Coleonema album* has been conducted into the biological activities of antimicrobial effects on the species (Eldeen & Van Staden, 2008). Research has also been conducted on Proteaceae species where the effects of fertilizer applications were tested on *Leucospermum parile* plants in nutrient-poor sandplain lowland fynbos ecosystem (Witkowski, 1990). Further research is required on fynbos plants for selection, application and concentration of growth regulator for cultivation purposes as it remains a challenge for fynbos growers.

1.3 Organic Concentrates and their role

The prolonged use of inorganic fertilisers' disturbs the physical properties of soil and affects organic matter content (Papenfus *et al*., 2013). Supplementing the soil with organic substances is an option to minimise the use of chemical fertilisers. The benefits of seaweeds as sources of organic matter and fertilizer nutrients have led to their use as soil conditioners for centuries (Wajahatullah *et al*., 2009). The use of organic substances is aimed at increasing root and overall plant growth which will increase the uptake of available nutrients in the soil. Kelpak® research has been conducted on canola, cereals, cucurbits, flowers, leafy vegetables, potatoes, rice, table grapes and tomatoes (Kelpak®, 2013). The beneficial effects seaweed concentrates have on the growth and yield of plants have been well documented (Crouch, 1990). Seaweed concentrates show good potential as supplementary soil conditioners (Papenfus *et el*., 2013).

Plants grown in soils treated with seaweed composts or concentrates applied either to the soil or foliage, exhibit a wide range of responses such as improved germination, root development, leaf quality, general plant vigor and resistance to diseases (Craigie, 2011). Besides eliciting a growth-promoting effect on plants, seaweeds also affect the physical, chemical, and biological properties of soil which in turn influence plant growth (Wajahatullah *et al*., 2009). Seaweeds and seaweed extracts enhance soil health by improving moisture-holding capacity and by promoting the growth of beneficial soil microbes. Seaweeds and seaweed products enhance plant chlorophyll content (Wajahatullah *et al*., 2009).

An extract from seaweed *Ascophyllum nodosum* applied to the soil or on foliage of tomatoes produced leaves with higher chlorophyll content than those of untreated controls (Wajahatullah *et al*., 2009). These processed seaweed products are marketed as biostimulants or growth regulators, sold as powders and liquid extracts and their use is well established for a variety of agricultural and horticultural crops (Stirk & van Staden, 2010). What is not known is the extent of applications of organic concentrates and their influence on fynbos garden plants.

1.4 Kelp as a product

The most common of the kelp species is *Ecklonia maxima* which is a large plant with a cylindrical stipe reaching up to 12 m and forming a flat palm-like hand bearing strap-shaped fronds. It has a hollow gas filled stipe with a buoyant swollen bulb at the top which holds the plant upright, allowing the fronds to float close to the surface of the water (Branch & Branch, 1981).

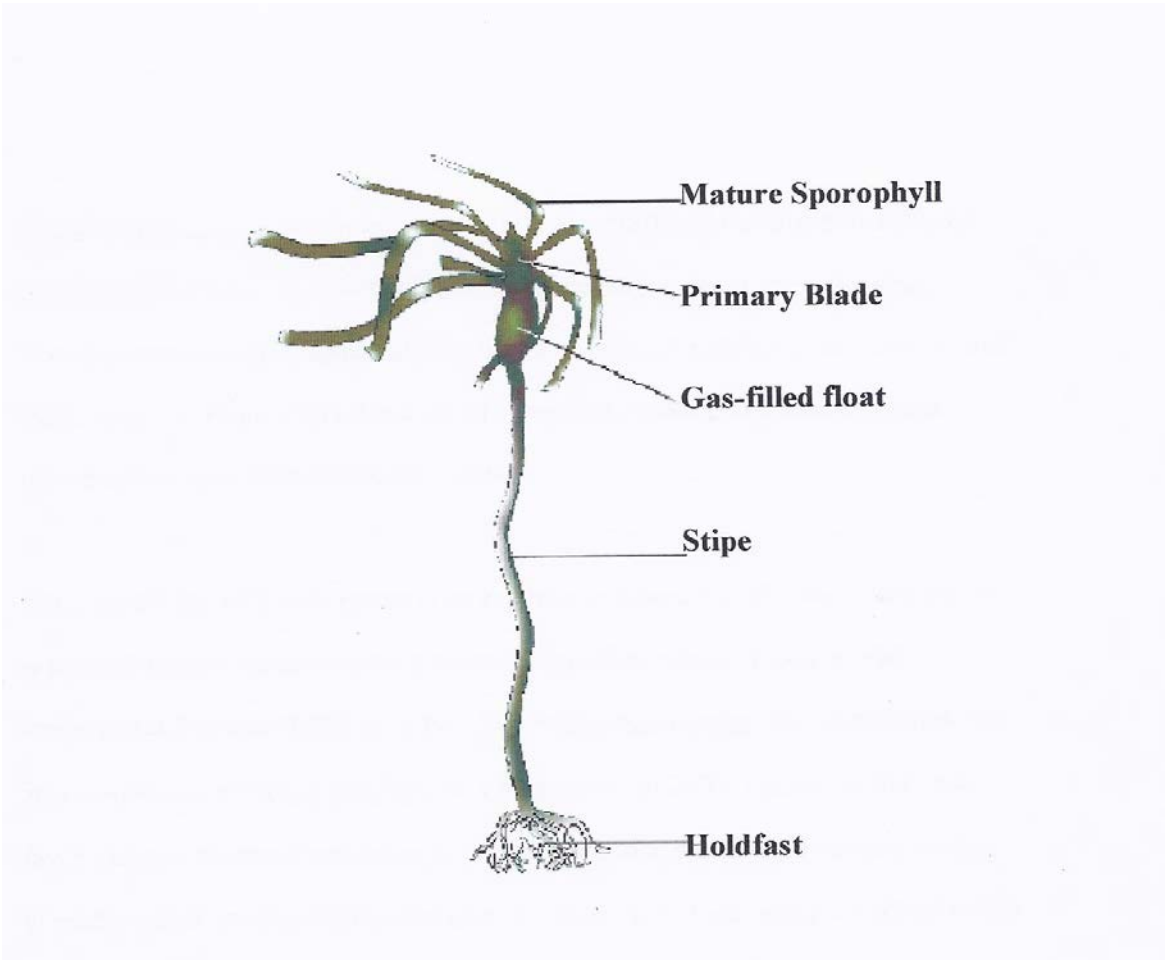

Figure 1.1: *Ecklonia maxima* mature sporophyte (Arendse, D. 2003)

It is one of four species found in the ocean waters of South Africa (Rothman, 2006). *Ecklonia maxima* is locally common and occurs from the coastal town Aasfontein, 15 km west of Cape Agulhas, westwards to as far as Swakopmund in Namibia (Arendse, 2003). This distribution is not continuous however, for the species is absent or is represented by occasional individuals along the northern and the greater part of the western shores of False Bay. The reason for scarcity in this region is owing to sea temperatures as normal growth temperature conditions must not exceed 14.6 degrees Celsius. The variability of sea temperature affects the abundance of *Ecklonia* maxima especially in the False Bay area (Featonby-Smith, 1984).

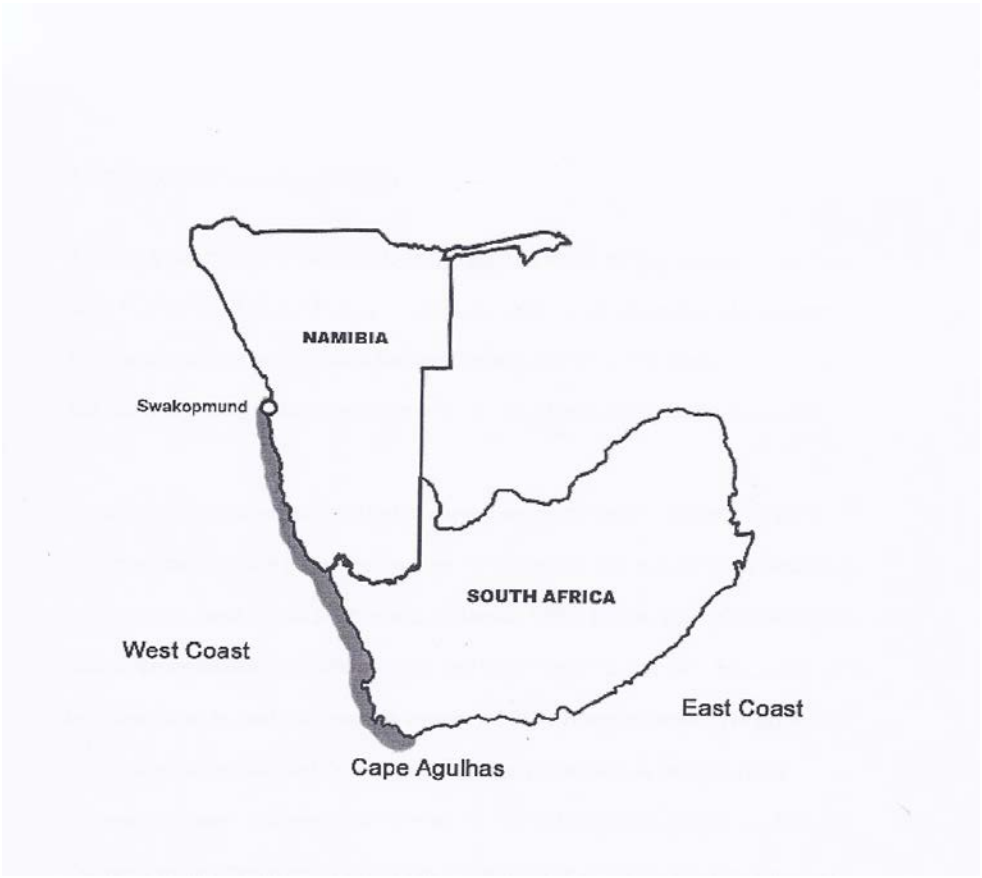

Figure 1.2: *Ecklonia maxima* distribution (Arendse, D. 2003)

This is of great importance in the ecology of the system as it means *Ecklonia maxima* always forms the top of the canopy of the kelp bed, shading other species. It also means that plants float when torn free and can thus be washed ashore where they of importance to the ecology of sandy beaches (Branch & Branch 1981).

Ecklonia has been harvested for many years in South Africa. The commercial exploitation of seaweed resources in South Africa is regulated by the Marine Living Resources Act of 1998 (Act No. 18 of 1998) Marine and Coastal Management are responsible to administer the act (Rothman, 2006). Prior to about 1980's, kelp was collected mainly as beach cast then dried and exported for extraction of alginate (Rothman, 2006). Alginate is used in a wide range of applications particularly in the food, industrial and pharmaceutical industry (Arendse, 2003). An average of 134 tonnes of wet mass of *Ecklonia maxima* was processed per annum for the production of liquid plant growth stimulant between 1984-1991 (Arendse, 2003). Trials conducted on wheat indicated that the local kelp product, applied at only 2 litres per hectare with an ordinary fertiliser blend increased production from 20 percent to 300 percent. It also increased the production of beans by 300 percent and improved germination and production of 12 other crops (Branch & Branch 1981). Studying these results it is important to establish how kelp as a growth regulator may influence the growth in fynbos species for nursery and garden cultivation purposes.

1.5 Cytokinins and Auxins

Kelp contains a natural and unique source of auxins and cytokinins, a group of plant growth regulators that have been proven to have a number of beneficial physiological effects on plants (Stirk & van Staden, 2010). Kelpak® possesses a very high auxin to cytokinins ratio, auxin 350 to cytokinins 1. Auxin dominance stimulates vigorous primary and secondary root development in the plant, thus enhancing plant hormone production resulting in an improved crop. Plant hormones acting as regulators of multiple physiological processes, cytokinins are especially important in regulating cell division, expansion and delaying senescence (Ghanem *et al*., 2011). Cytokinins are a group of plant growth regulators that elicit a wide range of physiological responses in plants (Stirk *et al*., 2004) at cellular level, being crucial for cell division as well at the tissue organ and whole plant level. Cytokinins influence development processes such as shoot apical dominance, branching, chlorophyll productions and root growth which have some of the highest concentrations of cytokinins (Stirk & van Staden, 2010). Research has shown that cytokinins could increase salt tolerance in wheat plants by interacting with other plant hormones, especially auxins and abcisic acid. Kinetin a biologically active compound and type of cytokinins was effective in increasing germination rates. Kinetin priming showed a consistent promoting effect in the field and improved growth and grain yield under salt stress (Mazid, *et al*., 2011).

Auxin, a natural plant hormone is manufactured in plant shoot tips and translocate downwards to the root zone. One of its functions is to signal plants to increase its root system (Kelpak®, 2013). Auxins sense plant cells and rapidly converted into a wide variety of growth responses and development. These include changes in growth direction, changes in shoot and root branching and changes in vascular differentiation (Leyser, 2001). A plant's roots system determines both the capacity of an organism to acquire nutrients and water, as well as providing a means to monitor the soil for a range of environmental conditions (Overvoorde, 2010). A study conducted on tomato fruit revealed that regular cell division first takes place in 10 – 14 days of development. However when fruit are induced by the natural auxin indole-3-acetic acid (IAA), cell division only lasts for 10 days (de Jong, 2009). The roots of fynbos are especially sensitive in its natural environment to disturbance where an organic layer of decomposed material provides moisture and absorption of nutrients by a network of adventitious roots (Manning, 2007). It is therefore of great importance to explore how auxin and cytokinins associated with organic growth regulators will interact under the production of fynbos species.

1.6 Kelpak® as a growth regulator

Since the 1960's *Ecklonia maxima* has been harvested for the production of seaweed concentrates. Tonnes of kelp are harvested fresh from the South African coast every year for the production of the seaweed concentrate Kelpak® (Stirk & van Staden, 2010).Kelpak® produced from the seaweed species *Ecklonia maxima* (Osbeck) Papenfuss is an organic approved natural biostimulator. Plants grown in soils treated with seaweed composts or concentrates applied either to the foliage or soil; display a wide range of responses such as general plant vigor, root development, improved germination, leaf quality, and resistance to diseases (Craigie, 2011). Seaweeds also affect the physical, chemical, and biological properties of soil which in turn influence plant growth (Wajahatullah *et al*., 2009).

Seaweed concentrate Kelpak® is used extensively in the horticulture, agriculture and the mariculture industry. Kelpak® was applied as a foliar spray and root drench to Eucalyptus tree seedlings grown in greenhouse conditions at various time intervals, application rates and dilution rates which significantly increased overall seedling growth (Rijkenberg, 1994). Canola, (oilseed rape) treated with Kelpak® indicated improved root development and an increase in seed and oil yield. Applications of Kelpak® in the Northern hemisphere improved winter hardiness in plants (Kelpak®, 2013). Kelpak® applied to ornamental plants improved root nutrient and moisture development. It also indicated an increase in foliar and longer flower stems and higher flower production (Kelpak®, 2013). In vegetable crops the application of Kelpak® increased the vigour of root and shoot growth. It further improved size and higher yields (Kelpak®, 2013). Clonal *in vitro* grown tomato seedlings were treated with seaweed concentrate from *Ecklonia maxima* with various dilution rates. The seaweed concentrate had significant increase in root fresh weight and root length had increased (Crouch, 1990).

A study was conducted to investigate if seaweeds that have no root systems, grown in culture, would benefit from the addition of Kelpak® or a combination of Kelpak® and fertilizer. A laboratory experiment was conducted using red alga *Gracilaria gracilis* in culture dishes containing a medium supplemented with seawater enriched with provasoli to which various concentrations of Kelpak® were added. *Gracilaria gracilis* in some of the Kelpak® treatments grew significantly better than the control (Robertson-Andersson *et al*., 2006). Trials were also conducted at Jacobsbaai Sea Products Ltd. Along the west coast South Africa. *Ulva lactuca,* green alga was grown in effluent from fish culture, with various concentrations of Kelpak® added once a week. The intermediate concentration produced the highest growth of *Ulva lactuca* in the effluent water, while the highest concentration inhibited the growth of *Ulva lactuca* (Robertson-Andersson *et al*., 2006). The results of this study suggested that Kelpak®

could be useful for commercial seaweed mariculture. In this context the measurement of seaweeds on ornamental plants could improve results in plant growth and serve as an indicator for the use of organic growth regulators in crop production.

1.7 Propagation and cultivation using Kelpak® at Kirstenbosch National Botanical Garden Nurseries

The economic, function and conservation use of fynbos has led scientists, conservationists and volunteers to collect a wealth of information on the ecology, geographical distribution and evolutionary history of Proteas (Schurr *et al*., 2012). The foundation for this knowledge was laid by the intense research on Proteaceae conducted in the 1980's (Schurr *et al*., 2012). Wild fires are a natural occurrence in fynbos and fire stimulated seed germination has been reported for a number of fynbos species (Brown, 1993). A study was conducted with 28 fynbos species for seed germination response to smoke and smoke extracts treatments. In nature plants can reproduce sexually from seeds and asexually or vegetatively (Toogood, 1999). Asexual propagation is very important to preserve the genetic integrity of species especially where particular characteristics of a plant need to be maintained which cannot be maintained through sexual reproduction (Bowes, 1999). Cutting production is more costly compared to seed, however their survival rates are higher than seedlings (Thompson, 2005).

Fynbos species at Kirstenbosch National Botanical Garden nurseries are propagated under greenhouse conditions and grown under greenhouse or shade net conditions (Viljoen, 2013 pers. comm 2013.). Rooted cuttings from the propagation bench are hardened off for three to four weeks prior to potting and during this phase the roots are drenched with Kelpak® as a method to cope against plant stress (Viljoen, pers. comm. 2013). Once the Fynbos species are potted into Fynbos mix soil in 0.75l bags or 1 pint bags respectively they are given again a soil drench of Kelpak®. However their growth was not scientifically measured or quantified and only overall visual growth and overall health appearance is noted (Mcquillan pers. comm. 2013). Horticulturists at Kirstenbosch were introduced to a planting disk in 2012 by Kelpak® which was a compressed solid disk of Kelp. The Kirstenbosch Horticulturists experimented with the planting disks and applied it to various Fynbos species while being re-potted into bigger bags sizes such as a two pint bag. The growth responses the disk had on the plant were not measured or quantified scientifically (Mcquillan, pers. comm. 2013). The growth responses that the Kelpak® planting disk has on indigenous fynbos species have received little attention and there have been few studies testing the effectiveness of seaweed growth regulator concentrations on the cultivation of containerized fynbos plants. The need to investigate the effects of liquid Kelpak® and Kelpak® Plantit® disk growth regulators on the

growth responses of vegetative cultivation is important for the successful growing of fynbos species in propagation nurseries such as Kirstenbosch National Botanical Garden Nurseries.

1.8 CONCLUSION

This review established a rationale for the study with the importance to conserve and grow fynbos as a garden plant. The study undertook to identify new knowledge that can unlock the problems of growing fynbos thereby increasing the conservation of fynbos species and the use of it for gardening. Three fynbos species, *Coleonema album, Erica verticillata* and *Leucospermum cordifolium* and were selected for the study. Kelpak a growth regulator was developed from the seaweed *Ecklonia maxima* and used as source of seaweed concentrate in the agricultural sector. Kelpak® is known to increase the yield of agricultural crops but there was no scientific evidence to prove what effect it had on fynbos species. The testing of seaweed growth regulators in the cultivation of species will be of great significance for future production at nurseries.

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Chapter 2 : PROBLEM STATEMENT, HYPOTHESIS AND OBJECTIVES

2.1 PROBLEM STATEMENT

The Cape Floristic Region at the southern tip of Africa is home to one of the richest floras in the world, known as Fynbos. This unique vegetation type contributes to 80% of the species in this region (Privett & Lutzeyer, 2010). Fynbos occurs in acidic, (pH 4.4 – 7.0) soils that are deprived of nutrients especially nitrogen and phosphorus. Not all Fynbos species are easy to cultivate (Brown & Duncan, 2006). A few of the most attractive and ornamental species are almost impossible to grow in the average home garden, inevitably giving rise to disappointment (Brown & Duncan, 2006). The emphasis of gardening in South Africa is shifting from the traditional exotic European garden to a more indigenous South African garden (Van Jaarsveld, 1996). Fynbos is the ultimate vegetation type for any water wise gardener (City of Cape Town, 2008). An appropriate method of improving growth responses on fynbos species is required to encourage large scale production nurseries to maximise growth of these species. This will encourage gardening enthusiasts to garden with more native plants.

Kelpak® produced from the seaweed species *Ecklonia maxima* is an organic approved natural biostimulator. It contains a unique source of plant growth regulators Auxins and Cytokinins that have been proven to have a number of beneficial physiological effects on plants (Papenfus *et al*., 2013 and Stirk & van Staden, 2010). Research around the world has proven Kelpak's ability to increase the health, quality and yield in a wide variety of crops for farmers (Kelpak, 2013). Kelpak research has been conducted on canola, cereals, cucurbits, flowers, leafy vegetables, potatoes, rice, table grapes and tomatoes. This study aims at evaluating the effects of Kelpak® growth regulator on the growth responses of three fynbos species, *Coleonema album* (Rutaceae), *Erica verticillata* (Ericaceae) and *Leucospermum cordifolium* (Proteaceae).

2.2 HYPOTHESIS

It is hypothesized that liquid concentrations of Kelpak® might be absorbed first to show earlier growth, but the disk applications might show extended growth results.
It is hypothesized that a higher dosage application of Kelpak® will produce a higher yield compare with the applications of lower dosages on the same species.
Various rates can have a positive effect on the yield (the fresh weight) and root length of *Coleonema album, Erica verticillata and Leucospermum cordifolium*.

2.3 OBJECTIVES OF THE RESEARCH

2.3.1 Main Objectives

To identify the optimal applications of Kelpak® to produce an increase in growth of Fynbos species.

2.3.2 Specific objectives

- To determine the desired application of Kelpak® by analysing the physiological improvements/ growth response of the total shoot growth of *Coleonema album Erica verticillata* and *Leucospermum cordifolium.*
- To determine the desired application of Kelpak® by analysing the physiological improvements/ growth response of the total root growth of *Coleonema album, Erica verticillata* and *Leucospermum cordifolium.*
- To determine the desired application of Kelpak® by analysing the physiological improvements/ growth response of the chlorophyll content of *Coleonema album, Erica verticillata* and *Leucospermum cordifolium.*

Chapter 3：Coleonema album response to growth regulator Kelpak in nursery container production

Growth responses of *Coleonema album* to organic seaweed growth regulator in nursery container production.

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3.1 ABSTRACT

The white confetti bush (*Coleonema album*) is a popular indigenous medicinal and garden plant in South Africa. Due to its low soil acidity growing requirements not much research has been done on using growth regulators successfully during cultivation. Growth regulator, Kelpak® is used by many growers but has not been scientifically tested to determine its success on fynbos container production. This study assessed the plant root and shoot length, fresh and dry root and shoot weight. Ninety plants were divided into three different treatments in a randomised block design. A control with 10 plants received no treatment. A second treatment consisting of 40 plants received liquid Kelpak® and third treatment consisting of 40 plants received Kelpak® Plantit® disks. The aim of the study was to establish the effects of the different Kelpak treatments on the plants growth. The results indicated that root and shoot length, fresh and dry weight of plants grown in the Kelpak® Plantit® disks treatments were generally higher.

Keywords: Kelpak®, KNBG, potted plants, plant height, South African National Biodiversity Institute (SANBI),

3.2 INTRODUCTION

The term fynbos belongs to a unique vegetation type that is dominated by shrub species endemic to southern and South Western Cape of South Africa (Cowling & Richardson, 1995). This vegetation type contributes to 80% of the species found in this region (Privett & Lutzeyer, 2010), while the Cape region has arguably the richest composition of native fragrant plant species in South Africa (Moolla & Viljoen, 2008). It is within this epicentre of essential oil bearing plants, that the genus Coleonema is found and particularly abundant in the mountainous areas of the Cape (Moolla & Viljoen, 2008). *Coleonema album* is an indigenous medicinal fynbos species which occurs widely from Saldanha Bay to the Cape Peninsula and as far as Bredasdorp, in the Overberg region of South Africa. It is also commonly known as the Confetti Bush and belongs to the Rutaceae family, the ninth largest family in the Cape Floral Region (Trinder-Smith, 2006; Brown & Duncan, 2006). This species is a compactly branched spreading woody shrub that grows up to one meter. It has small narrow kneedle-like leaves that are 12 mm long and 1,5 mm wide (van Wyk *et al.* 2009). The

small white flowers are 6 to 7 mm in diameter. It is a coastal plant that is able to tolerate strong coastal winds at sea levels but also up to a maximum altitude of 750 m on Table Mountain (Trinder-Smith, 2006). Coleonema is one of the buchu's, a term used by the Khoi and San people for any fragrant plant that could be dried and powdered (Schwegler 2003). Buchus are natural deodorisers and the aromatic leaves are used by fishermen to remove the fishy smell by rubbing the twigs of *C. album* between their hands (Schwegler 2003) while the leaves can also be used as insect repellent or pot pourri purposes (SANBI, 2003, Bean & Johns, 2005). The aromatic oil of the species is exported internationally for perfumery and medicinal purposes (Fajinmi *et al.,* 2013).

C. album is a popular garden, hedge, container or topiary plant for the horticultural and landscape industries (SANBI, 2003). The demand for gardening plants in South Africa has shifted to more indigenous species such as *C. album* as they require less care after establishment for a water wise garden (Van Jaarsveld, 1996; Joffe, 2001; Branch and Jennings, 2008; Eldeen & Van Staden, 2008; Privet, 2011). Some fynbos species are not easily grown, especially some of the most attractive species (Brown & Duncan, 2006) as they are often found on low-nutrient soils under Mediterranean climate areas (Sinclair, 2012). These soils are fairly acidic, range from a pH of 4.4 to 7.0 (Gold, 1992) and are deprived of nutrients especially nitrogen and phosphorus. Fynbos species such as Coleonema has made several adaptations to enhance the uptake of nutrients from these soils. Some species of the genus Protea have developed an unusual form of root growth to adapt to nutrient poor soils where tufts of fine roots sprout from the surface roots after the first winter rains to absorb moisture and minerals from the soil (Manning, 2007). As the organic content of these soils are very low the amount of nutrients available or species require is often not known. The application of nutrients is important, to increase production and quality, however it remains uncertain what concentration of growth regulators is required to provide optimum conditions for nursery container plants. The aim of the study therefore was to investigate the effects of various concentrations organic growth regulator, Kelpak® and Kelpak® Plantit® disks on the growth responses of *C. album* (Thunb.) Bartl. & J.C. Wendl. (Rutaceae).

3.3 MATERIALS AND METHODS

3.3.1 The Experiment layout and treatments

Rooted cuttings of *C. album* were sourced from Veld and Fynbos wholesale nursery and were planted into 15 cm square plastic pots which contained a well-drained nutrient poor fynbos soil mix developed by (KNBG). The fynbos soil mix was made up of 8 parts, 6 mm - 12 mm fine milled pine bark, 1 part milfeed wet Consol® glass sand, 2 parts "Malmesbury Grof" sand and 1 part agricultural lime. The trials were conducted in the Disa House of the

Living Collections Nursery of Kirstenbosch National Botanical Gardens (KNBG), Newlands, Cape Town, Longitude 18.43209E and Latitude 33.99039S. This greenhouse covered with opaque polycarbonate sheeting which allow for a constant, controlled environment. The average temperature in the greenhouse ranged between 13 and 23 degrees Celsius. The greenhouse contained growing benches constructed of brick and 13 mm drainage chip on the surface on which the plants were placed.

The pots were labelled according to the treatments applied. The experiment was arranged using a block design, consisting of 90 plant samples. The experiment layout consisted of the control, liquid Kelpak® and Kelpak® Plantit® disks. The seaweed products are manufactured by Kelp Products (Pty) Ltd., Simonstown, City of Cape Town, South Africa. The control consisted of 10 plants which were arranged is numerical order 1-10. The liquid Kelpak® consisted of 40 plants divided into 4 groups, containing 10 plants and each group having a different treatment with group 1, 300 ml, group 2, 200 ml, group 3, 100 ml and group 4, 50 ml. consisted of 40 plants which were divided into 4 groups, containing 10 plants each and each group having a different treatment with group 1, 2 disks, group 2, 1 disk, group 3, ½ disk and group 4 ¼ disk.

The disks were cut into the various size classes according to the various treatments each plant would receive. The treatments that received the half and quarter size disks were cut using a guillotine. The pots were filled half way with soil and the disks were inserted on top and covered lightly. The rooted cuttings were then inserted and the rest of the soil added to fill the pot. The treatments that received the Kelpak® liquid dosage was administered using a measuring cylinder assuring accuracy. The control received no treatments and the plants that contained the control only received water.

3.3.2 Data collection

Prior to planting, pre-trial measurements were recorded of each individual cutting. A standard ruler was used to measure the root length and shoot length of each plant, measurements were taken in mm. The weight of each plant was measured with a Radwag AS 220/C/2 analytical scale in grams. Plant growth, in terms of plant height was measured on a weekly basis. Plant height was measured with a standard ruler, from the surface of the medium to the tip of the tallest leaf. Watering during the trial period was conducted, using a hand held hose with a rose head sprayer twice a week and during the 10^{th} week of the trial of the trial once a week. Each container received an average of 250 ml of water. The 18^{th} week of the trial final readings of the plants was conducted.

The plants were then moved to CPUT, Department of Horticulture research laboratory, Bellville campus. Plants were carefully harvested and their roots were rinsed with tap water. A standard ruler was used to measure the root length and shoot length of each plant. The

roots and shoots were then separated with secateurs from each other. The fresh weight of each root and shoot were weighed and recorded. The combined total weight in grams was captured. The roots and shoots were placed in a manila brown paper bag and placed in a laboratory oven at 55° C for 48 hours. The plants were then removed from the oven and the dry weight of each root and shoot weighed. The combined total was also recorded.

3.3.3 Statistical analysis of plants

Collected data was analysed using the One–way analysis of variance (ANOVA), with the computations being done using the software program STATISTICA [Software Program version 2010 (Statsoft Inc., Tulsa, OK, USA)]. The Fisher least significance (L.S.D.) was used to compare significant treatment means *at P*≤0.05 level of significance (Steel & Torrie, 1980).

3.4 RESULTS

The results from the Kelpak® growth regulator treatments were significant (P≤0.001) at week 2 on the shoot length of *C. album.* The liquid treatments from 50 – 300 ml were the most significant compared to the control with no concentration. The disk at this stage proved less significant compared to the control; however the 1 disk application in treatment 7 did show significance above the other disk treatments (Table 3.1). Week 4, 6 and 18 measured significantly similar (P≤ 0.01) compared with the control. Significance was observed during week 4 in the 200 ml and 50 ml treatments compared to the control. Shoot length significantly (P≤ 0.01) increased at week 4 in the liquid treatments of 50 – 300 ml Kelpak® (Table 3.1). A similar significance (P≤ 0.01) continued in week 6 which also saw liquid treatments from 50 ml – 300 ml Kelpak® exceed the control. Week 8,10,12,14 and 16 measured significantly (P≤ 0.05) similar compared with the control. Week 8 measured significant (P≤ 0.05) increase in shoot length in the 50 ml treatments compared to the other treatments. A similar significance (P≤ 0.05) continued in week 10 but changed to the 200 ml treatment 3 at week 14 where the liquid application far exceeded the disk treatments. Week 18 changed significantly (P≤ 0.01) with treatment 6, 7 and 9 more favourable. The results shows that the disk had an effect on shoot length over a longer growing period (Table 3.1).

In Table 3.2 the effect of different concentrations of Kelpak® on total wet and dry weights showed significance (P≤ 0.01) on shoots of *C. album*. The wet weight significantly (P≤ 0.01) increased from 200 – 300 ml concentrations of liquid Kelpak® compared to the control and the disk applications. The wet weights of treatments 5, 9 and treatments 7 and 8 were similar in results but significantly (P≤0.01) different to the control. The 100 ml application in treatment 4 showed significance above the latter treatments also indicating increase in wet shoot weight; however the ¼ disk application in treatment 9 indicated significance above the

other disk treatments (Table 3.2). The dry weight showed significance (P≤ 0.01) on shoots of *C. album* in treatment 3 and 4 with 100 -200 ml concentrations. The 300 ml application in treatment 2 indicated significance above the disk applications and the control. The results showed that the liquid Kelpak® had an effect on shoot weight of C. album over the growing period (Table 3.2).

The effect of different concentrations was most significant (P≤ 0.001) in treatment 4, 100 ml liquid Kelpak® on the dry weight of the plant roots. This significance was followed by all other treatments compared to the control (Table 3.3). The wet weight on the roots of *C. album* showed significance (P≤ 0.01) in all the treatments, both liquid and disk Kelpak® concentrations compared to the control (Table 3.3). The results show that both Kelpak® growth regulators had an effect on root weight of C. album over the growing period (Table 3.3).

Table 3.4 results shows that both the pre harvest and both post-harvest results were significantly different (P≤ 0.01) to the control. The total pre-harvest wet weight was significantly different (P≤ 0.01) in all the treatments compared to the control. These results were repeated exactly in the total post-harvest wet weight. The total post-harvest dry weight of the plants show the same significance (P≤ 0.01) compared to the control (Table 3.4). The 100ml application in treatment 4 indicated significance above the other treatments with treatments 2, 3 and treatments 5, 6, 7 and 9 similar in results. The ½ disk treatment 8 also indicated significant (P≤ 0.01) increase in plant weight compared to the control. The results show that the liquid Kelpak® had an effect on the total weight of *C. album* over the growing period (Table 3.4).

3.5 DISCUSSION

Kelpak® seaweed concentration is a natural and unique source of auxins and cytokinins, a group of plant growth bioregulators that have been proven to have a number of beneficial physiological effects on plants (Stirk *et al.,* 2004). Seed germination in certain species can prove to be difficult, while seed dormancy, low seed viability, lack of germination cues and immature seed are the challenges faced during seed propagation. All the above mentioned challenges are common to the Rutaceae family to which *C. album* belongs (*Fajinmi et al.,* 2013). In this study the rationale was to investigate which Kelpak® concentration would increase root and shoot growth over an 18 week period of *C. album* plants propagated vegetatively.

The liquid seaweed growth regulator (50 ml – 200 ml) had a significant improvement over a ten week growth period however half a disk were far more significant than the liquid at 18 weeks on the vegetative growth in shoot length. The changed developed from weeks 16 to

18 significantly (P≤ 0.01) with treatment 6, 7 and 9 more favourable. The results indicate that the liquid Kelpak® applications were absorbed by the plants much faster than the disks and indicated much better growth at the initial stages compared to the disk that had an effect on shoot length over a longer growing period. This indicated that the liquid concentrate is available to the plant immediately for uptake through the shoot and roots compared to the disk which had to be broken down or dissolved in the soil. According to Crouch *et al.,* (1990) plant hormones present in seaweed concentrates are largely responsible for improved growth and vigour in plants. The end of the trial also indicated aesthetically that the shoot growth had been healthier and more vigorous. Plant hormone cytokinins influence development processes such as shoot apical dominance, branching, chlorophyll productions and root growth (Stirk & van Staden, 2010). The liquid growth regulator had a far better effect over the short term, while the disk proved to have impact at the end of the trial.

The liquid Kelpak® growth regulator at 200 – 300 ml showed far more improvement on the wet and dry weights of the shoots of *C. album* over the growing period. This supports the fact that liquid concentration is more effective in the initial stages of growth as plants react to the liquid sooner to influence long term results. A study conducted using lettuce plants found that Kelpak® had little effect on the growth of the plants using half or single strength solutions but increased the yield of plants receiving double the strength solution (Crouch *et al*, 1990). It is possible for this reason that liquid concentrations are easier absorbed. It should also be noted that many fynbos species, such as *C. album* contains volatile oils which could inhibit or promote uptake of nutrients either through leaves or movement through stems.

The results showed growth in root development and growth measured in the wet and dry weight of the samples. At KNBG once fynbos species are potted into a fynbos soil mix, plants are given a soil drench of Kelpak®. However plant growth has not been scientifically measured or quantified and results rely only on overall visual growth and overall health appearance (Mcquillan pers. comm. 2013). Plants can either be over or under fertilized. Kelpak® in high concentrations on cuttings exposed for too long, have produced lower and poor rooting results, suggesting that Kelpak® was too toxic under these experiments (Jones & van Staden, 1997). Both the liquid and disks had a significant greater effect on root development of *C. album.* However Jones and van Staden (1997) obtained favourable rooting results through favourable rooting conditions using various parameters.

Research studies have also indicated that Kelpak enhanced growth under greenhouse conditions of cucumber and tepary beans (Papenfuss *et al.,* 2013). Although *C. album* was grown in greenhouse, the environmental conditions were not trailed in this study. The research did conclude that both Kelpak® liquid and disks had a significant growth impact on the total wet weight of *C. album* whereas the liquid Kelpak® had an effect on the total dry

weight of *C. album* over the growing period in the greenhouse. According to Crouch (1990), organic constituents improve soil structure by improving soil aeration and water retention. Alginates, fucoidin and similar elements in seaweed enhance soil structures making conditions favourable for root growth. The research conducted by Crouch (1990) could not substantiate if soil conditions was a factor in the enhancement of root growth. Although the total weight of the plant increased it could not be determined if other factors were working in conjunction with Kelpak® to improve overall plant weight. The fact that most fynbos species such as *C. album* are also classified as more woody plants could further be contributing or inhibitory factors for absorption of growth regulators through either roots or leaves.

3.6 CONCLUSION

In conclusion the use of liquid Kelpak® and Kelpak® Plantit® disks had significantly increased the shoot, root growth and total weight of plants in *C. album* grown in 15 cm pots over an 18 week period. The liquid Kelpak® indicated higher growth rates in the initial growing stages of C*. album* as the liquid was immediately available to the plant. The Kelpak® Plantit® disks had better influence on the growth over a longer period as the disk dissolved or broke down at a slower rate which eventually became available to the plant. It is recommended that future studies continue with research using the *C. album* to determine if Kelpak growth regulators have an influence on the nutrients of fynbos soils. It is also recommended that further research be conducted on using *C. album* to determine if Kelpak® growth regulators are affected by changes in the greenhouse growing environment and or the composition of the plant cells.

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Table 3.1: Effect of different concentrations of liquid Kelpak® and Kelpak® Plantit® disks on shoot length of *Coleonema album.*

Treatment	Week 2	Week 4	Week 6	Week 8	Week 10	Week 12	Week 14	Week 16	Week 18
1 = Control	**76.50± 5.64bc**	**86.16± 7.19b**	**100.00± 10.11b**	**113.00± 11.66d**	**122.83± 11.03b**	**132.66± 11.55c**	**135.83± 12.48c**	**140.50± 13.99bc**	**146.50± 14.05ab**
2 = 300ml	**56.00± 16.13a**	**61.00± 17.20ac**	**76.33± 19.73a**	**88.16± 20.58abc**	**98.33± 24.72abc**	**106.66± 27.49abc**	**110.50± 30.19abc**	**116.83± 33.95abcd**	**123.33± 36.94ac**
3 = 200ml	**51.16± 17.19a**	**57.83± 18.15a**	**73.66± 19.49a**	**84.66± 18.15ab**	**88.66± 18.61a**	**91.83± 18.01a**	**92.83± 19.20a**	**95.33± 18.21d**	**99.16± 20.67c**
4 = 100ml	**52.16± 10.59a**	**60.16± 12.20ac**	**73.33± 13.90a**	**84.66± 14.37ab**	**90.83± 13.52a**	**96.33± 15.21a**	**100.16± 17.90ab**	**106.6667± 22.55364ad**	**114.66± 28.68ac**
5 = 50ml	**44.83± 9.88a**	**54.66± 8.04a**	**67.66± 19.21a**	**79.16± 20.67a**	**88.66± 22.22a**	**100.00± 22.09ab**	**105.16± 21.46ab**	**114.1667± 21.66abd**	**123.33± 22.76ac**
6 = 2 disks	**75.83± 18.77bc**	**77.50± 21.21bc**	**82.83± 24.35abc**	**95.16± 32.31abcd**	**103.66± 35.78abc**	**114.00± 38.84abc**	**120.33± 41.34abc**	**130.1667± 43.13abc**	**140.83± 42.84ab**
7 = 1 disk	**61.00± 13.43ab**	**70.33± 12.87bc**	**83.33± 11.34abc**	**94.66± 14.29abcd**	**101.33± 15.37abc**	**109.33± 17.09abc**	**118.33± 17.42abc**	**131.3333± 19.80abc**	**146.66± 22.35ab**
8 = ½ disk	**76.16± 17.03bc**	**80.50± 17.36b**	**95.33± 24.69bc**	**107.50± 25.24bcd**	**114.5000± 27.04b**	**125.66± 30.70bc**	**135.16± 32.93c**	**147.16± 34.02c**	**161.33± 33.85b**
9 = ¼ disk	**80.50± 15.06c**	**81.16± 15.86b**	**101.5± 11.23b**	**110.83± 11.33cd**	**120.00± 10.73b**	**124.33± 11.89bc**	**128.50± 10.80bc**	**135.83± 9.26abc**	**143.50± 7.63ab**
F-Statistic	**5.33*** **	**3.64** **	**2.88** **	**2.36***	**2.29***	**2.23***	**2.33***	**2.534***	**2.95** **

Values presented are means ± SE. *, **, ***, ns = significant at P≤ 0.05, P≤ 0.01, P≤ 0.001, ns = not significant respectively.
Means followed by the same letter(s) are not significantly different from each other at P≤ 0.05.

Table 3.2: Effect of different concentrations of liquid Kelpak® and Kelpak® Plantit® disks on total wet and dry weights on shoots of *Coleonema album.*

Treatment	Wet Weight	Dry Weight
1 = Control	**1.16±0.32d**	**0.31±0.07e**
2 = 300ml	**0.58±0.24a**	**0.17±0.07abc**
3 = 200ml	**0.53±0.16a**	**0.17±0.05ab**
4 = 100ml	**0.62±0.19ac**	**0.16±0.05a**
5 = 50ml	**0.73±0.27abc**	**0.21±0.08abcd**
6 = 2 disks	**0.92±0.38bcd**	**0.26±0.11cde**
7 = 1 disk	**0.96±0.15bd**	**0.25±0.03bcde**
8 = ½ disk	**1.03±0.38bd**	**0.29±0.11de**
9 = ¼ disk	**0.80±0.07abc**	**0.23±0.02abcde**
F-Statistic	**4.03****	**3.16****

Values presented are means ± SE. **, P≤0.01, Means followed by the same letter(s) are not significantly different from each other at P≤0.05.

Table 3.3: Effect of different concentrations of liquid Kelpak® and Kelpak® Plantit® disks on total wet and dry weights on roots of *Coleonema album.*

Treatment	Wet Weight	Dry Weight
1 = Control	**2.26±0.77b**	**0.36±0.12c**
2 = 300ml	**1.08±0.44a**	**0.15±0.06ab**
3 = 200ml	**1.18±0.37a**	**0.16±0.05ab**
4 = 100ml	**1.06±0.19a**	**0.11±0.02a**
5 = 50ml	**1.43±0.68a**	**0.20±0.11b**
6 = 2 disks	**1.22±0.45a**	**0.15±0.06ab**
7 = 1 disk	**1.47±0.28a**	**0.18±0.03ab**
8 = ½ disk	**1.48±0.69a**	**0.17±0.09ab**
9 = ¼ disk	**1.18±0.19a**	**0.15±0.02ab**
F-Statistic	**3.26****	**5.40*****

Values presented are means ± SE. **, ***, P≤0.01, P≤0.001 respectively. Means followed by the same letter(s) are not significantly different from each other at P≤0.05.

Table 3.4:
Effect of different concentrations of liquid Kelpak® and Kelpak® Plantit® disks on total pre and post-harvest wet and dry weights of *Coleonema album.*

Treatment	Pre harvest Total wet weight	Post-harvest Total wet weight	Post-harvest Total dry weight
1 = Control	**2.49±0.90b**	**3.42±1.07b**	**0.68±0.18c**
2 = 300ml	**1.13±0.22a**	**1.66±0.67a**	**0.32±0.13ab**
3 = 200ml	**1.13±0.10a**	**1.72±0.51a**	**0.34±0.09ab**
4 = 100ml	**1.34±0.38a**	**1.69±0.33a**	**0.27±0.07a**
5 = 50ml	**1.40±0.70a**	**2.17±0.94a**	**0.41±0.20ab**
6 = 2 disks	**1.53±0.61a**	**2.15±0.83a**	**0.42±0.16ab**
7 = 1 disk	**1.65±0.23a**	**2.44±0.42a**	**0.44±0.06ab**
8 = ½ disk	**1.24±0.48a**	**2.52±1.05a**	**0.47±0.20b**
9 = ¼ disk	**1.19±0.51a**	**1.98±0.23a**	**0.38±0.04ab**
F-Statitic	**4.03****	**3.38****	**3.95****

Values presented are means ± SE. ** at P≤0.01, Means followed by the same letter(s) are not significantly different from each other at P≤0.

Chapter 4 : The effect of organic seaweed concentrates on the growth parameters of *Erica verticillata* for conservation and pot plant production in South African National Botanical Gardens.

The effect of organic seaweed concentrates on the growth parameters of *Erica verticillata* for conservation and pot plant production in South African National Botanical Gardens.

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4.1 ABSTRACT

Erica's are popular indigenous and garden plants in South Africa. They attract an array of pollinators such as bees, birds and butterflies. *Erica verticillata* with its striking pink floral tubular flowers is an ideal Erica for an indigenous coastal garden. These Erica's grow in acid soils and not much research has been conducted on using growth regulators successfully during cultivation. Growth regulator, Kelpak® is used by many growers but has not been scientifically tested to determine its success on pot plant production in botanical gardens. The aim of the study was to assess whether Kelpak® growth regulators had an effect on the growth of *E. verticillata*. Ninety Plants were divided into three different treatments in a randomised block design. A control with 10 plants received no treatment. A second treatment consisting of 40 plants received liquid Kelpak® and third treatment consisting of 40 plants received Kelpak® Plantit® disks. The results indicated that the liquid Kelpak® and Kelpak® Plantit® disks influenced the growth of *E. verticillata*. Seradix 2 was attributed to the successful production of *E. verticillata*.

Keywords: CFR, Kelpak®, KNBG, plant height, Plantit® disks, SANBI, Seradix 2

4.2 INTRODUCTION

4.2.1 Erica verticillata

The Cape Floristic Region (CFR) is one of the world's hottest biodiversity hotspots, which is associated with an endemic bird area and a global priority for conservation action (Da Bush, 2010). An intriguing aspect of the CFR is the relatively small number of taxa that have differentiated into extraordinary species-rich families (e.g. the genus *Erica* includes 658 species within the CFR) (Dreyer, 2014). Erica is by far the largest genus in the Cape flora (Oliver & Oliver, 2000). Floral distinction in Erica is extensive and includes floral types that are pollinated by birds and insects as well as by wind (Van der Niet *et al*., 2014). There are

approximately 70 species of Erica that are pollinated by birds, mainly the short-billed orange breasted sunbird (*Anthobaphes violacea*), the only nectar-feeding bird closely associated with *Erica* species (Geerts & Pauw 2011).

Erica verticillata (Ericaceae) commonly known as the Cape Flat Erica or Whorled Heath is a narrow endemic species restricted to the southern areas of the Cape Flats Sand Fynbos (CFSF) vegetation of the Cape Peninsula (Schumann et al., 1992; Gibbs, 2014). The species is very tough, hardy and averaging between 1.5 and 2 m in height, but older plants have been recorded growing to 3 m. It flowers intermittently throughout the year with peak flowering periods from January to March with beautiful pink, tubular flowers arranged in neat whorls organised in distinct groups up the main stems and near the tips of the branches (SANBI, 2013).

4.2.2 Conservation importance of *Erica verticillata*

The Cape Flats, *E. verticillata* competed with the pressures of urban expansion, small scale farming, draining of wetlands, alien vegetation and possibly flower picking for the famous Cape Town flower market (Gibbs, 2014). It was considered to be extinct in the wild by the second half of the 20th century; however it was re-discovered in Protea Park Botanical Garden, Pretoria in 1984 and since then, distributed to various other botanical gardens around the world (Oliver & Oliver, 2000). It has been successfully conserved by the Royal Botanic Gardens, Kew and other European botanic gardens for a number of years. Kirstenbosch National Botanical Garden (KNBG) has propagated and grown *E. verticillata* to become a flagship conservation species being re-introduced in CFSF reserves within the city of Cape Town. These suburban areas include Kenilworth Racecourse Conservation Area, Rondevlei Nature Reserve and the lower Tokai Park under management of the South African National Parks. The red listed status of *E. verticillata* remains extinct in the wild until after survival of three burn cycles without replanting, its' status will be re-assessed (SANBI, 2013). Success stories include the re-introduction of more than 2000 seedlings of *E. verticillata* to Prinskasteel wetland in Tokai where it occurred in a narrow band on the Cape lowlands at the foot of the Table mountain chain from the Black River in Mowbray and Rondebosch all the way to Zeekoeivlei near Muizenberg (Da Bush, 2010). The future of *E. verticillata* will depend on *ex situ* conservation and commercialised cultivation for its survival (Winter & Botha, 1994). It could be beneficial to use the correct organic seaweed concentrate to improve the cultivation success of *E. verticillata.*

4.2.3 Growing *Erica verticillata*

Erica verticillata is one of the relatively few South African *Erica* species that is comparatively tough, disease resistant and recommended for planting in both gardens and containers (Hitchcock *et al.,* 2013). It has also become popular garden plant that attracts bees, beetles, bumblebees, hawk moths and sunbirds for nectar and pollination (Hitchcock *et al.,* 2013). Few fynbos species are easy to grow and with over 700 species of Erica, there are at least 50 species that have good potential for garden plants (Brown & Duncan, 2006).

The Whorled Heath is easily propagated from heel cuttings, selected from side shoots usually taken in autumn, although they have been rooted successfully throughout the year at KNBG (SANBI, 2013). Vegetative propagation is important to preserve the genetic identity of species where particular characteristics of plants need to be maintained (Bowes, 1999). Cuttings should preferably be rooted in compartment trays in a medium consisting of equal parts of 6 mm milled pine bark and polystyrene or perlite granules with intermittent mist. Rooting is enhanced by using a rooting hormone for roots to develop in three to six weeks' time. Once the cuttings are well rooted they are removed from the propagation benches and placed on hardening off benches and weaned for three weeks. Rooted cuttings are planted and established in a well-draining potting mix. Producing Erica species vegetatively is more costly compared to seed, however their survival rates are higher than seedlings (Thompson, 2005). Plants propagated from cuttings taken from mature plants will flower in the first season whereas plants grown from seed usually only flower in their third year (SANBI, 2013). At this stage they should be fed with diluted (50%) liquid, organic seaweed-based fertilizers (SANBI, 2013).

E. verticillata does not readily produce seed unless two or more different clones of this species are in relatively close proximity for cross pollination (Hitchcock *et al.,* 2013). The seed should preferably be sown in late summer or autumn in seed trays containing a fynbos soil mixture, lightly covered with sand, and then subjected to smoke treatment before the trays are watered. This allows for maximum smoke penetration into the medium containing the seed. Smoke is generated in a drum using dry and semi-dry fynbos plant material and is pumped into a sealed plastic tent stretched over a frame containing the trays of *E. verticillata* seed. The smoke is allowed to settle onto the medium, leaving a brownish film, after which the trays are removed from the tent and watered. The Marsh Erica germinates after three to four weeks. Seedlings, once germinated will benefit from diluted (50%) liquid, organic seaweed-based fertilizers (SANBI, 2013).

Fynbos of the Cape Floristic Region (CRF) is often cited as shrub lands on low-nutrient, well drained sandy soils under 'Mediterranean' climate zones which have unusually diverse floras (Sinclair, 2012; SANBI, 2013). Fynbos occurs in acidic, (pH 4.4 – 7.0) soils that are deprived

of nutrients especially nitrogen and phosphorus. *E. verticillata* grows in acidic soils but prefers more water than other *Erica* species (Hitchcock *et al.,* 2013). Furthermore the symbiotic relationship of *Micorrhiza* sp*.* fungi is important for the survival of Erica species during the hot dry summers of the Western Cape. Micorrhiza enters the roots and sends out a network of hyphae strands that increase the water-absorbing capacity of the plant during dry periods (SANBI, 2013). *Erica* species must be provided with specific conditions under which to thrive naturally (Van Jaarsveld, 2010) and it is best to plant them during the autumn or winter in the winter-rainfall areas of South Africa. Pruning should be carried out on mature plants only after flowering in order not to cut away new buds. *Erica* species should be regularly fed with organic liquid fertilizers or control-release fertilizers low in phosphorus. This practice is followed at KBNG however the growth parameters and success on individual species such as *E. verticillata* has never been scientifically recorded. The aim of this study therefore was to investigate the effects of various concentrations organic growth regulator, Kelpak® and Kelpak® Plantit® disks on the growth responses of *E. verticillata* (Ericaceae).

4.3 MATERIALS AND METHODS

4.3.1 The Experiment layout and treatments

Erica verticillata was vegetatively propagated using tip cuttings. Cuttings were dipped in Seradix No.2, rooting hormone powder and placed in a propagation medium consisting of 1 part fine milled pine bark and 1 part polystyrene. Cuttings were placed under bottom heat at 25 degrees Celsius and misting, in an environmentally controlled greenhouse. Cuttings took between 3-4 weeks to root. Rooted cuttings were moved out of the greenhouse and hardened off in a shade house (50% shading) for two weeks. They were then planted into 15 cm square plastic pots which contained a well-drained nutrient poor fynbos soil mix developed by (KNBG). The fynbos soil mix was made up of 8 parts, 6 mm - 12 mm fine milled pine bark, 1 part milfeed wet Consol® glass sand, 2 parts “Malmesbury Grof” sand and 1 part agricultural lime. The trials were conducted in the Disa House of the Living Collections Nursery of Kirstenbosch National Botanical Garden, Newlands, Cape Town, Longitude 18.43209E and Latitude 33.99039S. This greenhouse covered with opaque polycarbonate sheeting which allow for a constant, controlled environment. The average temperature in the greenhouse ranged between 13 and 23 degrees Celsius. The greenhouse contained growing benches constructed of brick and 13 mm drainage chip on the surface on which the plants were placed.

The pots were labelled according to the treatments applied. The experiment was arranged using a block design, consisting of 90 plant samples. The experiment layout consisted of the control, liquid Kelpak® and Kelpak® Plantit® disks. The seaweed products are manufactured

by Kelp Products (Pty) Ltd., Simonstown, City of Cape Town, South Africa. The control consisted of 10 plants which were arranged is numerical order 1-10. The liquid Kelpak® consisted of 40 plants divided into 4 groups, containing 10 plants and each group having a different treatment with group 1, 300 ml, group 2, 200 ml, group 3, 100 ml and group 4, 50 ml. consisted of 40 plants which were divided into 4 groups, containing 10 plants each and each group having a different treatment with group 1, 2 disks, group 2, 1 disk, group 3, ½ disk and group 4 ¼ disk.

The disks were cut into the various size classes according to the various treatments each plant would receive. The treatments that received the half and quarter size disks were cut using a guillotine. The pots were filled half way with soil and the disks were inserted on top and covered lightly. The rooted cuttings were then inserted and the rest of the soil added to fill the pot. The treatments that received the Kelpak® liquid dosage was administered using a measuring cylinder assuring accuracy. The control received no treatments and the plants that contained the control only received water.

4.3.2 Data collection

Prior to planting, pre-trial measurements were recorded of each individual cutting. A standard ruler was used to measure the root length and shoot length of each plant, measurements were taken in mm. The weight of each plant was measured with a Radwag AS 220/C/2 analytical scale in grams. Plant growth, in terms of plant height was measured on a weekly basis. Plant height was measured with a standard ruler, from the surface of the medium to the tip of the tallest leaf. Watering during the trial period was conducted, using a hand held hose with a rose head sprayer twice a week and during the 10^{th} week of the trial of the trial once a week. Each container received an average of 250 ml of water. The 18^{th} week of the trial final readings of the plants was conducted.

The plants were then moved to CPUT, Department of Horticulture research laboratory, Bellville campus. Plants were carefully harvested and their roots were rinsed with tap water. A standard ruler was used to measure the root length and shoot length of each plant. The roots and shoots were then separated with secateurs from each other. The fresh weight of each root and shoot weighed and recorded. The combined total weight in grams was captured. The roots and shoots were placed in a manila brown paper bag and placed in a laboratory oven at 55° C for 48 hours. The plants were then removed from the oven and the dry weight of each root and shoot weighed. The combined total was also recorded.

4.3.3 Statistical analysis of plants

Collected data was analysed using the One–way analysis of variance (ANOVA), with the computations being done using the software program STATISTICA [Software Program version 2010 (Statsoft Inc., Tulsa, OK, USA)]. The Fisher least significance (L.S.D.) was used to compare significant treatment means *at P*≤0.05 level of significance (Steel & Torrie, 1980).

4.4 RESULTS

The effect of different concentrations was most significant (P≤ 0.001) in treatment 8, ½ disk Kelpak® on the dry root weight of *E. verticillata* (Table 4.1). The liquid treatments 3, 4, 5 and disk treatments 6, 7 and 9 were similar in results but indicated higher dry weights than the control. The liquid application of 300 ml was also significantly (P≤ 0.001) different to other treatments and higher than the control (Table 4.1).

The effect of different concentrations of Kelpak® was significant (P≤ 0.05) on the post-harvest root length of *E. verticillata* (Table 4.2). The applications of 2 disks and 50 ml treatments indicated the most significant (P≤ 0.05) post-harvest root lengths compared to the other treatments and the control (Table 4.2). Liquid applications of treatments 3 and 4 and disk treatment 8 were similar in results but also indicated a significant (P≤ 0.05) difference to the control (Table 4.2). The significant (P≤ 0.05) difference of disk treatments 7 and 9 were similar in results but also indicated longer post-harvest root lengths compared to the control. The applications of liquid treatments had much better increase in root length than the disks on *E. verticillata*.

Table 4.3 is based on the research of propagating and growing *E. verticillata* for ex situ conservation. At week one fresh side shoots were collected from healthy vigorous *E. verticillata* bushes. Heel cuttings were selected and treated with Seradix 2 rooting hormone to initiate callusing for rooting. The 4th week indicated successful rooting results and juvenile plants were removed from the heated beds and placed in a hardening-off area, to acclimatize, up until week 7. In week 8 the juvenile plants were removed from the plug trays, measured and weighed before being potted into the KNBG nursery fynbos soil mix. The plants also received various liquid and disk Kelpak® treatments and a control that received no treatment in week 8. From weeks 9 to 21 the plants were monitored for general health and treated with preventative fungicides and an insecticide. Plant growth in terms of height was also measured in these weeks. Week 22 indicated the harvesting of the plants, where root length, shoot length, root weight, shoot weight and total plant weight was measured.

4.5 DISCUSSION

Many efforts have been done to find effective methods for the protection of rare ornamental plants (Khoshbakht & Hammer, 2007). *E. verticillata* listed as extinct in the wild (EW) on the IUCN Red List is one species that is depended on *ex situ* conservation, propagation and growing for commercial sale for its survival (Winter & Botha 1994). According to Hitchcock *et al.,* (2013) *E. verticillata* does not freely produce seed unless two or more different clones are in close proximity, for intraspecific cross-pollination. It is further reported by Hitchcock *et al.,* (2013) that seed production is low when compared to other wild Ericas. In this research the aim was to investigate which Kelpak® application increased root growth of vegetatively propagated *E. verticillata* in potted plants over a trial period. Physiological responses indicated by agricultural plants treated with seaweed concentrates are thought to be due to auxins and cytokinin plant hormones found in Kelpak® (Robertson-Andersson *et al.,* 2006). Auxins orchestrate almost every aspect of plant growth and development (Overvoorde *et al.,* 2010).

The results indicate that the ½ disk application influenced the dry root weight more than other Kelpak® treatments over the trial period. This would indicate that the disk applications were absorbed by the plants much faster than the other treatments as the disk broke down or dissolved much faster in the soil allowing the plant to absorb the nutrients at a quicker rate. According to Stirk and van Staden (2010), roots have the highest concentration of cytokinins and in mature and lateral roots elevated levels of cytokinins appear. This study would support the research that Kelpak® had a significant ($P \leq 0.001$) effect on the dry root weight of *E. verticillata*. Featonby-Smith and van Staden (1984) reported that cytokinins are involved in nutrient mobilization in vegetative grown plants. Their study also indicated that seaweed concentrates increased the dry mass of plants by 24% in comparison to the control.

Hammer and Khoshbakht (2007) stated that rare and threatened ornamental plants are often displayed in gardens and are protected and serve as a source of cultivated material which can be eventually taken for the reintroduction to the wild. Plant hormone cytokinins control a range of plant activities shown to increase meristemic cells in roots and shoots, (Miransari & Smith, 2014). The applications of Kelpak® 2 disks and 50ml treatments indicated the best post-harvest root lengths compared to the other treatments and the control. Auxins are produced in a plant's shoot tips and move downwards to the root zone and one of its functions is to signal a plant to increase its root system (Kelpak®, 2013).

Crouch *et al.,* (1990), indicated that plants receiving half or single applications of Kelpak had little effect on the yield of plants but double concentrations increased plant yield. In this study both the smallest application, 50 ml and the double application, 2 disks had positive effects

on the post-harvest root length of *E. verticillata*. According to Hitchcock *et al.,* (2013), *E. verticillata* is one of the easiest Ericas to grow from cuttings and using the double or low applications rates will increase root growth of this species thereby strengthening the growth of the plant for ex situ re-introduction to natural habitats for conservation and preservation of this species.

Bowes (1999) reported that an advantage of asexual propagation is very important to preserve the genetic integrity of species especially if particular characteristics of the plant need to be retained. Sexual reproduction cannot maintain these characteristics as each individual off spring is genetically different. Survival rates of vegetatively propagated material would be higher according to Thompson (2005) but more expensive than producing seedlings. For successful rooting of *E. verticillata* practices of KNBG were followed such as using Seradix 2 rooting as it which contains the auxin indole-3-butyric acid (IBA). Plants during photosynthetic periods synthesize endogenous auxin, which is transported to the base of the cutting and promotes root formation (Leakey, 2004). A study conducted by Tsobeng *et al.,* (2013) established that Seradix 2 increased the rooting percentage of a tree species *Pentaclethra macrophylla.* According to Leakey (2004), cuttings treated with auxins root better than untreated cuttings. Cuttings of *E. verticillata* were placed in compartmental plug trays containing a propagation mix of 50% fine milled pine bark and 50% polystyrene granules. These cuttings rooted in late summer under intermittent mist in raised greenhouse benches. These benches maintained a surface temperature of 24°C this is ideal for Erica propagation as stated by Brown and Duncan (2006). *E. verticillata* was produced from heel cuttings as heel cuttings provide a firm rooting base so that the roots are well protected against possible root rot (Browse, 1979). The rooted cuttings were removed from the hotbeds and placed in a well-lit area to harden-off for 3 weeks. Hardening off allows plants to adapt from being in a protected, stable greenhouse environment to changeable outdoor conditions. Watering of these rooted juvenile plants was conducted twice a day during this period. These juvenile plants were treated with a preventative fungicide to avoid any root rot. Once the juvenile plants were adapted to the outdoor environmental conditions these plants were ready to be potted into the KNBG nursery fynbos soil mix.

4.6 CONCLUSION

In conclusion the use of liquid Kelpak® and Kelpak® Plantit® disks had significantly increased the dry root weight and post-harvest root length of *E. verticillata* grown in 15 cm pots over a period. The Kelpak® Plantit® disks indicated higher growth rates in the dry root weight of *E. verticillata* but both the liquid and the disk had a positive effect on the post-harvest root length. The results also conclude that the successful rooting of *E. verticillata* was attributed to rooting hormone Seradix 2 under greenhouse heating conditions. It is

recommended that this study continue with research using the *E. verticillata* to determine if Kelpak® growth regulators have an influence potential growth development. It is also recommended that further research be conducted to determine the potential of sea weed growth regulators on the establishment of replanting *E. verticillata* in the wild for conservation purposes. This in itself can unlock the potential to increase natural flowering and the production of more seed in saving the species in the wild.

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Table 4.1: Effect of different concentrations of liquid Kelpak® and Kelpak® Plantit® disks on the wet and dry root weights of *Erica verticillat*a.

Treatment	Wet Weight	Dry Weight
1= Control	**1.145±0.66c**	**0.14±0.11c**
2=300ml	**1.00±0.39bc**	**0.09±0.03b**
3= 200ml	**0.63±0.34a**	**0.05±0.02ab**
4= 100ml	**0.73±0.20ab**	**0.06±0.01ab**
5= 50ml	**0.62±0.15a**	**0.04±0.01ab**
6= 2 disks	**0.73±0.24ab**	**0.05±0.02ab**
7= 1 disk	**0.74±0.29ab**	**0.05±0.01ab**
8= ½ disk	**0.64±0.18a**	**0.03±0.01a**
9= ¼ disk	**0.81±0.20abc**	**0.05±0.01ab**
F-Statistic	**2.01 *ns***	**4.02*** **

Values presented are means ± SE *** at P≤0.001, respectively. Means followed by the same letter(s) are not significantly different from each other at P≤0.05. NS = not significant.

Table 4.2: Effect of different concentrations of liquid Kelpak® and Kelpak® Plantit® disks on the pre and post-harvest root lengths of *Erica verticillat*a.

Treatment	Pre-harvest Root Length	Post-harvest Root Length
1= Control	**47.71±17.62ab**	**158.28±54.15b**
2=300ml	**38.00±7.32a**	**221.85±24.81c**
3= 200ml	**45.28±23.15a**	**195.71±38.20abc**
4= 100ml	**62.71±18.56b**	**193.71±26.73abc**
5= 50ml	**45.28±10.22a**	**172.57±31.69ab**
6= 2 disks	**47.42±12.47ab**	**172.00±47.48ab**
7= 1 disk	**45.28±17.28a**	**200.00±44.27ac**
8= ½ disk	**47.00±16.87ab**	**185.00±18.61abc**
9= ¼ disk	**43.71±13.98a**	**208.57±19.92ac**
F-Statistic	1.20 *ns*	2.13*

Values presented are means ± SE* P≤0.05 respectively. Means followed by the same letter(s) are not significantly different from each other at P≤0.05. NS= not significant

Table 4.3: A proposed production schedule for the propagation and cultivation of *Erica verticillata* grown at 13 - 23 °C in a controlled green house for ex situ conservation.

Week	Development stages	Actions	Treatments	Media	Watering schedule	Temp of hotbeds and greenhouse	Growth Measurements
1	Fresh shoot cuttings collected from mature *E. verticillata*	Propagate cuttings in 200 compartment plug trays.	Heel cuttings dipped in Seradix 2 rooting hormone and dipped in fungicide Captan to prevent damping off.	50 % fine milled pine bark and 50 % polystyrene.	Misting every 10 minutes for 10 sec burst of spray. Water pH 6.55	Hotbed 24°C and greenhouse ambient temp. 28°C	Each heel cutting was cut to exactly the same size: 40mm
4-7	Cuttings rooted in week 4 and a juvenile plant established	Cuttings removed from hotbeds and moved to hardening off area for 3 weeks	Cuttings are treated with a preventative fungicide Captan	Cuttings remain in propagation media	Hardening off area rooted cuttings is watered twice per day, morning and afternoon. Water pH 6.55	Greenhouse temperature 25°C	No growth measurements taken.
8	Juvenile plant	Plants potted	Trial Treatments: 10 x plants-control 40 x plants-liquid Kelpak 40 x plants-Kelpak Plantit disks	KNBG-fynbos soil mix, pH 6.5	Plants watered twice per day, average watering 250ml per plant. Water pH 6.55	Temp 23°C, Plants grown under opaque polycarbonate covered house with well-ventilated front and back sides to allow good airflow movement.	Rooted plants weighed, roots washed and the length measured in mm. Shoot length measured in mm.
9-21	Maturing plant	Overall conditions and health of plants monitored	The plants were sprayed with Dimeldex fungicide and Khorhinor pesticide	KNBG-fynbos soil mix	Plants watered twice per day during the initial growing stages and then to once a day, average watering 250ml per plant. Water pH 6.55	Greenhouse temp. 23°C	Weekly readings of plant height was recorded using a standard ruler
22	Mature plant	Plants ready for harvesting	No treatments	Greenhouse temp. 23°C	Plants watered two times a day. Water pH 6.55	Greenhouse temp. 23°C	Height, root length, wet and dry shoot weight, wet and dry root weight and total weight measured

Chapter 5 : The effect of Kelpak® growth regulators on the growth of *Leucospermum cordifolium* flowering potted plants for Garden Centre production.

The effect of Kelpak® growth regulators on the growth of *Leucospermum cordifolium* flowering potted plants for Garden Centre production.

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5.1 ABSTRACT

The pincushion, *Leucospermum cordifolium* is a popular cut flower and very attractive indigenous garden plant in South Africa. Due to its low soil acidity growing requirements not much research has been done on using growth regulators successfully during cultivation. Growth regulator, Kelpak® is used by many growers but has not been scientifically tested to determine its success on fynbos container production. This study assessed the plant root and shoot weights and the total plant weights of *L. cordifolium*. Ninety plants were divided into three different treatments in a randomised block design. A control with 10 plants received no treatment. A second treatment consisting of 40 plants received liquid Kelpak® and third treatment consisting of 40 plants received Kelpak® Plantit® disks. The aim of the study was to establish the effects of the different Kelpak® treatments on the plants growth. The results indicated that root and shoot weight and total weights of plants grown in the Kelpak® Plantit® disks treatments were significantly higher.

Keywords: CFR, KNBG, potted plants, plant height, roots, SANBI, shoots

5.2 INTRODUCTION

5.2.1 Fynbos

The Cape Floral Region (CFR) at the southern tip of Africa contains over 9,000 plant species in an area of only 80,000 km2 (Moran & Hoffman, 2012). This floral region, the smallest of 6 floral kingdoms contains distinctive vegetation, fynbos (Fajinmi, 2012). Fynbos is dominated by shrubs with an unusual mixture of plant types of different shapes and sizes (Esler *et al.,* 2014). The word 'fijnbosch' is derived from Dutch and describes the narrow-leaved bushes which characterises this vegetation type (Dorrat-Haaksma & Linder, 2012). A characteristic of the Cape fynbos are the prevalent Proteaceae species (Anderson *et al*., 2014), are possibly one of the oldest flowering groups of plants (Vogts, 1982). Leucospermum which are members of the Proteaceae family are just as popular as the Protea and easily recognisable by their flower heads with long exposed styles. These styles form a mass

together which resembles a pincushion, commonly named the pincushion Protea (Vogts, 1982).

5.2.2 *Leucospermum cordifolium*

The economic, functioning and conservation use of fynbos has led scientists, conservationists and volunteers to collect a wealth of information on the ecology, geographical distribution and evolutionary history of Proteas (Schurr *et al.,* 2012). The foundation for this knowledge was laid by the intense research on Proteaceae conducted in the 1980's (Schurr *et al.,* 2012). *L. cordifolium* is a rounded spreading shrub up to 1.5 m high and 2 m in diameter, hard green leaves, with a single main stem and several horizontally drooping stems (SANBI, 2013). The beautiful flowering blooms decorate the shrub for 6-8 weeks during flowering season in spring (Vogts, 1982). The flowers produce large, hard, nut-like seeds, which fall to the ground and are collected by ants stored in the soil, and germinate only after a fire (SANBI, 2013, Paterson-Jones 2007). *L. cordifolium* is an excellent garden plant as a focal point or planted in groups while the flowers make excellent cut flowers which last up to four weeks (Joffe, 2001) with perfect long-stemmed blooms. This species also features in many popular hybrids (Rebelo, 1995). The pincushion Protea with its attractive striking flower head has many colours ranging from yellow, to orange, through to a deep red-pink. The smaller foliage, which can be soft, smooth and hairy are more delicate than other members of the Proteaceae family. These characteristics together with its hardiness and ease of rooting caused Leucospermum to be identified as being suitable as a potted plant for the European horticultural markets (Hoffman & Du Plessis, 2013).

5.2.3 Growing *Leucospermum cordifolium*

Seed is sown in autumn when the nights become cooler; using fresh seed produces best results (SANBI, 2013). Seed should be soaked in a solution of water and hydrogen peroxide which has been added, at the ratio of 1% of the total volume. This mixture softens the outer seed coat which permits the seed to be oxygenated. The seeds are treated with a preventative fungicide prior to being sown in an open seed bed or seed tray. The seeds are sown in a well-drained acidic soil mixture and covered lightly with a fine milled pine bark. Seeds germinate after four weeks and are potted in batches at different stages as the seed germinate at different times.

Vegetative propagation by cuttings is made from the beginning of November to March. Vegetative propagation is important to maintain the genetic integrity of species and propagation by cuttings where particular characteristics of a plant need to be maintained

cannot be conserved through sexual reproduction (Bowes, 1999).The material selected should be semi-hardwood, 6-10 cm long and of the current season's growth. A rooting hormone is used to stimulate rooting and the cuttings are placed in a growing house with bottom heat (25ºC) and intermittent mist (SANBI 2013). According to Thompson (2005) plants that are produced vegetatively are more costly compared to seed, however their survival rates are higher than seedlings.

In nature soils are mainly deprived of nutrients especially nitrogen and phosphorus (Brown & Duncan, 2006). Fynbos grows in poor infertile sandy soils that are not suited for agricultural crops. It occurs mainly on acidic soils with exceptions on alkaline soils and dune sands close to the coast (Esler *et al.,* 2014). *L. cordifolium* grows in acid, nutrient poor soils and occurs only in the winter rainfall area with its wet winters from May to September and hot, dry summers from December to the end of February (SANBI, 2013). The organic content of most fynbos soils are very low in the amount of nutrients available or species requirement is often not known. It is there very difficult to estimate the nutrient type and concentrations to grow Leucospermum commercially and testing organic growth regulators would be a first option. This study found very little documentation related to organic growth regulators and their effect on fynbos species. The aim of the study therefore was to investigate the effects of Kelpak® and Kelpak® Plantit® disks growth regulators on the growth responses' of *L. cordifolium*.

5.3 MATERIALS AND METHODS

5.3.1 The Experiment layout and treatments

Leucospermum cordifolium was vegetatively propagated using tip cuttings. Cuttings were dipped in Seradix No.2, rooting hormone powder and placed in a propagation medium consisting of 1 part fine milled pine bark and 1 part polystyrene. Cuttings were placed under bottom heat at 25 degrees Celsius and misting, in an environmentally controlled greenhouse. Cuttings took between 3-4 weeks to root. Rooted cuttings were moved out of the greenhouse and hardened off in a shade house (50% shading) for two weeks. They were then planted into 15 cm square plastic pots which contained a well-drained nutrient poor fynbos soil mix developed by (KNBG). The fynbos soil mix was made up of 8 parts, 6 mm - 12 mm fine milled pine bark, 1 part milfeed wet Consol® glass sand, 2 parts "Malmesbury Grof" sand and 1 part agricultural lime. The trials were conducted in the Disa House of the Living Collections Nursery of Kirstenbosch National Botanical Garden, Newlands, Cape Town, Longitude 18.43209E and Latitude 33.99039S. This greenhouse covered with opaque polycarbonate sheeting which allow for a constant, controlled environment. The average temperature in the greenhouse ranged between 13 and 23 degrees Celsius. The greenhouse

contained growing benches constructed of brick and 13 mm drainage chip on the surface on which the plants were placed.

The pots were labelled according to the treatments applied. The experiment was arranged using a block design, consisting of 90 plant samples. The experiment layout consisted of the control, liquid Kelpak® and Kelpak® Plantit® disks. The seaweed products are manufactured by Kelp Products (Pty) Ltd., Simonstown, City of Cape Town, South Africa. The control consisted of 10 plants which were arranged is numerical order 1-10. The liquid Kelpak® consisted of 40 plants divided into 4 groups, containing 10 plants and each group having a different treatment with group 1, 300 ml, group 2, 200 ml, group 3, 100 ml and group 4, 50 ml. consisted of 40 plants which were divided into 4 groups, containing 10 plants each and each group having a different treatment with group 1, 2 disks, group 2, 1 disk, group 3, ½ disk and group 4 ¼ disk.

The disks were cut into the various size classes according to the various treatments each plant would receive. The treatments that received the half and quarter size disks were cut using a guillotine. The pots were filled half way with soil and the disks were inserted on top and covered lightly. The rooted cuttings were then inserted and the rest of the soil added to fill the pot. The treatments that received the Kelpak® liquid dosage was administered using a measuring cylinder assuring accuracy. The control received no treatments except for water.

5.3.2 Data collection

Prior to planting, pre-trial measurements were recorded of each individual cutting. A standard ruler was used to measure the root length and shoot length of each plant, measurements were taken in mm. The weight of each plant was measured with a Radwag AS 220/C/2 analytical scale in grams. Plant growth, in terms of plant height was measured on a weekly basis. Plant height was measured with a standard ruler, from the surface of the medium to the tip of the tallest leaf. Watering during the trial period was conducted, using a hand held hose with a rose head sprayer twice a week and during the 10^{th} week of the trial of the trial once a week. Each container received an average of 250 ml of water. The 18^{th} week of the trial final readings of the plants was conducted.

The plants were then moved to CPUT, Department of Horticulture research laboratory, Bellville campus. Plants were carefully harvested and their roots were rinsed with tap water. A standard ruler was used to measure the root length and shoot length of each plant. The roots and shoots were then separated with secateurs from each other. The fresh weight of each root and shoot weighed and recorded. The combined total weight in grams was

captured. The roots and shoots were placed in a manila brown paper bag and placed in a laboratory oven at 55° C for 48 hours. The plants were then removed from the oven and the dry weight of each root and shoot weighed. The combined total was also recorded.

5.3.3 Statistical analysis of plants

Collected data was analysed using the One–way analysis of variance (ANOVA), with the computations being done using the software program STATISTICA [Software Program version 2010 (Statsoft Inc., Tulsa, OK, USA)]. The Fisher least significance (L.S.D.) was used to compare significant treatment means *at P*≤0.05 level of significance (Steel & Torrie, 1980).

5.4 RESULTS

The effect of different concentrations of Kelpak® growth regulators on fresh and dry weights indicated significance (P≤0.01) on shoots of *L. cordifolium*. The wet shoot weight significantly (P≤0.01) increased from ½ disk - 50ml concentration of liquid Kelpak® compared to the control (Table 5.1). The wet weights of treatments 7 and 9 indicated the same results but were significantly (P≤ 0.01) different to the control. Liquid applications 2 and 4 also produced similar results which were also higher than the control. The disk applications had much more an effect on the wet shoot weight of *L. cordifolium* compared to the liquid application.

The dry shoot weight significantly (P≤0.01) increased from ½ disk to ¼ disk treatment compared to other treatments and the control (Table 5.1). Treatment 6, 2 disks showed significance (P≤ 0.01) on the dry shoot weight of *L. cordifolium* compared to the control (Table 5.1). Treatments 4, 5 and 7 displayed similar results but indicated higher dry shoot lengths than the control. The disk applications provided more effective dry shoot weight results than the liquid applications.

The effect of different concentrations of Kelpak® growth regulators on the dry weights indicated significance (P≤ 0.01) on the roots of *L. cordifolium* (Table 5.2). The dry root weight significantly increased (P≤ 0.01) with applications of disk treatments 7 and 9. The application of treatment 5, 50 ml also significantly (P≤ 0.01) increased dry root weights compared to the control. Liquid treatments 3, 4 and disk treatments 6 and 8 also improved dry root weight compared to the control. The application of 300 ml liquid Kelpak® had the lowest effect of all treatments on the dry root weight but in comparison to the control it was higher (Table 5.2).

In Table 5.3, treatment 5, 50 ml liquid Kelpak® had the most significant (P≤ 0.05) difference on the total post-harvest wet weight compared to other treatments and the control with no concentration. The 1 disk application in treatment 7 indicated significance (P≤ 0.05) above the other disk treatments and the control (Table 5.3).The disk treatments of 6 and 9 indicated

similar results but showed significance (P≤ 0.05) above the other treatments and the control (Table 5.3). Although the application of treatment 8 provided the lowest result of all the disk applications the significance (P≤0.05) was recorded above the control. Treatments 8 and 9 indicated significant differences (P≤ 0.01) between other treatments and the control with no concentration on the total post-harvest dry weight of *L. cordifolium* (Table 5.3). Liquid applications 5, 6 and disk applications 7 and 8 provided similar results but indicated significant (P≤ 0.01) increase in total post-harvest dry plant weight compared to the control. The disk applications have provided more effective growth increases on the total dry weight of the plants in comparison to the liquid applications.

5.5 DISCUSSION

Research on fynbos and the commercialization of Proteaceae species, Leucospermum, Serruria, Leucadendron, Protea and other species for the cut flower market, has been well documented (Reinten *et al.,* 2011). Hoffman and Du Plessis (2013) reported that *L. cordifolium* has become a popular potted plant especially in the European flower markets. The use of seaweed concentrates as growth regulators is well established in the agriculture and horticulture industries (Stirk *et al.,* 2004). Kelpak® a natural seaweed concentrate contains plant hormones auxins and cytokinins (Sosnowski *et al.,* 2013) which stimulates prolific adventitious root formation, improves nutrient status and improves the top growth, which increases the quality of crops (Rekanović *et al.,* 2010). In this study the rationale was to investigate which Kelpak® application would increase the root, shoot and total weight growth of *L. cordifolium* vegetative propagated plants over a trial period. The effect of different concentrations of Kelpak® growth regulators on fresh and dry weights indicated improved growth results on the shoots of *L. cordifolium*.

This study is in agreement with Papenfus *et al. (2*013) where Kelpak® had an increase in wet and dry shoots of *Abelmoschus esculentus* seedlings. The 2 disk, ½ disk and 50ml liquid Kelpak® applications indicated the best wet shoot weight results of all treatments which is comparable with a study conducted by Basak (2013), indicating that seaweed concentrates especially Kelpak® stimulates stronger vegetative growth and shoot elongation. It is possible that the plant cell structure of L. *cordifolium* was more conceivable to the absorption of lower liquid levels and smaller disks parts of the seaweed concentrate, whereas stronger liquid concentrations could have inhibited growth. Similarly Kelpak® applications of ½ disk and ¼ disk treatments increased the dry shoot weight of *L. cordifolium* compared to other treatments. According to Sosnowski *et al.,* (2013) the use of a biostimulator caused a significant increase in dry matter yield, number of shoots, and leaf blade length of *Festulolium braunii.* The smaller applications also indicate that these disks break down much

faster in the soil which allowed the roots to absorb it much quicker and hence having an effect on dry shoot weight.

The 50 ml, 1 disk and ¼ disk applications indicated the best growth results for *L*. *cordifolium* dry root weights. Rekanović *et al.,* (2010), states that seaweed concentrates stimulates root growth and uptake of nutrients from soil causing faster growth and development of plants. A study conducted by Matysiak (2011) indicated that plant hormones, auxins and cytokinins found in Kelpak® intensify plant growth and increase the root weight. These findings would collaborate with the current study which indicated that the liquid and disk applications of Kelpak® had an effect on the dry root weight of *L. cordifolium*.

The liquid application of 50 ml Kelpak® increased the total post-harvest wet weight compared to other treatments and the control with no concentration of *L. cordifolium*. This indicated that the liquid concentrate is available to the plant immediately for uptake through the roots compared to the disk which has to be dissolved in the soil before uptake by the roots. The ½ and ¼ disk applications of Kelpak® indicated the best results among treatments and the control on the total post-harvest dry weight of *L. cordifolium*. Although the disk provided better results in the post-harvest dry weights the dosage or application ratios were smaller in comparison to the other applications indicating that once these disks started to dissolve in the soil, they were immediately available to the plant. Coenen and Lomax (1997) reported that cytokinins and auxins act synergistically together to regulate physiological responses in plants. These Kelpak® applications elicited a physiological response in the total wet and dry post-harvest weights of *L. cordifolium*.

5.6 CONCLUSION

In conclusion the use of liquid Kelpak® and Kelpak® Plantit® disks had significantly increased the wet and dry shoot weights, dry root weights and post-harvest wet and dry total plant weights of *L. cordifolium* grown in 15 cm pots over the growth period. The Kelpak® Plantit® disks indicated higher growth rates in the dry shoot weights of *L. cordifolium* but both the liquid and the disk had a significant effect on the wet shoot weight. The liquid and the disks were also responsible for the improved dry root weight. The liquid application indicated the best post-harvest wet weight but the disks improved the post-harvest dry weights. It can there be confirmed that organic seaweed concentrates such as Kelpak® is effective on the growth development of *L. cordifolium.* Little is known what impact the growth regulator will have on flowering formation. It is therefore recommended that further research be conducted using *L. cordifolium* to determine if organic growth regulators will increase the number of flowering buds to promote flowering of potted plants.

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Table 5.1: Effect of different concentrations of liquid Kelpak® and Kelpak® Plantit® disks on the wet and dry shoot weights of *Leucospermum cordifolium.*

Treatment	Wet Weight	Dry Weight
1= Control	**9.18±1.54c**	**2.39±0.68d**
2= 300ml	**8.75±2.18bc**	**2.23±0.72cd**
3= 200ml	**9.35±3.06c**	**2.14±0.53cd**
4= 100ml	**8.77±1.62bc**	**1.66±0.66abc**
5= 50ml	**6.82±1.16a**	**1.73±0.34abc**
6=2 disks	**6.59±0.80a**	**1.63±0.17ab**
7= 1 disk	**6.93±1.23ab**	**1.69±0.33abc**
8= ½ disk	**6.54±1.69a**	**1.31±0.68a**
9= ¼ disk	**7.03±1.59ab**	**1.42±0.39a**
F-Statistic	**3.218****	**3.3133****

Values presented are means ± SE **, P≤0.01 respectively. Means followed by the same letter(s) are not significantly different from each other at P≤0.05.

Table 5.2: Mean yield results of total fresh and dry weights of roots for *Leucospermum cordifolium* various treatments.

Treatment	Wet Weight	Dry Weight
1= Control	**5.39±2.39c**	**0.74±0.46c**
2= 300ml	**4.95±1.86bc**	**0.55±0.27b**
3= 200ml	**3.81±1.75abc**	**0.36±0.19ab**
4= 100ml	**4.05±2.03abc**	**0.34±0.16ab**
5= 50ml	**2.44±1.50a**	**0.24±0.14a**
6=2 disks	**3.44±1.96ab**	**0.34±0.20ab**
7= 1 disk	**2.80±1.74a**	**0.28±0.12a**
8= ½ disk	**3.95±1.17abc**	**0.39±0.10ab**
9= ¼ disk	**3.18±1.49ab**	**0.28±0.14a**
F-Statistic	**1.9682ns**	**3.3447****

Values presented are means ± SE **, P≤0.01 respectively. Means followed by the same letter(s) are not significantly different from each other at P≤0.05. NS = Not Significant.

Table 5.3: Mean yield results of total pre and post-harvest fresh and dry weights of *Leucospermum cordifolium* at various treatments.

Treatment	Pre harvest total weight	Post-harvest total weight wet	Post-harvest total weight dry
1= Control	**11.32±1.99ab**	**14.58±3.64e**	**2.73±0.59c**
2= 300ml	**11.26±3.95ab**	**13.71±3.73de**	**2.79±0.91c**
3= 200ml	**10.82±5.10ab**	**13.16±4.40cde**	**2.51±0.70bc**
4= 100ml	**11.08±3.12ab**	**12.82±3.44bcde**	**2.01±0.64ab**
5= 50ml	**9.04±3.20ab**	**9.27±2.49a**	**1.98±0.41ab**
6=2 disks	**8.71±3.28a**	**10.04±2.53abc**	**1.97±0.22ab**
7= 1 disk	**7.13±1.73a**	**9.73±2.39ab**	**1.97±0.37ab**
8= ½ disk	**21.79±2.05b**	**10.50±2.37abcd**	**1.71±0.74a**
9= ¼ disk	**8.12±2.57a**	**10.21±2.88abc**	**1.71±0.51a**
F-Statistic	**0.89*ns***	**2.75***	**3.2939****

Values presented are means ± SE*, **, P≤0.05, P≤0.01 respectively. Means followed by the same letter(s) are not significantly different from each other at P≤0.05. NS = Not Significant.

Chapter 6 : GENERAL RECOMMENDATIONS AND CONCLUSIONS

6.1 GENERAL CONCLUSIONS AND RECOMMENDATIONS

The introductory chapter established a rationale for the study with a review on the importance to conserve and grow fynbos. The study undertook to identify new knowledge and information that may be used to solve growing problems of fynbos species thereby increasing the conservation of fynbos species and the use of it for gardening and conservation. Three fynbos species, *Coleonema album, Erica verticillata* and Leucospermum *cordifolium* were selected for the study as they are commercially available in retail garden centres. In nature these species grow in acidic nutrient poor soils and do not prefer to be grown in nutrient rich soils.

The uses of seaweed concentrates in the agricultural industry have been documented for many years. Kelpak® a growth regulator was developed from the seaweed *Ecklonia maxima* and used as source of seaweed concentrate in the agricultural sector. This reference study indicated that Kelpak® increased the growth yield of agricultural crops but there was no scientific evidence to prove what effect it had on fynbos species. It was also documented that Kirstenbosch National Botanical Garden used Kelpak® in the growing production of their plants but no scientific evidence had been recorded. Kelpak® contained a unique source of natural plant hormones, auxins and cytokinins which have been proven to have a number of beneficial physiological effects on plants. This study therefore recommended that the effects of Kelpak® on the growth of the three fynbos species be investigated.

Chapter two concluded that the Cape Floristic Region is home to a unique vegetation type known as Fynbos. Fynbos was cited as the ultimate vegetation type for any water wise gardener and that a shift in gardening with traditional European plants to more indigenous plants was taking place. It is recommended that appropriate growing methods to improve the growth of fynbos using Kelpak growth regulators® are required to encourage large scale production nurseries to maximise growth of these species. This will encourage gardening enthusiasts to garden with more indigenous plants.

Chapter three investigated which Kelpak application increased the plant root and shoot length, fresh and dry root and shoot weight of *Coleonema album* over a trial period. The aim of the study was to collect data which could establish which application provided the best growth results for *C. album*. The study found that there had not been any documented use of Kelpak® on the growth of *C. album*. In conclusion the use of liquid Kelpak® and Kelpak® Plantit® disks had significantly increased the shoot, root growth and total weight of plants in *C. album* grown in 15 cm pots over an 18 week period. The liquid Kelpak® indicated higher

growth rates in the initial growing stages of C*. album* as the liquid was immediately available to the plant. The Kelpak® Plantit® disks had better influence on the growth over a longer period as the disk dissolved or broke down at a slower rate which eventually became available to the plant. It is recommended that future studies continue with research using the *C. album* to determine if Kelpak® growth regulators have an influence on the nutrients of fynbos soils. It is also recommended that further research be conducted on using *C. album* to determine if Kelpak® growth regulators are affected by changes in the greenhouse growing environment and or the composition of the plant cells.

Chapter four investigated which Kelpak® application increased root growth of *E. verticillata* over a trial period in pots. Although Kelpak® was used extensively in the agricultural trade the study established that there had not been any documented use of Kelpak® on the growth of *E. verticillata*. In conclusion the use of liquid Kelpak® and Kelpak® Plantit® disks had significantly increased the dry root weight and post-harvest root length of *E. verticillata* grown in 15 cm pots over a period. The Kelpak® Plantit® disks indicated higher growth rates in the dry root weight of *E. verticillata* but both the liquid and the disk had a positive effect on the post-harvest root length. The results also conclude that the successful rooting of *E. verticillata* was attributed to rooting hormone Seradix 2 under greenhouse heating conditions. It is recommended that this study continue with research using the *E. verticillata* to determine if Kelpak® growth regulators have an influence potential growth development. It is also recommended that further research be conducted to determine the potential of sea weed growth regulators on the establishment of replanting *E. verticillata* in the wild for conservation purposes. This in itself can unlock the potential to increase natural flowering and the production of more seed in saving the species in the wild.

Chapter five investigated which Kelpak® application increased root and shoot growth of *Leucospermum cordifolium* over a trial period. The objective of the study was to establish the best application rates for plants grown in containers using Kelpak® growth regulators. Kelpak® is used extensively in the agricultural trade to increase growth and yield of crops but no scientific evidence was documented on the use of Kelpak® on the growth of *L. cordifolium*.In conclusion the use of liquid Kelpak® and Kelpak® Plantit® disks had significantly increased the wet and dry shoot weights, dry root weights and post-harvest wet and dry total plant weights of *L. cordifolium* grown in 15 cm pots over the growth period. The Kelpak® Plantit® disks indicated higher growth rates in the dry shoot weights of *L. cordifolium* but both the liquid and the disk had a significant effect on the wet shoot weight. The liquid and the disks were also responsible for the improved dry root weight. The liquid

application indicated the best post-harvest wet weight but the disks improved the post-harvest dry weights. It can therefore be confirmed that organic seaweed concentrates such as Kelpak® is effective on the growth development of *L. cordifolium.* Little is known what impact the growth regulator will have on flowering formation. It is therefore recommended that further research be conducted using *L. cordifolium* to determine if organic growth regulators will increase the number of flowering buds to promote flowering of potted plants.

This study was aimed at identifying new knowledge that may be used to overcome the various challenges of growing fynbos species. Fynbos is part of the Cape Floral Kingdom and many of its species is listed on the IUCN Red Data list. This study concludes that the application of organic seaweed growth regulator was successful in growing fynbos species *Coleonema album*, *Erica verticillata* and *Leucospermum cordifolium* in nursery containers. It further recommends that extended growth periods for these and other species be researched as it remains unknown what the response of organic seaweed growth regulators on fynbos species in more permanent containers would be. It is hoped that the findings of this study will support the wholesale production industry and botanical gardens in the efforts of propagating and cultivating endangered Red Data species through sustainable conservation practices.

Chapter 7 :REFERENCES

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