EFFECT OF GROWTH RETARDANT CHEMICALS ON STOCK PLANT GROWTH AND SUBSEQUENT ROOTING OF VERBENA (*Verbena hybrida*) CUTTINGS

M.Sc. Thesis

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January, 2011
Jimma University

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M.Sc. Thesis

Submitted to the School of Graduate Studies

Jimma University College of Agriculture and Veterinary Medicine

In Partial Fulfillment of the Requirements for the Degree of

Master of Science in

Horticulture (Floriculture)

BY

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January, 2011
Jimma University

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As thesis research advisor, I hereby certify that I have read and evaluated this thesis prepared, under my guidance, by Ismael Yassin, entitled **Effect of Growth Retardant Chemicals on Stock Plant Growth and Subsequent Rooting of Verbena *(Verbena hybrida)* Cuttings.** I recommend that it can be submitted as fulfilling *thesis* requirement.

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DEDICATION

I would like to dedicate this M.Sc. thesis work to my family.

STATEMENT OF THE AUTHOR

First, I declare that this thesis is my work and that all sources of materials used for this thesis have been duly acknowledged. This thesis has been submitted in partial fulfillment of the requirements for M.Sc. degree in Horticulture (Floriculture) at Jimma University College of Agriculture and Veterinary Medicine and is deposited at the university library. I solemnly declare that this thesis is not submitted to any other institutions anywhere for award of any academic degree, or certificate.

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ACKNOWLEDGEMENTS

I have special thanks to the almighty Allah for giving me health and encouragement to do everything.

I am profoundly indebted to my advisors, Mr. Nigussie Kassa and Dr. Ali Mohammed for their unreserved advice, guidance and valuable suggestions in each and every step of my research work and thesis write up. Without the encouragement, insight and professional expertise of my advisors, the completion of this work would not have been possible.

I would like to thank Jimma University College of Agriculture and Veterinary Medicine for providing the fund required for the study. I am grateful to Florensis Ethiopia Plc for their support during my research by providing all the necessary materials. In addition, my thanks also go to all staff of the company for providing their unreserved assistance.

I earnestly acknowledge the contribution of Debre Zeit Agricultural Research Center for allowing me to use equipment for measuring of fresh and dry weight with sincere appreciation to my friend Desta Fekadu for facilitating the support of the research center.

I want to thank Jimma University College of Agriculture and Veterinary Medicine staff members, specially, Mr.Mulatu Wakjira, Mr.Desalegn Weldesenbet, Mr.Abebe Nigussie, Mr.Amsalu Nebiyou, and Mr.Gezahegne Berecha for their kind assistance during thesis write-up by providing valuable literatures and comments.

Last, by no means least, special thanks are reserved to my family and relatives for their strong support throughout my life and my academic career.

BIOGRAPHICAL SKETCH

Ismael Yassin Mohammed was born on June 10, 1984 in Kombolcha town. He completed his elementary school at Assai Primary School from 1990 to 1998 and secondary education at Bole Senior Secondary School and Lideta Catholic Cathedral School from 1999 to 2002.

In 2003, he joined Jimma University and graduated in 2006 with BSc. degree in Horticulture. After graduation, he joined Top Flowers Plc. as a greenhouse and pack-house supervisor for production and post harvest handlings of cut Rose. Finally, in October 2009, he rejoined Jimma University College of Agriculture and Veterinary Medicine to pursue his MSc. degree in Horticulture (Floriculture).

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LIST OF ABBREVIATIONS

ANOVA	Analysis of Variance
CDP	Copalyl diphosphate
CRD	Completely randomized design
GA_s	Gibberellins
GGPP	Geranylgeranyl diphosphate
IAA	Indole-3-aceti acid
LSD	Least significant difference
NADH	Nicotine adenine dinucleotide
P	Probability
PLC	Private Limited Company
RH	Relative Humidity
SAS	Statistical Analysis System

EFFECT OF GROWTH RETARDANT CHEMICALS ON STOCK PLANT GROWTH AND SUBSEQUENT ROOTING OF VERBENA (*Verbena hybrida*) CUTTINGS

ABSTRACT

Verbena hybrida and its cultivars are some of the most popular bedding plants in the landscape and are in the best top ten of bedding plants in terms of annual sale and market acceptance. Like several bedding plants, Verbena hybrida may begin to stretch, developing thin, long and less branched stems after transplanting which forces growers to use some sort of growth retardation. Chemical growth retardation is the most commonly used method for commercial greenhouse production. Effects of chemical growth retardants may vary by location, species, and variety. Growth retardants are widely used in cutting producing farms in Ethiopia without any scientifically justified concentration levels. Consequently a two phase greenhouse experiment was conducted at Florensis Ethiopia plc situated at Koka from November 2009 to February 2010 with the objective of determining the growth response of stock plants and subsequent rooting of cuttings of Verbena hybrida cultivar Vegas Scarlet to individual and combined application of Cycocel and Alar. The experiment was laid down as 4x4 factorial experiment involving two different growth retardants as factor, with four different levels in a complete randomized design (CRD) with three replications. The two growth retardants used were Alar at concentrations of 0g L^{-1}, 1g L^{-1}, 2g L^{-1}, and 3g $L^{-1}$and, Cycocel at concentrations of 0ml L^{-1}, 0.5ml L^{-1}, 1ml L^{-1}, and 1.5ml L^{-1}. On stock plants it was vividly observed that interaction of Alar and Cycocel considerably affected most of the parameters except average leaf area, stem diameter and number of cuttings. Combination of 1ml L^{-1} CCC and 2g L^{-1} Alar significantly produced the maximum number of main branches (14.35). However, combined application of 1.5ml L^{-1} CCC and 3g L^{-1} Alar resulted in the minimum internode length (3.88cm), stem length (10.15cm), canopy diameter (48.44cm), root dry weight (12.39g), root fresh weight (72.86g), shoot fresh weight (196.74g), and shoot dry weight (35.81g). Conversely, maximum values for internode length (7.56cm), stem length (21.35cm), canopy diameter (60.33cm), shoot fresh weight (229.80g), and shoot dry (57.84g) were obtained from stock plants without any treatment. Application of 1ml L^{-1} CCC with 0g L^{-1} Alar resulted in the maximum root fresh weight (82.01g), whereas maximum root dry weight (16.17g) was obtained from application of 1.5ml L^{-1} CCC with 0g L^{-1} Alar. As the concentration of Cycocel increased from 0ml L^{-1} to 1.5ml L^{-1} average leaf area revealed a decline trend from 8.26 $cm^2$to 6.97 cm^2. The same trend has also been observed from Alar applications (8.16cm^2 to 6.83cm^2). Significantly maximum number of cuttings was achieved at a concentrations 1ml L^{-1} CCC (75.0) and 2g L^{-1} Alar (70.4). Regarding stem diameter, 1.5ml L^{-1} CCC and 3g L^{-1} Alar showed the maximum value (1.45mm and 1.40mm, respectively). The minimum stem diameter was obtained from 0ml L^{-1} CCC (1.27 mm) and 0g L^{-1} Alar (1.37mm). Significant persistent effect of the treatments on subsequent rooting was observed only on number of roots per cutting. Maximum number of roots per cutting (9.37) was recorded from cuttings collected from stock plants treated with the combination of 0.5ml L^{-1} CCC and 2g L^{-1} Alar. Whereas cuttings collected from non treated stock plants produced less number of roots per cuttings (7.80). Generally, the investigation verified the influence of Alar and Cycocel on growth of Verbena hybrida and cutting yield without causing a significant reduction on rooting performance of cuttings. Hence, application of 1ml^{-1}L Cycocel, which demonstrated positive influence on cutting production, can be recommended for use by commercial growers. However, further researches are imperative regarding the economic aspects and other production factors like type of media used for rooting purpose, time and type of application of retardants to come up with pertinent and comprehensive recommendations.

1. INTRODUCTION

In Ethiopia, floriculture is becoming very promising business opportunity and growing very fast. Currently there are more than 250 projects of floriculture industry in Ethiopia. The total flower production has shown increment and the number of exporters had increased from just five in 2002 to more than 100 in 2008. During the same period, foreign exchange earnings abruptly increased from US $1.5 million to in excess of US $125 million and it is expected to go up to 200 million US $ in the current fiscal year (Danse *et al*., 2009). This sub-sector has also played an important role in generating employment by creating job opportunities for more than 50,000 peoples (Taylor, 2010). Additionally, Ethiopia is advancing fast by exporting large volume of floriculture product mainly to the European Union countries. This very fast growth suggests the immense potential of the sector to become one of the major foreign currency sources in the near future (EHPEA, 2011; Danse *et al*., 2009).

Cut flower production is the major component of floriculture industry in Ethiopia which covers an estimated 80% of the production area (EHPEA, 2011; Danse *et al*., 2009). Another category of the industry comprises propagators, who are mainly subsidiaries of European breeding companies. Until 2004, Ethiopian exports of unrooted and rooted cuttings were negligible. From 2005 onwards, however, some breeding companies have set up propagation facilities in Ethiopia for production of planting material (Joosten, 2007). At the moment, there are five cutting propagating farms in Ethiopia producing pot plant and bedding plant cuttings which covers about 20% of the production area of the floriculture industry (EHPEA, 2010).

Worldwide consumption of bedding and potted plants is increasing rapidly. The total consumption in 1990 was about 14.2 billion dollars, at some 21% higher than the value in 1985. In 1995, this market increased to about 19 billion dollars and in 2001, this value increased to about 23 billion dollars. Further growth is expected due to the growing expandable income in many regions of the world. The US, with about one third of the consumption, has the largest share of the total world consumption, followed by Germany (about 20%), Italy and, France (Nigussie, 2005). Armitage (2007) signifies the use of bedding plants by saying gardens have no life without bedding plant.

Many of the cultivated bedding and potted plants reveal undesirable stretch growth habits. To keep them shorter, compact and more branched growth regulation is important (Kessler, 1998). One way of controlling excessive plant growth is treating plants with chemical growth retardants. Chemical growth retardants are very useful tools for controlling the height of bedding plants and also to create more branched stock plants for maximizing cutting yield (Cox, 2007). For many species and cultivars of bedding plants the treatment with such chemicals is an obligatory commercial procedure (Anita *et al*., 2003). Chemicals, including Bonzi®, Sumagic®, Alar®, Cycocel®, Alar®/Cycocel® mixed use, and Ethephon®, are most commonly used and effective in controlling growth of numerous horticultural crops, including many herbaceous perennials (Burnett *et al*., 2000, Erwin and Warner, 2003; Latimer, 2009).

Growth retardant chemicals are highly specific. There is no obvious correlation between taxonomic classification and plant response to a particular compound. Even different cultivars of the same species may vary greatly in the responsiveness to applied chemicals (Cathey, 1964). Scientific literatures referring to the effect of different growth retardants on growth of bedding plants are often divergent regarding retardant concentration as well as the time and frequency of application (Anita *et al.*, 2003).

Plant growth retardants are justified having persistence effect on growth and quality of bedding plants (Banko *et al*, 2003). The residual effects of these chemicals can influence adventitious root formation and can even last until the plants become fully established (Whipker, 2001). Most studies with chemical growth retardants in greenhouse crops are primarily concerned with effects on the crop during production. However plants followed through the marketing channels or post-production are rarely tested on residual or carry over effects from plant growth retardants (Arnold and McDonald, 2000).

Verbena hybrida and its cultivars are some of the most popular herbaceous bedding plants in the landscape due to their floriferous and durable nature (Burnett *et al*., 2000). Some cultivars of *Verbena hybrida* are already under greenhouse cultivation in Ethiopia as stock plants. Nearly 4-5 million cuttings of *Verbena* are being exported annually. However, *Verbenas* can quickly grow up and often requires repeated pruning or transplanting to a larger pot for maintenance in greenhouse environment. For greenhouse grower, excessive growth of verbena can lead to

blow-over, plants outgrowing their pots, excessive drying between irrigations, leggy and less branched plants (Burnett *et al*., 2000). This production problem may require the application of growth retardants because these compounds often offer inexpensive and effective alternative to other mechanisms (Warren, 1990).

Plant growth regulators are widely used in cutting producing farms in Ethiopia without any scientifically justified concentration levels for Ethiopian condition. Surprisingly, the application of growth retardants carried out irrespective of cultivar types. Effects of growth retardants on *Verbena hybrida* have not been previously studied in Ethiopia. Due to the above mentioned facts, growers frequently receive complaints from their clients on matters pertaining to the product quality (personal observation and communication) which includes poor rooting performance and stretching growth during propagation.

One of the limitations of chemical growth retardants is misapplications leading to catastrophic results, which in turn lower plant quality and yield. Common consequences include phytotoxicity, delayed flowering, ruined growth habit, and stunted growth (Cavins *et al*., 2001). Hence, growers should adjust application of growth retardants to the existing conditions. In the absence of specific recommendation the grower must run a trial (Cox, 2007). Determining the effect of growth retarding chemicals on the growth responses of stock plants and rooting performance of cuttings obtained from stock plants of V*erbena hybrida* and establishing the optimum application rate is mandatory to intensify production in terms of quality and quantity through viable and economically feasible manner and to be competitive in the global market. Moreover, it will encourage other investors to join this emerging business. Thus, this study was initiated to determine the effect of individual and combined applications of Cycocel and Alar on growth of stock plants and subsequent rooting ability of cuttings of *Verbena hybrida* cultivar Vegas Scarlet.

2. LITRETURE REVIEW

2.1 Bedding Plants

Bedding plants are herbaceous plants that are normally grown in outdoor beds to provide colorful blooms or foliage for a landscape (Beaulieu, 2009). These plants are (usually annuals) massed with others to produce the maximum in visual appeal by means of providing the five basic elements of landscape design (color, scale, line, form and texture). A landscape designer skillfully arranges each bedding plant in relation to the accompanying annuals, perennials, shrubs and trees (Boodley, 1998). Bedding plants are therefore plants that provide temporary colour or texture to the landscape and means of providing quick colour in the garden and no other group of plants can bring such exuberant colour (Dole and Wilkins, 2005).

It is difficult to strictly define bedding plants because annuals and perennials are grown together (Nelson, 1998). From greenhouse production point of view, bedding plants are a heterogeneous group of plants started under controlled conditions that share a common production methodology. Bedding plants include a wide range of plant species and cultivars that may have multiple applications. These include herbaceous annuals and perennials, biennials, herbs, ground covers, vegetables, small fruits (strawberry), and a few woody species (Kessler, 1998).

The main use of bedding plants in the past has been in formal bedding schemes but they can bring life to any part of the garden in just a few months. They are also widely used in containers, pots, planters, window boxes and hanging baskets. As gardens become smaller and patios more important, their use in this way will become more popular. Bedding plants are especially useful in new gardens where recently planted permanent trees and shrubs are still small and where much space remains to be filled. Bedding can also be used to fill up gaps that sometimes occur in beds of hardy annuals, in newly planted herbaceous borders or in mixed plantings of shrubs, perennials and bulbs (Armitage, 2007).

2.2 Verbena

Verbena is a genus in the family Verbenaceae. It contains approximately about 250 species of annual and perennial herbaceous or semi-woody flowering plants, six or seven of which are in cultivation. The majority of the species are native to the New World from Canada south to southern Chile, but some are also native in the Old World, mainly in Europe (Boodley, 1998). *Verbena* ×*hybrida* results from the crossing of *V. peruviana*, *V. incisa*, *V. phlogiflora*, and *V. teucroides*. The demand for this species is high, and additional species are being incorporated every year. This species is the most common of all verbenas in the market (Armitage, 2007).

Verbenas are a very useful species due to wide variation in plant height (15-90cm high) and different growth habit. It has both spreading and upright growth habit (Rao, 2004). *Verbena hybrida* tends to have a loose, sprawling growth habit that makes it suitable for pots and hanging baskets. Once the verbena plants are established, they require little care and will spread out to cover the bed space allotted for them (England, 2007). The toothed or cut leaves usually grow opposite each other and reach a length of 2 to 7 cm. During growing season, slender stalks arise and bear flattened clusters of flowers. The flowers are small, with five petals, and borne in dense spikes. They may be multi-colored, red, rose, peach, pink, purple, lavender, or blue, usually with a white eye (Boodley, 1998). Verbena can propagate from seed, cuttings or division (Rao, 2004). The seedlings are transplanted at three pair of leaves stage but if the seedlings are overgrown before transplanting, they may be pinched back at the time of transplanting (Dole and Wilkins, 2005).

Boodley (1998) suggested that for growing verbena in greenhouse, temperature should be at least 20-22^{o}C and the plants need sufficient humidity (60-65%) during the period of vegetative growth. Verbenas do best in sunny sites but will tolerate a wide range of soils as long as it is well- drained. Many verbenas will tolerate partial shade. These plants are useful for edging paths, growing on bank, in beds, in pots, and in hanging baskets (England, 2007). They are easy to grow both in ground and pots, and extensively used as garden display and ground cover. Verbena is in the best top ten of bedding plants in terms of annual sale and market acceptance (Burnett *et al*., 2000).

2. 3 Plant Growth Regulation

Plant growth regulation refers to any compounds or process used to produce a specific type of growth response, such as inhibition or promoting of internode elongation, encouraging of formation of lateral branches or root development. Growth regulating compounds or processes usually affect plant growth through alternation of endogenous plant growth regulators (plant hormones) levels (Dole and Wilkins, 2005; Kumar and Prasad, 2005). Plant growth regulators can be divided into two groups: the natural growth regulators (plant hormones) and chemical growth retardants. The five major classes of naturally occurring plant growth regulators are: auxins, cytokinins, gibberellins, abscisic acid and ethylene. They usually exist in plants and crops at a concentration lower than 1μM; above this they are generally considered supra optimal (Naqvi, 2002).

These plant hormones generally regulate plant growth and development by affecting cell division, elongation and differentiation. They also mediate various physiological responses to help plants adapt to stresses. Each class of hormone has a multiplicity effects, but has unique physiological functions (Naqvi, 2002; Huang, 2007;Carvalho *et al*, 2008). A natural plant hormone is defined as an organic compound synthesized in one part of a plant that, in very small concentrations, is translocated to another location, where it causes a physiological response (Naqvi, 2002). Therefore, an exogenous application of hormone may lead, not simply to a response by a single tissue, but also may be accompanied by a change in hormone concentration, and frequency and availability of a receptor protein which could amplify the hormonal signal (Puglisi, 2002).

2.4 Chemical Growth Retardation

Chemical growth retardants are synthetic compounds which are used to reduce plant growth in a desired way without changing developmental patterns or being phytotoxic (Magnitskiy, 2004). The selection of the word "retardant" for this type of compound implies a special action by the chemicals. These chemicals imitate or influence the activities of natural growth regulators (Carvalho *et al*, 2008). Treated plants are not ultimately stunted or completely suppressed from growing; rate of development and vigor of the plants are unaffected (Nelson, 1998). Growth

retardant Chemicals are the most commonly used and important growth regulators in floriculture. For commercial greenhouse production of bedding plants there is a need to be a fast response of plants from any treatment of height control. Chemical growth retardants are given priority for commercial production because of their very fast response compared with other cultural, biological and physical methods (Bailey and Whipker, 1998).

Additionally, cultural practices used by growers are not always possible due to factors such as cost as it is not conducive to automation and requires manual labor or the presence of mixed crop types in a single greenhouse. Thus ornamental growers may decide to use chemical applications in order to manipulate plant growth characteristics (Whipker, 2001). Chemical growth retardants may be helpful in manipulating the growth of bedding plants until improved shorter cultivars can be obtained via breeding and/or biotechnology (Anderson and Davis, 1989b).

Many floriculture crops undergo a triphasic pattern of plant growth: 1) slow initial growth that occurs immediately after propagation or after a pinch, 2) rapid vegetative phase and elongation, and 3) slow final reproductive growth during which the flowers develop. The effective use of chemical growth retardants requires that the chemicals be applied prior to or during the rapid growth phase. Growth retardants cannot shrink plants after they are grown and late application of a growth retardant will have a limited effect on the final plant height (Dole and Wilkins, 2005).

When used in appropriate concentrations, these compounds influence the plant architecture in a typical fashion, which according to Grossmann (1990) is characterized by phenomena such as; inhibition of shoot growth (plant height, internode elongation, leaf area) with unchanged number of internodes and leaves and with intensified green leaf pigmentation, and maintained or slightly promoted root growth. Opposing suggestion was mentioned by Cathey (1964) which confirmed applications of growth retardants measurably inhibited root formation or delayed root development.

Normally chemical growth retardants are applied during cloudy weather. The effectiveness of Cycocel and Alar sprays is increased when conditions favor slow evaporation. Plants to be

sprayed should also be fully turgid during application (Cox, 2007). Alar and Cycocel are water soluble and take a long time (more than six hours) to move through plants with waxy cuticle. They need to be applied at a time when the leaf surface will stay wet for several hours, such as in the morning, evening, in humid conditions, or on cloudy days (Carey, 2008).

2.5 Description of Chemical Growth Retardants

2.5.1 Cycocel or CCC [(2-chloroethyl) trimethylammonium chloride)

Cycocel is a growth retardant which is available in liquid formulation. Chlormequat chloride is the active ingredient in Cycocel and it constitutes 11.8% of per liter of the product. It is used extensively to control shoot growth on many floricultural crops. Cycocel effectively control the stem elongation of a wide variety of bedding plant crops grown in packs, pots, hanging baskets, and plug trays (Banko *et al*., 2009). Cycocel may be applied as a spray or drench and is commonly applied as a foliar spray (Carey, 2008). Cycocel typically used at the rate of 1.5 to 5 ml L^{-1}on *Begonia*, geranium (*Pelargonium*), hibiscus (*Hibiscus rosa-sinensis*), poinsettia (*Euphorbia pulcherima*), and other crops. The recommended label of Cycocel for *verbena* is 0.4 to 3 ml L^{-1} spray (Bailey and Whipker, 1998; Banko *et al*, 2009). Because foliar uptake is slow, Cycocel is most effective if the foliage moist for 8 to 12 hours. Cycocel is not persistent and needs to be applied more than once if the cropping period is long (Andersen, 1989b).

2.5.2 Alar [(butanedioic acid mono (2, 2-dimethylhydrazide)]

Alar is also known under the brand name B-Nine. It's sold as a soluble powder containing 85% of active ingredient or daminozide plus a wetting agent (Kumar and Prasad, 2005). Alar is an effective height retardant labeled for use in *azalea, pot chrysanthemum, gardenia, hydrangea* and many bedding and foliage plants (Delaune, 2005). Currently, it is one of the most frequently applied growth retardant, which has found a wide application primarily in growing pot plants as well as many bedding ornamentals (Anita *et al*, 2003; Banko *et al*, 2009). It is very mobile and moves to all parts of the plant after being applied which is advantageous if the crop has a tight canopy (Carey, 2008). Therefore, uniformity of spray application is not as critical as it is with

some other chemicals. It is rapidly broken down in the media, so it is not effective as a drench (Nelson, 1998). Growth retardants vary greatly in activity and persistence. The least active and shortest lived is Alar which is safe on the greatest variety of ornamental plants and is active for about 10 days (Carey, 2008). In general, it is less phytotoxic and has a short-term effect that seldom results in over stunting of treated plants (Delaune, 2005). Alar is commonly applied at concentrations of 1.25 to 5g L^{-1} (Carey, 2008). For verbenas, 2.5-5 g L^{-1} Alar is the recommended rate (Bailey and Whipker, 1998; Kumar and Prasad, 2005; Banko *et al*, 2009).

2.6 Plant Growth Retardants Mode of Action

Modern Plant growth retardants have a variety of modes of action and affect plants in different ways. Some growth retardants are synthetic versions of plant hormones and mimic their actions. Others inhibit the biosynthesis, reception, or metabolism of plant hormones and thus block the activity of plant hormones (Naqvi, 2002; Carey, 2008). Most of the available growth retardants are anti-gibberellins (Grossman, 1990; Bailey and Whipker, 1998; Hartmann, 2002; George *et al*., 2008).

Disrupting the biosynthesis of gibberellins is an effective way of limiting stem elongation due to cell expansion and elongation. Chemicals that are commonly used to inhibit gibberellins include: A-Rest (ancymidol), Cycocel (Chloromequat chloride), Alar (daminozide), Bonzi (paclobutrazol), and Sumagic (uniconazole) (Erwin and Warner, 2003).

Over hundred gibberellins are known today, but only a few are biologically active. Most are intermediates and precursors to active gibberellins. Gibberellins synthesis is primarily carried out in young tissues such as shoot apices, new leaves, developing fruits, developing seeds, and young roots (Rademacher, 2000). The pathway of gibberellins synthesis has three stages. The gibberellins pathway begins with geranylgeranyl diphosphate (GGPP) in the proplastids. Two cyclization reactions take place, first forming copalyl diphosphate (CDP) and ultimately producing *ent*-kaurene. Two enzymes catalyze the cyclization reactions in stage one of Gibberellins synthesis: copalyl diphosphate synthase and *ent*-kaurene synthase. The second stage of gibberellin biosynthesis takes place in the endoplasmic reticulum. A series of oxidations, involving several cytochrome P450 monooxygenases, take place to produce *ent*-

kaurenoic acid from *ent*-kaurene. Intermediates of this process include *ent*-kaurenol and *ent*-kaurenal. The final product of Stage two is GA12-aldehyde, which is the first form of gibberellin produced in all plants (Krug, 2004; George *et al*., 2008).

From GA12-aldehyde, all other gibberellins are produced during stage three. The specific gibberellins formed and the processes used are species specific. Stage three generally takes place in the cytosol and involves at least one hydroxylation and one oxidation reaction. Dioxygenase enzymes including 3β-hydroxylase catalyze these reactions. During this stage biological active gibberellins are produced (George *et al*., 2008).

Chemical growth retardants however, do not all disrupt the gibberellin biosynthesis pathway in the same manner and can be further organized into categories defined by the mode through which they disrupt the gibberellin biosynthesis pathway (Rademacher, 2000; Krug, 2004). Cycocel inhibits copalyl diphosphate synthase in GA-synthesizing plants and fungus during stage one, while the synthesis of *ent*-kaurenoic acid is reduced to a lesser extent (Rademacher, 2000). Alar is in a group of its own, and interferes with 2- oxoglutarates dependent dioxygenases causing the inhibition of gibberellins in stage three of the pathway (Rademacher, 2000; Erwin and Warner, 2003; Krug, 2004). Mixed aapplication of Alar and Cycocel frequently recommended because the two products have different sites of inhibition in the GA production process, such a mix can be highly effective at suppressing stem elongation. A larger sub-group of chemical growth retardants are the N-heterocyclics, compounds with a nitrogen containing heterocycle. This group includes A-Rest, Bonzi, and Sumagic. N-heterocyclics inhibit gibberellin biosynthesis in stage two of the pathway (Krug, 2004).

It was also suggested that treating plants with growth retardants contribute to inefficient energy metabolism in plants. They inhibited oxidation of nicotinamide adenine dinucleotide (NADH) and reduction of cytochrome c, the first and the final steps in mitochondria electron transport chain, respectively (Bai and Chaney, 2001). Oxidation of NADH is dependent on cytochrome P450 and Fe-S protein (Buchanan *et al*., 2000). The last one is supposedly affected by growth retardants (Bai and Chaney, 2001). These authors speculated that lowering energy metabolism in growth retardant treated plants is a height reduction mechanism, which is an alternative to GA biosynthesis inhibition.

Growth retardants inhibit biosynthesis of sterols in plants and fungi by blocking oxidative 14α-demethylation reactions in the course of sterol biosynthesis (Fletcher *et al*., 2000; Rademacher, 2000). Biosynthesis of sterols is an important process for cell division suggesting that its inhibition represents another mechanism of growth retardation in treated plants (Asami and Yoshida, 1999).

From other growth retardants, Ethephon has a unique mechanism to modify plant growth. This compound has demonstrated the capacity to manipulate the shape, size and flowering of ornamental plants especially for pot production and to increase the number of lateral branches on many ornamental plant species (Banko *et al*., 2009). Ethephon mode of action can be explained as it promotes the production of ethylene in the plant which inhibits cell elongation (Huang, 2007). The Ethephon compound has a central phosphorus atom that is attacked by water or hydroxyl ions, which leads to the simultaneous elimination of chlorine and the liberation of ethylene (Puglisi, 2002).

2.7 Effect of Alar and Cycocel on Growth of Bedding Plants

The features that make growth retardant chemicals valuable for ornamental plant production are their effect on plant height, branching patterns, time of flowering, number of flowers, color intensity of foliage and flowers. Inhibition of GAs biosynthesis makes applications of growth retardants to plants effective in height control of various ornamental crops. These compounds regulate plant growth by affecting growth of the main and lateral shoots, internode length and leaf area (White, 2003; Magnitskiy, 2004). According to Delaune (2005) resulting stems from growth retardants application are thicker.

Controlling plant height and internode elongation is an essential aspect in producing of greenhouse bedding plants (Whipker, 2001). White (2003) stated that internode elongation is highly related with plant height where it increases, a rise also expected from plant height. Chemical growth retardants are reported to have inhibitory role on both internode length and plant height thus often used to suppress stem extension and produce a more compact, higher quality plant (Blanchard *et al*., 2008). Many greenhouse grown floricultural crops, bedding

plants and vegetable transplants tend to grow taller than desired and require height control measures. Since greenhouse growing plants are grown in crowded conditions with ideal levels of water, fertilizer, temperature, and light, as a result the plants grow quickly and have a tendency to stretch. Growers often use growth retardants to slow down the growth of a crop by restricting stem elongation through reducing the plant hormones that trigger cell expansion, and/or cell division (Dole *et al*., 1999; Carey, 2008).

Blanchard *et al.* (2008) evaluated the effectiveness of the combined application of Alar and Cycocel on reducing stem elongation of *Verbena hybrida* 'Obsession Lilac'. The study included spray concentrations ranged between 0.75 to 1.5 $ml^{-1}L$ for Cycocel and 1.25 to 5.0 $g^{-1}L$ for Alar. Internode elongation and plant height were considerably reduced with a spray application of tank mix Cycocel (1.5ml L^{-1}) and Alar (2.5 g L^{-1}) when compared with control treatments.

In addition, Burnett *et al.* (2000) justified chemical growth retardants, Sumagic, and Alar/Cycocel tank mixes, and Ethephon are effective in regulating the growth of *Verbena canadensis* under greenhouse conditions. There was a trend for plants to be smaller than controls with increasing rates of all chemicals applied to this species. The study showed that Alar and Cycocel tank mixes appreciably reduced stem height of *Verbena canadensis* at all tested concentrations (2.5 g L^{-1} with 1.5ml L^{-1}, 5 g L^{-1} with 1.5ml L^{-1}, and 7 g L^{-1} with 1.5ml L^{-1}, respectively).

Andersen and Davis (1989a), after their experiment "Effect of growth retardants on growth and flowering of *Verbena rigida* bedding plants'' suggested the combined application of Alar and Cycocel . Alar and Cycocel applied at rates of 2, 2.5, 3 g L^{-1} and 0.5, 0.7, 1 ml L^{-1} respectively to *Verbena rigida* reduced plant height with the most adequate reduction observed from the combination of 3 g L^{-1} Alar and 1 ml L^{-1} Cycocel.

Such an effect by Cycocel, Alar and other growth retardants has been widely described in other bedding and/or ornamental plants. A report by James *et al.* (2002) indicated that combined application of Alar and Cycocel affected final plant height for three cultivars of Poinsettias and two cultivars of Pansy. A mixed use of Alar and Cycocel at concentrations of 4.5 $g^{-1}L$ / 1.5$ml^{-1}L$

respectively, reduced the height of the three cultivars of poinsettia between 16 and 21% compared with the control. Similar concentration of Alar and Cycocel reduced plant height by 29-37% of the two cultivars of pansies.

Also report by Baden *et al.*(1999) in *Veronica* 'Sunny Border Blue', *Sedum* 'Autumn Joy', *Monarda didyma* 'Marshall's Delight', and *Phlox paniculata* 'David' confirmed that Alar and Cycocel mixed applications at a concentration of 5 $g^{-1}L$ / 1.5$ml^{-1}L$, respectively effectively reduced the height of the four species by 17% to 31% relative to the untreated control during greenhouse production. Finally, the authors recommended combination of Alar and Cycocel application for commercial growers wishing to control height in these cultivars.

Unlike the above reports, Gibson and Whipker (1999) showed that all combinations of Alar and Cycocel treatments (2.5 $g^{-1}L$ with 1.5$ml^{-1}L$, 2.5 $g^{-1}L$ with 3$ml^{-1}L$, 5 $g^{-1}L$ with 1.5$ml^{-1}L$, and 5 $g^{-1}L$ with 3$ml^{-1}L$, respectively) haven't provided additional control of plant height of *Brassica juncea* var. *rugosa* 'Red Giant'when compared to similar doses of individual Alar foliar sprays. However, no additional control was observed with Alar rates greater than 2.5 $g^{-1}L$.

Latimer *et al.* (1999) reported the effects of Alar on plant height of some perennial bedding plants. Accordingly, foliar spray of 5 $g^{-1}L$ Alar with multiple applications was very effective in reducing the height of *Salvia greggii, Gaura lindheimeri, Salvia leucantha,* and *Heliotropium arborescens.* Whereas, the tank mix of Alar and Cycocel applied at 5 $g^{-1}L$ with 1.5 $ml^{-1}L$ respectively, had less effect on plant height than did Alar.

Influence of foliar application of Alar and Cycocel on the height of Chrysanthemum cultivar 'Revert'was investigated by Karlovic *et al.* (2004). Alar was applied in concentrations of 1, 2 and 3 g L^{-1} and control (without treatment) while Cycocel was used in concentrations of 2, 3, 4 ml L^{-1} and a control (without treatment). The concentrations used differed significantly in their effects on plant height. 2 g L^{-1} Alar was the most efficient concentration in decreasing the upward growth than the applied concentrations with Cycocel.

Barbosa *et al.* (2005) investigated effect of growth retardants on development and ornamental quality of potted *Zinnia elegans* Jacq. and reported that Alar spray (5.0 g L^{-1}) reduced internode length and plant height of 'Yellow Marvel' *Z. elegans*. Alar sprays also presented similar results on 'Lilliput', but at lower concentrations (2.5 and 3.75 g L-1). The study also showed that Cycocel at 1.0 g L^{-1} significantly reduced plant height and side branches length.

Baden *et al.* (1998) made evaluation on nine perennial bedding plants treated with three plant growth retardants (Alar, Cycocel and Sumagic). From the study they demonstrated that combination of Alar and Cycocel (5 g L^{-1} with 1.5 ml L^{-1}, respectively*)* effectively reduced plant height of *Salvia greggii, Lantana camara,* and *Gaillardia grandiflora.* Besides, eight of the nine species tested were responsive to Alar at 5 g L^{-1}.

Ornamental plant growers often need to encourage branching in certain crops that form long runners such as V*erbena* and L*antana* (Carey, 2008). Puglisi (2002) stated that increased branch number is very important especially for the management of stock plants for generating cuttings. With increasing number of branches, the number of harvestable cuttings is expected to show escalation (Faust and Lewis, 1997). According to Healy *et al.* (1979) branching is the lifeblood of vegetatively propagated crops.

Plant growth retardants can be used to enhance branching on different bedding plants and also numerous ornamentals. For these reason they are frequently called "chemical pinchers" because they generally inhibit the growth of the terminal shoots and enhance the growth of lateral buds, thereby increasing the development of lateral branches (Latimer, 2009). Growth retardants work by interrupting apical dominance, which triggers lateral buds to grow and fill in the plant (Carey, 2008). A plant is said to display apical dominance when only one shoot predominates (Nelson, 1998). Apical dominance is a curious phenomenon and in retrospect apical dominance controls branching (cutting production) (Wilkins, 2001). Inhibitory role of growth retardants in apical dominance and its favoring of lateral branching were demonstrated by Yeang and Hillman (1984) and Abeles *et al*. (1992). During interruption of apical dominance, the tip of shoots which are the sources for auxin production will be disabled to allow lateral shoots to develop freely (Nelson, 1998).

Abbas *et al.* (2007) studied the effect of growth retardants to break apical dominance in *Rosa damescena* and concluded that the growth retardants were effective in reducing the size of plants and increasing branch number. Moreover, the intensity of the action depends upon the concentration of the chemicals used. Regarding branch number, it was observed that Alar at 1 $g^{-1}L$ attained significant superiority over the rest of treatments by producing maximum number of lateral shoots whereas Cycocel at 5 $ml^{-1}L$ produced maximum number of lateral shoots.

Anita *et al*. (2003) carried out an experiment to determine the effect of Alar on growth of bedding plants and sprayed *Tagetes patula, Impatiens walleriana,* and *Petunia hybrid* with 1.275 and 2.550 g L^{-1}. Application of 1.275 g L^{-1} showed stronger effect on growth of all the three species with stimulated shoot branching and reduced plant height which had a significant effect on the plant decorative value.

It was noticed on *Hebe x fransiscana*, a common pot plant in Europe, application of Cycocel and Alar resulted in higher number of branches at concentration level of 2 $ml^{-1}L$ and 4.5 $g^{-1}L$ respectively. But better branching results from spraying of bonzi with a concentration range of 0.3 to 0.4 $g^{-1}L$ (Adriansen and Kristensen, 1988).

Glady *et al.* (2004) worked with *Coreopsis verticillata* 'Moonbeam', *Veronica spicata* 'Sunny Border Blue', *Dianthus* 'Cinnamon Red Hots' and *Salvia nemorosa* 'May Night' to investigate if growth retardants could be used as a tool to maintain vegetative stock plants and increase the number of cuttings harvested. Weekly application of 0.5 ml L^{-1} Cycocel on *Salvia nemorosa* yielded 26% more vegetative cuttings than untreated plants. The study also demonstrated that weekly application of Cycocel on *Coreopsis verticillata* and *Veronica spicata* at 1.5 $ml^{-1}L$ yielded 32% and 30% more vegetative cuttings than controls respectively. Cycocel was not effective at maintaining more cutting production for *Dianthus* using the rates and frequencies in the experiment.

Similarly, Carpenter and Carlson (1972) also indicated increment on number of geranium cuttings using Cycocel. Cycocel when applied as a spray at 1.2 ml L^{-1} significantly increased cutting yield from all concentration levels in the range of 0.7 to 1.5 $ml^{-1}L$. Application of 1.5

$ml^{-1}L$ also enhanced cutting production compared with the controls but more phytotoxic effects on the foliage was observed. .

Chemical growth retardant treatments result in stock plants with thickened stems. These qualities allow plants to survive shipping and handling operations (Dole *and* Wilkins, 2005). Such an effect contrasts with available data in the literature concerning their effect stem diameter of plants. Barras-Ali *et al.* (2007) reported that stem diameter of *Chrysanthemum morifolium* is directly proportional to Alar concentration. Stem diameter was considerably increased by foliar spray of Alar at a concentration of 1.25, 2.5 and 5 $g^{-1}L$ with the greatest diameter obtained at 5 $g^{-1}L$.

Several studies emphasized that plants treated with growth retardants often exhibit leaves smaller in size and darker green in colour. The greening effect caused by plant treatments with growth retardants can be explained by an increase in chlorophyll content per leaf unit area due to a reduction in leaf area (Barras-Ali *et al.,* 2008). According to Grossmann (1990), reduction in leaf area is a typical response of plants from application growth retardant chemicals. Meanwhile, some literatures reported the other way, in favour of increasing of leaf area from application plant growth retardant.

Reduction in leaf area was reported by Gibson and Whipker (2004) where Cycocel has shown to decrease leaf area on stock plants of fuchsia (*Fuchsia* x *hybrida*), geranium (*Pelargonium* x *hortorum*), and lantana (*Lantana hybrida*) with increasing concentration where the highest reduction obtained at a concentration of 1 ml L^{-1} for all the species.

Similarly, Barras-Ali *et al.* (2008) studied the effect of growth retardant Alar on some anatomical and chemical changes in *Chrysanthemum morifolium.* Application of Alar was *made* at four concentrations, 0, 1.25, 2.5 and 5g L^{-1}. The study demonstrated leaf area was inversely proportional to Alar concentration where the highest leaf area observed from application of 5g L^{-1} Alar.

Amarender and Veena (2007) undertaken an experiment to exploit the potential benefits of plant growth retardants (Cycocel and Alar) over growth traits of China aster (*Callistephus*

chinensis L. nees). The growth retardants viz., Cycocel (0.5, 1, 1.5, 2 and 2.5 ml L^{-1}) and Alar (0.15, 0.3, 0.6, 0.9 and 1.2 g L^{-1}) were used in the investigation. In contrary to the above findings, Cycocel at a concentration of 2 ml L^{-1} resulted in maximum leaf area. Also, with increase in concentrations of Alar, there was a gradual increase in leaf area where the maximum value recorded from 1.2 g L^{-1}.

According to Basra (2000), plant width or canopy reduction is one of the responses from plants after treatment with growth retardants. This inhibition of excessive growth is helpful for creating compact plants especially for pot plants to adjust to the size of pots. Burnett *et al*. (2000) showed that Alar and Cycocel tank mixes at a concentration of 5 g L^{-1} with1.5 ml L^{-1} respectively recorded the maximum reduction of canopy diameter in *verbena Canadensis*. In addition, according to Banko and Stefani (1988) Alar application *to Zinnia elegans* 'Yellow Marvel' at rate of 5 g L^{-1} effectively reduced canopy size.

Some reports suggested that the root systems of treated plants were less developed than untreated ones which contribute for the decline of root fresh and dry weight. Pink dombeya (*Dombeya burgessiae*) was tested for its potential as a flowering potted plant, using the growth retardant Cycocel with concentration rates of 0.5, 1, 2 and 3 ml L^{-1}. This finding by Laubscher *et al. (*2010) depicted that the highest concentration of Cycocel caused the largest reduction in the root fresh and dry weight of *Dombeya burgessiae* with the control yielded the largest weight.The fresh and dry weights of aerial part of the plants also severely decreased with the increased Cycocel concentrations. The highest concentration caused the largest reduction, with plants in this treatment only weighing 38% of the total fresh weight of the control and 35% of the total dry weight.

In *Salvia greggi* and *Salvia leucantha,* Latimer *et al*. (1999) made similar observation with Cycocel and Alar mix applications. Both species was very responsive to a tank mix of 5 g L^{-1} Alar and 1.5 ml L^{-1} Cycocel with reduction in root fresh and dry weight 32 and 18% respectively in comparison with the control. .

According to Andersen and Davis (1989a), shoot fresh and dry weight was negatively and linearly related to Alar (2, 2.5, 3 g L^{-1}) and Cycocel (0.5, 0.7, 1 ml L^{-1}) concentration. The

study also revealed mixed use of Alar and Cycocel at 3 g L^{-1} and 1 ml L^{-1} to *Verbena rigida* drastically reduced fresh and dry weight in comparison with non treated controls. However, regardless of mixed application, no reduction in root fresh and dry weight was noted. Reduction was only noticed from individual application of the two growth retardants.

Observation by Banko and Stefani (1988) on *Zinnia elegans* 'Yellow Marvel' revealed reduction of shoot dry weight with treatment of Alar at rate of 5 g L^{-1}. Similarly application of 0.45 g L^{-1} and 0.5 ml L^{-1} bonzi showed significant reduction on shoot fresh weight on *Begonia semperjlorens and* vinca *(Catharanthus roseus),* respectively.

Poole and Ying (1965) studied effect of growth regulators on growth of *Chrysanthemum morifolium* 'bluechip'. From the study they found out that among levels of Cycocel that has been involved in the experiment (0, 5, and 10 ml L^{-1}) t he highest level of Cycocel which is 10 ml L^{-1} reduced both shoot fresh and dry weight.

2.8. Effect of Chemical Growth Retardants on Adventitious Root Formation

Adventitious roots are post-embryonic roots which, differently from lateral roots, arise from the stem and leaves and from non-pericycle tissues in old roots. In these organs, hormones (specially auxin), sugars, temperature, mineral salts, and light conditions may induce groups of cells to redefine their fate, resulting in adventitious rooting (Altamura *et al.*, 2004).

Adventitious root formation is the primary regenerative process required in most cutting propagation. It is the prerequisite to successful cutting propagation. Propagation by stem cuttings requires only that a new root system to be developed, because the potential shoot system is already present (Dole and Gibson, 2006). The formation of adventitious roots is dependent on plant cells to dedifferentiate and develop into a root system. The process of dedifferentiation is the capability of previously developed, dedifferentiated cells to initiate cell divisions and form a new meristematic growing point. Adventitious roots form naturally on the various plant parts (Hartman *et al.,* 2002).

Plant scientists have been interested in the chemical control of adventitious rooting for many years. The first major discovery regarding the chemical control of rooting was that auxins could dramatically promote rooting. Although this finding was of considerable theoretical and practical importance, it soon becomes apparent that auxins did not promote rooting on all types of cuttings and under all circumstances. Hence, it appeared that other factors in addition to auxin were important on rooting. This finding, coupled with an increased understanding of plant growth substances in the past 20 to 25 years seems to have led to the screening of many types of compounds for their ability to promote rooting (Davis *et al*., 1988).

It has been well known that the production of adventitious roots is controlled by growth substances. Auxins are the main hormones for promoting rooting, and play a direct role in this process whereas GA has been widely reported to inhibit adventitious root formation in cuttings of a variety of species. Since plant growth retardants inhibit GA biosynthesis, so decreasing the endogenous GA levels seemed likely that such compounds could enhance adventitious root formation. Although the rationale behind using these compounds has not always been clear, the justification for testing them has usually been that they either 1) antagonize the activity of or synthesis of gibberellins which normally inhibit the rooting, or 2) reduce shoot growth which may compete with the base of the cuttings for assimilates to the detriment of rooting. In some cases the root promoting properties were discovered fortuitously during basic studies on responses of plants to growth retardants (Davis *et al*., 1988; Read and Yang 1991).

Researchers and propagators have long known that plant growth retardant treatments applied to stock plants have a dramatic effect on subsequent propagation efficiency and could favorably influence rooting of cuttings. This is highly probable to, growth retardants can cause modifications in endogenous hormone levels, thus influencing the rooting process of cuttings taken from such stock plants (Read, 1988).

In some studies it was evident that gibberellins, in many physiological processes, had effects opposite to those obtained with auxins. Adventitious root formation, which was stimulated by auxin, was inhibited by gibberellins (Kato, 1958).

According to Read and Yang (1989) sprays of stock plants with growth retardant chemicals Alar and Cycocel have caused an increased propagation potential. In *dahlia*, both chemicals caused improved rooting of cuttings from treated stock plants. Additional studies by Hoysler and Read (1969) illustrated that spraying of 2.5 g L^{-1} Alar on stock plants of *chrysanthemum, dahlia,* and *poinsettia* increased rooting ability of the cuttings. Conflicting observation was made by the same author where Alar sprays caused greater branching, but cuttings taken from Alar treated stock plants rooted more poorly in *Cordyline* and *Poinsettia.* Read and Yang (1989) concluded that it is highly probable that growth retardants can cause modifications in endogenous hormone levels, thus influencing the rooting process of cuttings taken from stock plants.

Indole butyric acid (IBA) is one of the most widely used rooting promoters, but plant growth retardants may also promote the formation of adventitious roots. For example, rooting of *Petunia* is affected by Bonzi and Alar, but the magnitude of effect of the growth retardants was less than that of IBA. Since growth retardants inhibit the biosynthesis of GA, it was thought that IBA and growth retardants could have a synergistic effect on rooting when applied simultaneously or sequentially (Pan and Zhao, 1994).

Alar is a growth retardant mostly used on a number of herbaceous ornamental species and has been reported to be a strong promoter of adventitious rooting in several species. According to Read and Hoyser (1971) 1 to 5 g L^{-1} spray of Alar on stock plants of several ornamental species resulted in increased root number, weight and length of the cuttings. Also, cuttings from treated plants rooted faster than the controls. In contrast to these finding, Beck and Sink (1974) reported that Alar had no effect on rooting of *Euphorbia spp*. cuttings up to concentration of 2.5 g L^{-1}. From these variable effects, it may be said that Alar induced effects on rooting are species dependent (Davis *et al*., 1988).

Cycocel is used extensively throughout the world to control shoot elongation on many ornamental crops. The effect of Cycocel treatment on rooting has been somewhat variable. Most investigators have reported that Cycocel treatment promoted rooting but others have reported no effect or inhibition. These discrepancies might be explained by differences in application methods and variable responses from different plant species (Davis *et al*., 1988).

3. MATERIALS AND METHODS

3.1 Description of the Study Site

The study was conducted at Florensis Ethiopia P.L.C. from November 2009 to February 2010 under greenhouse condition. Florensis Ethiopia is located at Koka town, South-Central Ethiopia 110 km away from Addis Ababa. Geographically, the area is situated at 8^{o}, 26'N latitude and 39^{o}, 02' E longitude at an altitude of 1595 meters above sea level (m.a.s.l.). The mean annual rainfall is 700 mm while the mean minimum and maximum temperatures are 10.4 and 29.7 ^{0}C, respectively (Wikipedia, 2009). In the greenhouse the temperature and relative humidity were kept in the range of 18 to 30^{o}C and 55 to 70%, respectively and in the propagation unit temperature and relative humidity were maintained in the range of 24 to 35^{o}C and 75 to 85%, respectively using a computerized system (Priva greenhouse systems) found at the farm (Appendix fig. 1 and 2).

3.2 Experimental Materials

Rooted cuttings of Verbena *(Verbena hybrida*) cultivar Vegas Scarlet were used for the study to develop into stock plant. The breeder of this cultivar is Florensis breeding department which is located in Quedlinburg, Germany. Vegas Scarlet is well known for its vigorous growth and selected for this experiment because of the following main reasons i) it is popular in the international trade because of its red flower color ii) it is widely grown at Florensis Ethiopia plc and is one of the crops which cover up the majority of the export volume of the company, and iii) because of its vigorous growth difficult to get sufficient compact cutting.

The two plant growth retardants used for the experiment were Cycocel (BASF Asia Pacific Pvt. Ltd) and Alar (Chemtura Chemical Co.). Red ash was used as media for growing the stock plants and the cuttings which were taken from stock plants were sticked in to tray plugs filled with peat moss for rooting. Red ash and peat moss were selected because the farm practically uses these medias for production of *Verbenas* and the other crops. The important feature of red ash is it has more pore space for aeration and good ability to absorb water meanwhile peat moss

is known for its excellent water retention and high cation exchange capacity. Hydra foam is incorporated with peat moss to complement its moderate aeration activity because of its fine structure. All the necessary materials for the experiment were supplied by Florensis Ethiopia plc.

3.3 Experimental Design and Treatments

The experiment was executed in two phases. The first phase of the experiment was laid out in a 4x4 factorial arrangement with a complete randomized design (CRD) consisting of three replications. The treatments consisted of two different growth retardants as two factors (each with four different levels). The two growth retardants used for the study were Alar (daminozide) at concentrations of 0g/L, 1g/L, 2g/L, and 3g/L (**A_0**, **A_1**, **A_2**, and **A_3**, respectively) and**,** Cycocel (chlormequat chloride) at concentrations of 0ml/L, 0.5ml/L, 1ml/L, and 1.5ml/ L (**C_0**, **C_1**, **C_2**, and **C_3**, respectively). In total, there were sixteen treatment combinations, which were randomly assigned to the experimental plots. The concentration levels for both Alar and Cycocel were based on the farms actual practice and previous small scale trials made at the farm as per the recommendations of the products.

In first phase of the experiment, rooted cuttings of the respective cultivar were planted on round plastic pots having a capacity of 1.5litres filled with red ash as a growing media. Each pot accommodated only one cutting and there were sixteen pots per treatment per replication and 48 potted cuttings for a single treatment. Hence, the total number of potted cuttings for the whole treatment was seven hundred sixty eight (768). The experimental pots in each replication were arranged close to each other but with 30cm gap between each plot.

When the rooted cuttings developed sufficient foliage and when the leaves fully expanded to the edge of the pot (Six weeks after planting), the potted plants were sprayed with the randomly assigned treatments. Spraying of the treatments was done using hand sprayers. Each plot was sprayed uniformly with respective treatments until the foliage of the plants became sufficiently wet. The spray of stock plants with plant growth retardants was done early in the morning at a week interval for eight consecutive weeks. Glady *et al*,(2004) stated that weekly application of plant growth retardants can be adequate to manage vegetative stock plants for

cutting propagation. Application was done in the morning merely because of cooler condition in the morning which is reportedly know to increase the effectiveness of Cycocel and Alar sprays due to slow evaporation and full turgidity of plants (Cox, 2007). Apart from this, early morning application generates effective height control since a large percentage of the daily stem elongation occurs early in the day just after sunrise (Dole and Wilkins, 2005).

Other management practices and follow-ups were implemented uniformly to all the stock plants as per the operational procedures of the farm at Florensis Ethiopia plc. Fertilizers were applied through fertigation using the microtube (spaghetti) system based on the recommended rate of the farm. Data regarding the growth of the stock plants were taken at weekly interval for eight consecutive weeks starting a week after the first application of the treatments. On the other hand, data such as shoot fresh and dry weight and root fresh and dry weight collected only once (at the end of the 8^{th} week) after uprooting the sampled plants.

For the second phase of the experiment, after taking the last data on stock plants, twenty cuttings having one pair of leaves and length of 2 cm were collected from each plot. The size of the cuttings was determined on the basis of the farm export standard. The cuttings were subsequently rooted under mist in a propagation house using ‘‘winstrip’’ (rooting flat tray) filled with peat moss. The rooting of cuttings was done with the same design and layout as that of phase one of the experiment. All the cuttings received the regular management practices as per the operational procedures of the farm. When all the cuttings formed well developed shoots with three pairs of leaves, data pertaining to different parameters were taken to evaluate the rooting performance of the cuttings.

3.4. Data Collected

3.4.1 First phase experiment

The following parameters were recorded and analyzed from randomly sampled six stock plants.

Internode length | cm |

The distance between nodes of the main stem of stock plants was measured using a ruler and the average value was recorded.

Main stem length | cm |

Main stem length of stock plants was measured using a ruler from the crown (the point where the root and stem meet) to the uppermost point of the stem.

Stem diameter | mm |

The stem diameter of the main stem was measured from the base of stock plants using standard (digital) Verneir Caliper. Measurement was taken 5cm above from the surface of the media.

Number of main branches

Number of only main branches on the main stem of stock plants was counted.

Average leaf area |cm^2|

Leaf area was measured and averaged by arbitrarily taking ten leaves from top & medium positions of the stock plants. Measurement was taken using square paper from intact leaves without detaching from the stock plants.

Canopy diameter |cm|

Canopy diameter or width of stock plants was measured at the widest point using hand meter. Measurement was done from both North to South and East to West directions and the average value was taken.

Root fresh weight |g|

Root fresh weight of stock plants was measured using digital sensitive balance (EX-2000, maximum precision level of 0.1mg) after uprooting and thorough washing of the roots.

Root dry weight |g|

The measured roots for fresh weight were placed into an oven (70^{o}C) for 24 hour for drying to a constant weight and then the dried roots were weighted using digital sensitive balance (EX-2000, maximum precision level of 0.1mg).

Shoot fresh weight |g|

Shoot fresh weight (above ground portion excluding only the roots) of stock plants was measured using digital sensitive balance (EX-2000, maximum precision level of 0.1mg).

Shoot dry weight |g|

After taking the fresh weight of the shoots, the samples were subjected for drying to a constant weight using an oven (70^{o}C) for 24 hour then shoot dry weight was measured using digital sensitive balance (EX-2000, maximum precision level of 0.1mg).

Number of cuttings

After taking fresh and dry weights of the randomly assigned plants, from the remained ten stock plants in each experimental plot, total number of available cuttings was taken four times in a week interval. The first picking of cuttings was done one week after the last application of the treatments. For analysis, the total number of cuttings of the month was taken into consideration.

3.4.2 Second phase experiment

The following parameters were recorded and analyzed from randomly sampled ten rooted cuttings.

Number of roots per cutting

Total number of adventitious roots per cutting of cuttings was counted.

Root fresh weight |g|

Root fresh weight of the cuttings was measured using digital sensitive balance (EX-2000, maximum precision level of 0.1mg).

Root dry weight |g|

The measured roots for fresh weight were placed into an oven (70^{o}C) for 24hrs of drying and then root dry weight was recorded when the weight was constant using digital sensitive balance (EX-2000, maximum precision level of 0.1mg).

Mean Root length |cm|

Root length of the cuttings was measured from the crown of the plant to the final tip of the roots. Measurement was taken from all developed roots and averaged.

Mean percentage of the rooted cuttings |%|

The amount of cuttings rooted from the total cuttings planted was determined by calculating using percentage. The following formula was used to for determining the percentage.

$$PR = \frac{\text{Number of cuttings rooted}}{\text{Total number of cuttings planted}} \times 100$$

Root volume |cm^3|

The average root volume was measured by immersing the roots of each cutting in a beaker containing 1000ml of water. The volume of the root was determined by observing the displacement of the water by the root, so that the difference was taken as the volume of the root**.**

3.5. Statistical Analysis

The data of all parameters considered in the study were subjected to the Analysis of Variance (ANOVA) using SAS version 9.2. computer software (SAS Institute Inc., 1999) after the data were checked for meeting the various ANOVA assumptions (Montgomery, 2005). The model used for two factor analysis of variance was:

$$\gamma_{ij}=\mu+\tau_i+\beta_j+(\tau\beta)_{ij}+\varepsilon_{ij}$$

i= 1, 2, 3,…a

j= 1, 2, 3,….b

Where,

- μ = is the overall mean effect,
- τ_i = is the effect of the i^{th} level of factor A (Alar),
- β_j = is the effect of the j^{th} level of factor B (Cycocel)
- $(\tau\beta)_{ij}$ = is the effect of the interaction between τ_i and β_j
- ε_{ijk} = is a random error component.

LSD procedures at 0.05 probability level of significance were used to determine differences between treatment means whenever the treatment effects were found to be significant and the bivariate correlation between response variables were also determined using the same software program.

4. RESULTS AND DISCUSSION

The results of the investigation on the influence of Alar and Cycocel on the growth of stock plants and subsequent rooting of cuttings of *Verbena hybrid*a cultivar Vegas Scarlet are presented and discussed in this particular chapter.

4.1 Effect of Cycocel and Alar on Growth of Stock Plants

4.1.1 Internode length

As indicated in Table 1 there was a highly significant (P<0.0001) interaction effect among different concentrations of Alar and Cycocel on internode length of stock plants.

Table 1. P Values for ANOVA for internode length, stem length, stem diameter, number of main branches, average leaf area and canopy diameter as influenced by Alar and Cycocel and their interaction

Source of Variation	DF	IL	SL	SD	NB	LA	CD
Alar	3	<.0001	<.0001	0.0149	0.0139	<.0001	<.0001
Cycocel	3	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001
Alar * Cycocel	9	<.0001	<.0001	0.9865^{ns}	0.0028	0.3638^{ns}	0.0387
CV (%)		1.78	2.96	1.86	5.65	4.50	1.50

^{ns}=non significant different; DF=Degree of Freedom; IL=Internode length; SL=stem length; NB=number of main branches; LA= average leaf area; CD=canopy diameter; CV=Coefficient of Variation.

As shown in Fig.1 (Appendix Table 11) the longest internode length (7.56cm) was recorded from non treated stock plants. On the other hand, the shortest internode length (3.88cm) was observed from the combined application of 1.5 ml L^{-1} CCC and 3 g^{-1} L Alar. Furthermore, an increase in either of the growth retardants decreased the mean length of stem internode of the treated stock plants.

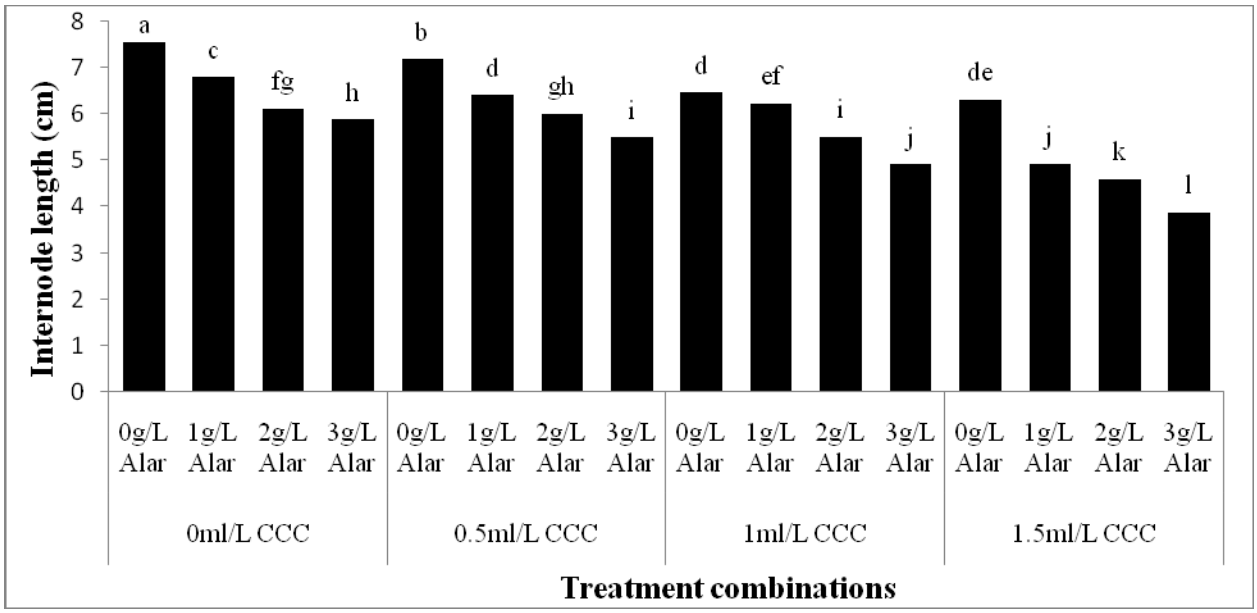

Fig. 1 Interaction effect of Cycocel and Alar on internode length.

The findings of this study was in line with the effect already seen in *Verbena hybrida* 'Obsession Lilac' (Blanchard *et al.*, 2008) and *Verbena rigida (*Andersen and Davis, 1989a) from combination of Cycocel and Alar, *Zinnia elegans (*Barbosa *et al.,* 2005) from individual application of Alar and Cycocel where decline in internode length was observed. The reduction of internode length suggests that the activity of subapical meristematic area in the stem, which is responsible for internode elongation, is influenced. Internode elongation is based on two cellular processes: cell division (based on cell number) and cell expansion or elongation which are mainly driven by gibberellins. Gibberellins are strongly influenced by growth retardants (Cathey, 1964; Rademacher, 2000; Puglisi, 2002;). Since Cycocel and Alar are antagonistic to gibberellins (GAs), the result obtained may also be attributed to the reduced level of gibbrerelins. Erwin and Warner (2003), Blanchard *et al.* (2008), and Carvalho (2008) also forwarded similar explanations for the occurrence of reduced internode length.

To better understand the elongation process, cell number and cell length were recorded in fully developed internodes of genus *Lilium* and *Campanula* grown under different concentrations of plant growth retardants by Carvalho *et al*. (2008). The study demonstrated that plants with higher concentrations had reduced stem elongation due to decreased cellular elongation as a result of both smaller cell length and cell width. On the other hand, Grossman (1990) was able

to demonstrate the number of mitotic figures on stems of *chrysanthemum* that, after treatment with growth retardants, the cell division activity in the subapical meristems was diminished which can support the reduction of internode length is due to cell division.

4.1.2 Main stem length

This study revealed that the interaction effect between Cycocel and Alar was highly significant ($P<0.0001$) on main stem length (Table 1). A similar scenario was observed with that of internode length where treatment combination of higher concentration of Cycocel and Alar produced the minimum main stem length (10.15cm) while non treated stock plants resulted in the maximum main stem length (21.35cm)(Fig. 2 or Appendix Table 11).

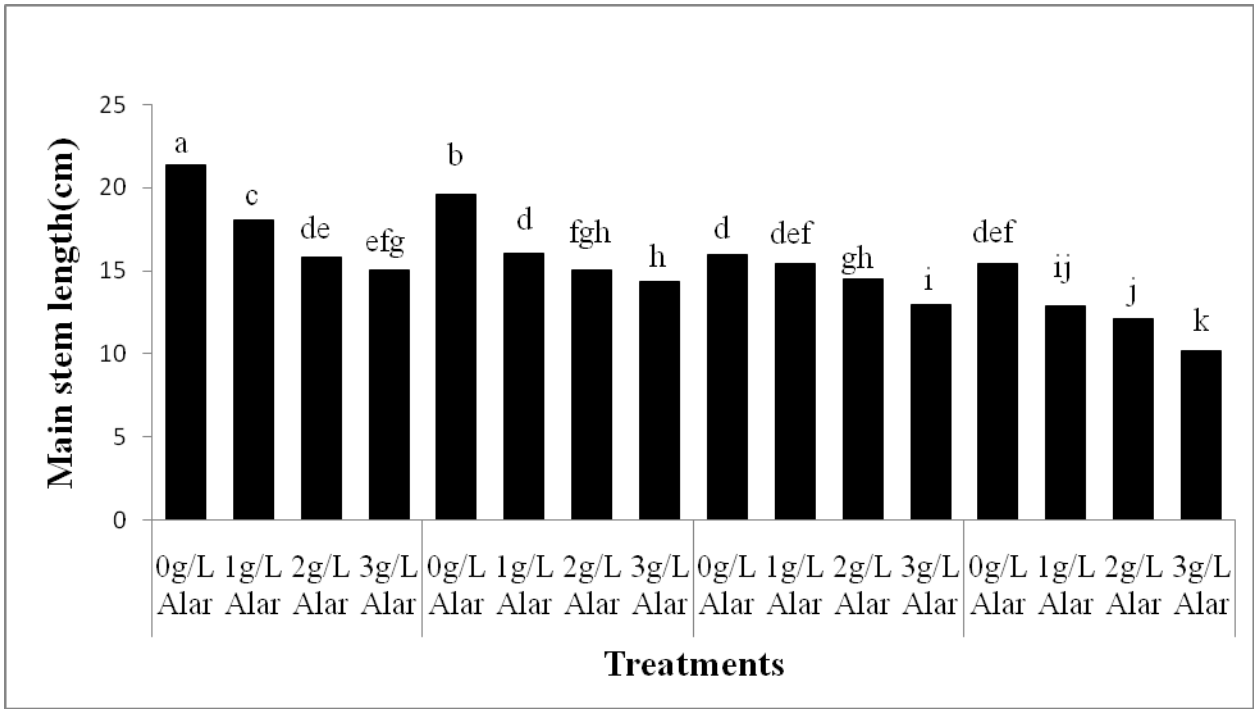

Fig. 2 Interaction effect of Cycocel and Alar on main stem length

Similar results were reported on *Verbena canadensis* (Burnett *et al*., 2000) and *Verbena* 'Obsession Lilac' (Blanchard *et al.,* 2008) and *Verbena rigida (*Andersen and Davis, 1989a) using combined application of Alar and Cycocel. Several authors have pointed out such an effect in other bedding or ornamental plants including *Poinsettia* and *Pansy* (James *et al.*,

2002), and *Veronica* 'Sunny Border Blue', *Sedum* 'Autumn Joy', *Monarda didyma* 'Marshall's Delight', and *Phlox paniculata* 'David' (Baden *et al.,* 1999) with Alar and Cycocel mix application. Karlovic *et al.* (2004) also obtained similar result on *Chrysanthemums* by applying only Alar.

The observed reduction in plant height might be due to anti-gibberellins activity of Alar and Cycocel which facilitates inhibition of cell division frequency and cell elongation in the sub apical meristematic zone of the stem. This fact is in conformity with James *et al.* (2002), Banko and Stefani (1988) and Barbosa *et al.* (2005).

According to Basra (2000), shorter stems have been related to decreased cell number, short cortical cells, and reduced xylem length. These may result from the combined effect of the two factors. Less height increase in the treated plants might also be due to reductions in the internode elongation. As internode length shows certain decline, stem length is also expected to decrease (Barrett and Nell, 1983).

4.1.3 Stem diameter

A highly significant ($P<0.01$) differences were observed among the different concentrations of Cycocel treatments in relation to stem diameter (Table 1). Regarding Alar, significant variation ($P<0.05$) was among the different concentrations. In contrast, the interaction effect between Cycocel and Alar was not statistically significant ($P>0.05$) in respect of stem diameter.

The result in Table 2 indicated that stem diameter was directly proportional to the concentration of both Cycocel and Alar. With higher concentrations, higher stem diameter was obtained. Among concentrations of Cycocel, application of 1.5 ml L^{-1} produced significantly the maximum stem diameter (1.45mm) which however was at par with treatment of stock plants with 1ml L^{-1} (1.43 mm). Stock plants with no application of Cycocel (0 ml L^{-1}), on the other hand, resulted in significantly lower stem diameter (1.27mm) followed by application of 0.5 ml L^{-1} (1.36mm). In case of Alar, application of 3 g L^{-1} gave significantly higher stem diameter (1.40 cm) nevertheless it was not significantly different from application of 2 g L^{-1}. Significantly

the least stem diameter (1.37 mm) was observed from non treated stock plants (0 g L^{-1}) and 1 g L^{-1} which were again comparable with application of 2 g L^{-1}.

Table2. Effect of Cycocel and Alar on stem diameter

Factors and Levels	N	Stem diameter (mm)
Alar		
0 g L^{-1}	12	1.37^{b}
1 g L^{-1}	12	1.37^{b}
2 g L^{-1}	12	1.38^{ab}
3 g L^{-1}	12	1.40^{a}
LSD		0.02
SE(±)		0.007
CV (%)		1.86
Cycocel		
0 ml L^{-1}	12	1.27^{c}
0.5 ml L^{-1}	12	1.36^{b}
1ml L^{-1}	12	1.43^{a}
1.5ml L^{-1}	12	1.45^{a}
LSD		0.02
SE(±)		0.007
CV (%)		1.86

Means followed by different letters are significantly different at the 5 % level of probability; ml=milliliter, g=gram; SE=Standard Error; CV=Coefficient of variation

An increase in stem diameter due to the influence of Alar and Cycocel agreed with the results obtained by Barras-Ali *et al.* (2007) in chrysanthemum using Alar. The result achieved may be due to the facts that as plants have limited vertical growth they tend to store more food or carbohydrate in their stem, because they use less energy for upward growth. Since the different concentrations of Cycocel and Alar had brought reduced stem length, the plants h ave resulted in higher stem diameter. According to Cathey (1964), the increasing effect of Alar and Cycocel could be due to the stimulation of cell production in the cambium, accompanied by a delay in cell differentiation, and to an increase in cell volume of the parenchymatous cortical cells. Barras-Ali *et al.* (2007) justified that increase in stem diameter might be due to transverse cell expansion and division in the sub apical tissues which deviates from the custom orientation of plants cells during cell expansion.

4.1.4 Number of main branches

Branch number is a major consideration in growing of stock plants for the purpose of producing more number of cuttings. More cuttings are expected from stock plants having more number of branches. In this study, a highly significant ($P<0.01$) interaction effect was observed among the different concentrations of Cycocel and Alar (Table 1).

The mean comparison of the treatment combinations revealed a substantial influence on the number of main branches (Fig.3 or Appendix Table 11). Accordingly, significantly maximum number of branches (14.35) was observed from treatment combination of 1 ml L^{-1} CCC and 2 g L^{-1} Alar, which however was not significantly different from combined application of 1 ml L^{-1} CCC and 1 gL^{-1} Alar. Conversely, the minimum number of main branches was observed from stock plants without treatment (5.73) which however was at par with treatment of 1g L^{-1} Alar X 1ml L^{-1} CCC and 3g L^{-1} Alar X 1ml L^{-1} CCC. Number of branches exhibited a trend of rising with increasing levels of combined application of the two growth retardants, but this rise started to show some decline beyond the treatment combination of 1 ml L^{-1} CCC X 2g L^{-1} Alar.

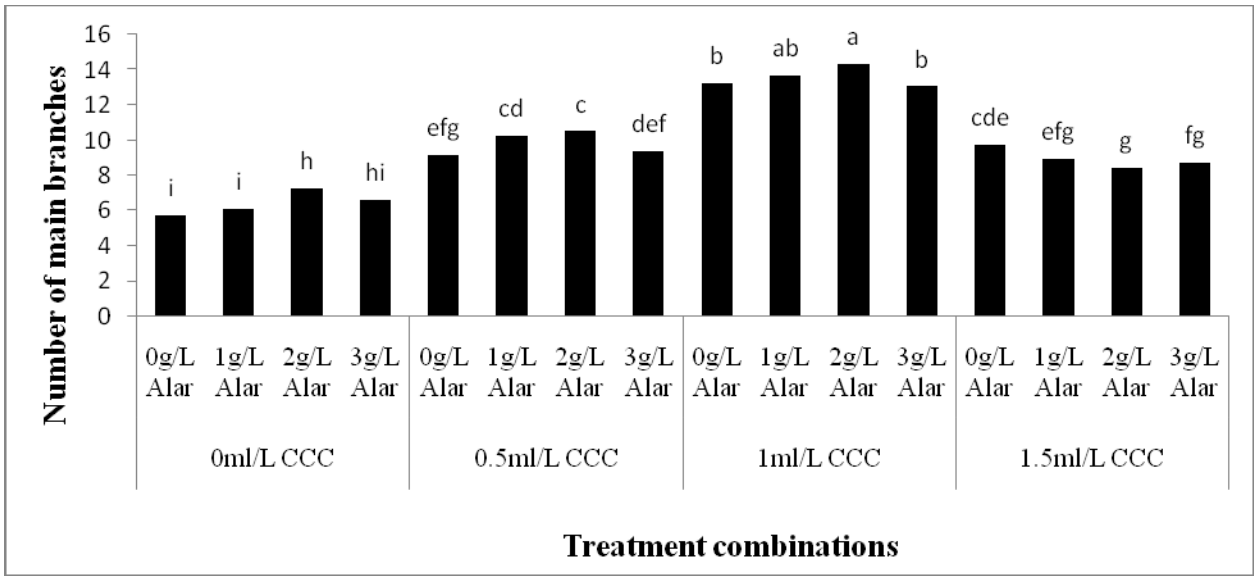

Fig. 3 Interaction effect of Cycocel and Alar on number of main branches.

This finding is in line with the observations on *Rosa damescena* (Abbas *et al.,* 2007) and *Hebe fransiscana* (Adriansen and Kristensen, 1988) from individual application of Alar and Cycocel and on *Tagetes patula, Impatiens walleriana,* and *Petunia hybrid using* only Alar. The increase

in the number of main branches per plant as a result of the combined application of Alar and Cycocel might be attributed to the synergetic effects of the two growth retardants in checking the apical dominance through reduced levels of endogenous production of auxins which in turn induced the sprouting of vegetative buds. Plant growth retardants work by interrupting apical dominance, which triggers lateral buds to grow and fill in the plant. In apical dominance, the shoot apex can prevent lateral bud growth. Such possible explanation was also forwarded by other workers (Abbas *et al*., 2007; Amarander, 2007; Carey, 2008;).

4.1.5 Average leaf area

Leaf area is a determinant factor in radiation interception, photosynthesis, biomass accumulation, transpiration and energy transfer by crop canopies. Therefore, leaf area is measured in many different studies and its accurate measurement is necessary for understanding crop responses to experimental treatments (Akram, and Soltani, 2007).

In the current study, highly significant differences were observed among the different concentrations of Cycocel ($P<0.0001$) and Alar ($P<0.0001$) in relation with average leaf area (Table 1). Conversely, the interaction between Cycocel and Alar was found non-significant ($P>0.05$).

The result on Table 3 confirmed average leaf area was indirectly proportional to concentrations of both Cycocel and Alar. There was a decline in leaf area as the concentration of Alar increased from 0 g L^{-1} (8.26 cm^2) to 3 g L^{-1} (6.97 cm^2). However 2 g L^{-1} (7.22 cm^2) and 3 g L^{-1} (6.97 cm^2) were not significantly different from each other. Likewise, a decline in leaf area was noticed as the concentration of Cycocel increased from 0 ml L^{-1} (8.16 cm^2) to 1.5 ml L^{-1} (6.83 cm^2).

Table3. Effect of Cycocel and Alar on average leaf area.

Factors and Levels	N	Average leaf area (cm^2)
Alar		
0 g L^{-1}	12	8.26[a]
1 g L^{-1}	12	7.70[b]
2 g L^{-1}	12	7.22[c]
3 g L^{-1}	12	6.97[c]
LSD		0.28
SE(±)		0.10
CV (%)		4.50
Cycocel		
0 ml L^{-1}	12	8.16[a]
0.5 ml L^{-1}	12	7.85[b]
1ml L^{-1}	12	7.32[c]
1.5ml L^{-1}	12	6.83[d]
LSD		0.28
SE(±)		0.10
CV (%)		4.50

Means followed by different letters are significantly different at the 5 % level of probability; ml=milliliter; g=gram; SE=Standard Error; CV=Coefficient of variation

A similar result was reported by Gibson and Whipker (2004) from application of Cycocel on *Fuchsia* x *hybrid, Pelargonium* x *hortorum,* and *Lantana hybrid* as Barras-Ali *et al.* (2008) did on *Chrysanthemum morifolium* from application of Alar. In contrary, Amarender and Veena (2007) observed gradual increase in leaf area after application of Alar and Cycocel. The result obtained from this investigation may probably be attributed to the inhibiting effect of Cycocel and Alar on gibberellins biosynthesis. In line with this context, Grossman (1990) and White (2003) pointed out the role of gibberellins in regulating longitudinal shoot and leaf growth.

4.1.6 Canopy diameter

An essential aspect of any crop production system is the development of a crop canopy that optimizes the interception of light, photosynthesis, and the allocation of dry matter to harvestable parts**.** The present experiment indicated that the interaction among the different

concentrations of Cycocel and Alar has imparted a significant (P<0.05) difference on canopy diameter of the treated stock plants (Table 1).

As indicated in Fig.4 (Appendix Table 11) the unreated stock plants produced the higher canopy diameter (60.33 cm) which still was not significantly different from the treatments of 0.5 ml L^{-1} CCC X 1 g L^{-1}Alar (59.39cm) and 0.5 ml L^{-1} CCC X 0 g L^{-1}Alar (59.20cm). On the other hand, significantly the lower canopy diameter was observed from application of 1.5 ml L^{-1} CCC X 3 g L^{-1} Alar (48.44cm).

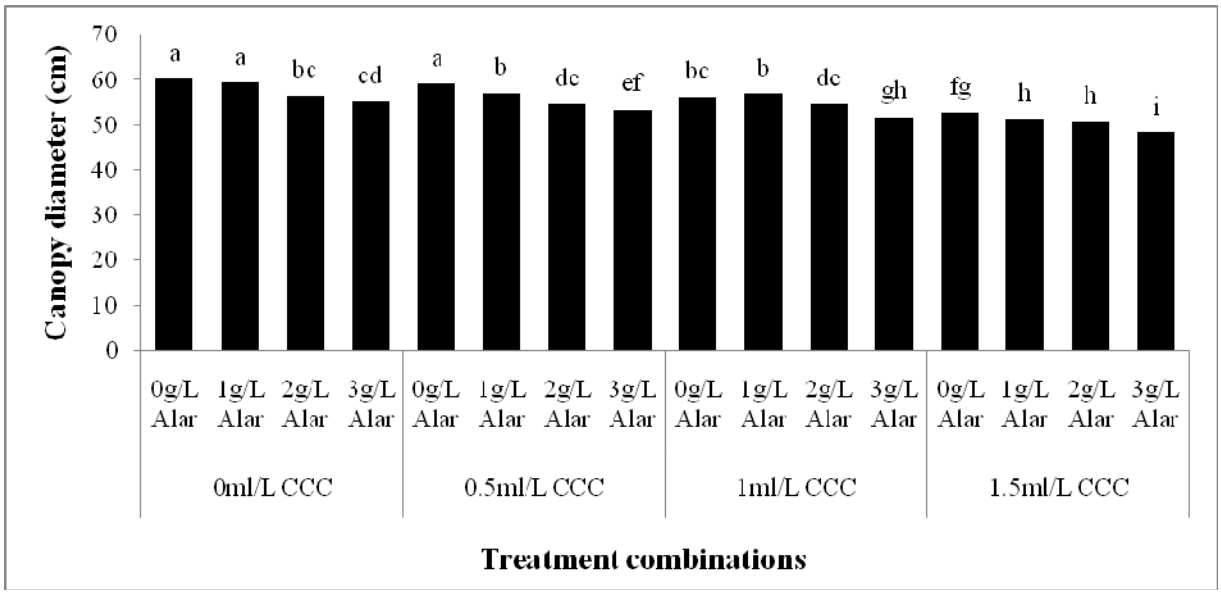

Fig. 4 Interaction effect of Cycocel and Alar on canopy diameter.

This finding is in agreement with observations reported on *Verbena canadensis* (Burnett *et al*., 2000) from combined application of Cycocel and Alar, and also Alar application *on Zinnia elegans* (Banko and Stefani, 1988). The apparent results were probably due to the dwarfing effect of Cycocel and Alar, reducing both plant height and width. Comparable justification is also made by Carvalho *et al*. (2008).

4.1.7 Root fresh weight

A highly significant (P<0.01) interaction effect among the different concentrations of Alar and Cycocel was observed on root fresh weight (Table 4).

Table 4. P Values for ANOVA for root fresh weight, root dry weight, shoot fresh weight, shoot dry weight, and number of main branches as influenced by Alar and Cycocel and their interaction

Source of Variation	DF	RFW	RDW	SFW	SDW	NC
Alar	3	0.0252	0.0259	<.0001	<.0001	0.0403
Cycocel	3	<.0001	<.0001	<.0001	<.0001	<.0001
Alar * Cycocel	9	0.0015	0.0264	0.0119	0.0005	0.4623^{ns}
CV (%)		1.27	2.91	1.24	2.24	1.25

ns=non significantly different DF=Degree of Freedom; RFW=root fresh weight; RDW=root dry weight; SFW=shoot fresh weight; SDW=shoot dry weight; NC=number of cuttings; CV=Coefficient of Variation

As indicated in Fig.5 (Appendix Table 11) the maximum root fresh weight (82.01g) was obtained from application of 1ml L^{-1} CCC X 0g L^{-1}Alar. However, this result was insignificant with the effect of applying 1.5ml L^{-1} CCC X 0g L^{-1} Alar (81.70g) and 0.5ml L^{-1} CCC X 0g L^{-1} Alar (81.20g). Whereas, significantly the minimum root fresh weight (72.86g) was obtained from combined application of 1.5 ml L^{-1} CCC and 3 g L^{-1} Alar, which was still not significantly different from 0.5 ml L^{-1} CCC X 3 g L^{-1} Alar (73.17g), 1ml L^{-1} CCC X 2g L^{-1} Alar (73.74g), 1ml L^{-1} CCC X 3g L^{-1} Alar (73.04g) and 1.5ml L^{-1} CCC X 2g L^{-1} Alar (73.35g).

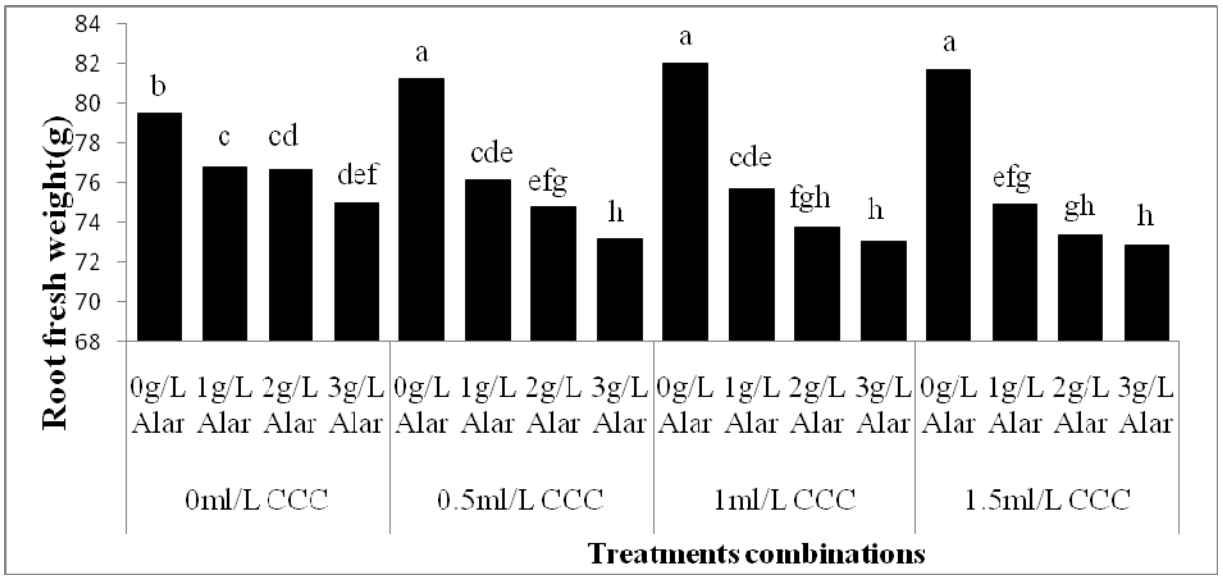

Fig. 5 Interaction effect of Cycocel and Alar on root fresh weight.

These results are found to be in compliance with findings reported on *Dombeya burgessiae* (Laubscher *et al.,* 2010), genus *Salvia* (Latimer *et al*., 1999) from mixed use of Alar and Cycocel, and *Verbena rigida* (Andersen and Davis, 1989a) from individual application. In all these cases, the authors have indicated the reduction of root fresh weight due to the effect of different growth retardants. According to Dalbro and Jindal (1977) plant growth retardants can modify endogenous auxin level in treated plants. Such effect can have a significant role in root development of treated plants. The interaction between Cycocel and Alar could influence the level of auxin that brings limited root growth. According to Cathey (1964) applications of growth retardants measurably inhibited root formation or delayed root development of chrysanthemum with application of Cycocel and Alar. As a result, the obtained reduction in root fresh weight could be related to the limited root growth.

Despite the above mentioned explanations, contradictory reports have also been mentioned. For instance, according to Latimer (1991) and Grossman (1990) root growth is less affected, or slightly promoted with main roots often longer and thicker by growth retardants application. Such discrepancies in respect of the effect of growth retardants on root fresh weight might arise from the concentration, type, frequency and time application of growth retardants.

4.1.8 Root dry weight

As depicted in Table 4, the effect of the interaction among the different concentrations of Cycocel and Alar on root dry weight was found to be significant ($P<0.05$).

The maximum root dry weight (16.17g) was obtained from the application of 1.5ml L^{-1} CCC X 0g L^{-1} Alar. But it was still insignificant with stock plants without treatment (15.94g), 0.5ml L^{-1} CCC X 0g L^{-1} Alar (16.01g), and 1ml L^{-1} CCC X 0g L^{-1} Alar (15.91g). On the other hand, the minimum value was observed from combined application of 1.5 ml L^{-1} CCC and 3 g L^{-1} Alar (12.39 g) which however was not significantly different from combined treatments 1.5ml L^{-1} CCC X 2g L^{-1} Alar (12.97g), and 1ml L^{-1} CCC X 3g L^{-1} Alar (12.84g) (Fig.6 or Appendix Table 11).

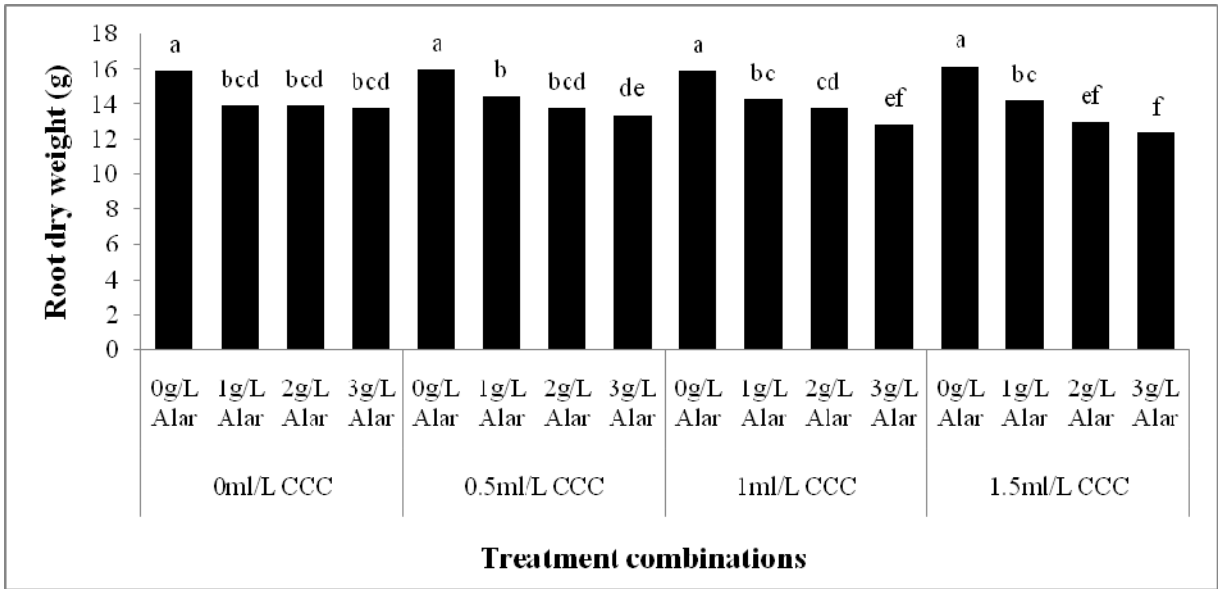

Fig. 6 Interaction effect of Cycocel and Alar on root dry weight.

The reduction of root dry weight due to the different combination levels of Cycocel and Alar has already been noted in genus *Salvia greggi* and *Salvia leucantha* (Latimer *et al*., 1999) and *Dombeya burgessiae* (Laubscher *et al.,* 2010). The reduction in dry weight with increasing levels of CCC and Alar can be related to the limited growth of root system and also with limited production of carbohydrate because of reduced leaf area with increasing level of the treatments.

4.1.9 Shoot fresh weight

According to the present study a significant interaction effect of Cycocel and Alar was observed for shoot fresh weight ($P<0.05$; Table 4).

As depicted in Fig.7 (Appendix Table 11) the maximum shoot fresh weight (229.80g) was recorded from the non treated stock plants. In contrast, the minimum shoot fresh weight (196.74g) was observed from the combined application of 1.5ml L^{-1} CCC and 3g L^{-1} Alar which nevertheless was not significantly different from the application of 1.5ml L^{-1} CCC X 2g L^{-1} Alar (198.48g) and 1.5ml L^{-1} CCC X 1g L^{-1} Alar (200.44g). The results of this study confirmed that shoot fresh weight was inversely proportional to the concentration of both Cycocel and Alar. Each additional amount of Alar and Cycocel applied resulted in an additional reduction of shoot fresh weight.

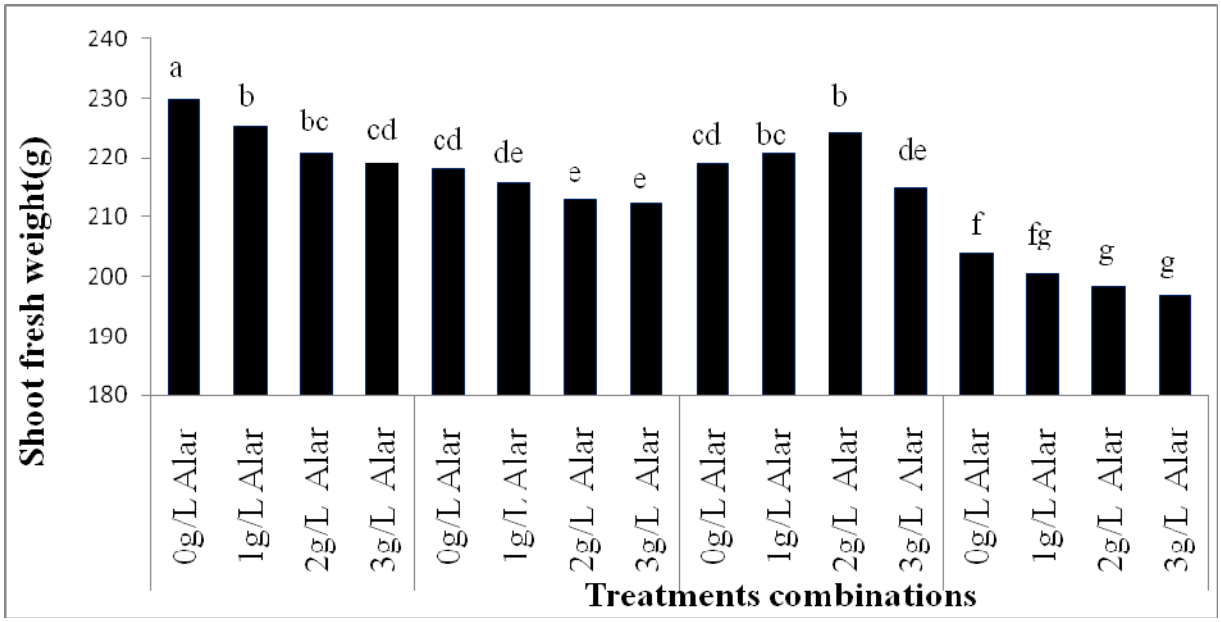

Fig. 7 Interaction effect of Cycocel and Alar on shoot fresh weight.

The current finding is in agreement with the previous reports of Andersen and Davis (1989a) on *Verbena rigida* by Alar and Cycocel combined application, Laubscher *et al.* (2002) on *Dombeya burgessiae* and Poole and Ying (1965) on *Chrysanthemum morifolium* after application of Cycocel. The observed reduction in fresh weight was probably due to the synergic effect of the two growth retardants in causing dwarfness by reducing plant height and width.

4.1.10 Shoot dry weight

There was a highly significant ($P<0.01$) interaction effect between different concentrations of Alar and Cycocel on shoot dry weight (Table 4).

Fig.8 (Appendix Table 11) showed that the untreated stock plants attained significant superiority from the rest of the treatments by exhibiting the maximum shoot dry weight (57.84g). In contrast, the minimum shoot dry weight was obtained from the combination of 1.5ml L^{-1} CCC and 3g L^{-1} Alar (35.81g) which yet was insignificant with 1.5ml L^{-1} CCC X 2g L^{-1} Alar (36.87g) and 1.5 ml L^{-1} CCC X 2 g L^{-1} Alar (37.15 g) (Table 12).

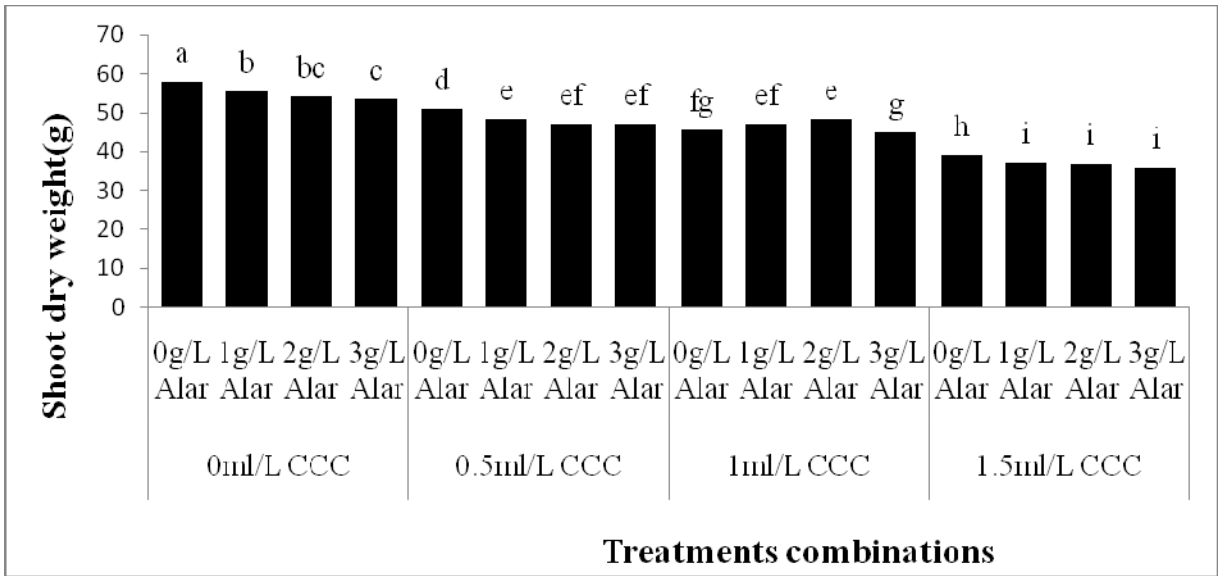

Fig. 8 Interaction effect of Cycocel and Alar on shoot dry weight.

The reduction in shoot dry weight obtained by Cycocel and Alar agreed with the results obtained on *Verbena rigida* by Alar and Cycocel combined application (Andersen and Davis, 1989a), *Zinnia elegans* with a treatment of Alar (Banko and Stefani, 1988), and *Chrysanthemum morifolium* by Cycocel (Poole and Ying, 1965). The observed variation in dry weight can be associated to less biomass accumulation in the plant tissue. This can be attributed to the reduction in average leaf area because of increasing concentration of Cycocel and Alar. Plants with higher leaf area are expected to have more vigorous growth since they can absorb more sunlight which can promote the process of photosynthesis than those having less leaf area (Akram and Soltani, 2007). Hence, with lower Average leaf area the plants are expected to intercept less solar energy which leads to limited or reduced production of carbohydrate. After all the plants will manage less biomass accumulations which will favour reduction in shoot dry weight.

4.1.11 Number of cuttings

The sustainability of cutting producing farms depends upon the total volume of cuttings produced. The yield of cuttings is very crucial to ensure the profitability of the floriculture business. From the conducted experiment, highly significant ($P<0.0001$) difference was

observed among the different concentrations of Cycocel. Similarly, a significant ($P<0.05$) variation was noticed from the spray of different concentrations of Alar. On the contrary, there was no significant ($P>0.05$) interaction effect between Cycocel and Alar in respect of the number of cuttings produced per stock plant (Table 4).

The results in Table 5 revealed among concentrations of Cycocel significantly maximum number of cuttings was obtained from stock plants treated with 1ml L^{-1} CCC (75.0) followed by 1.5 ml L^{-1} CCC (69.2). On the other hand, stock plants with no application of CCC (0ml L^{-1}) exhibited significantly the minimum number of cuttings (66.2). Concerning Alar, significantly the maximum number of cuttings acquired from 2g L^{-1} (70.4) which however was at par with 1g L^{-1} (69.9). Meanwhile the minimum number of cuttings (69.4) was obtained from treatments without Alar (0g L^{-1}). Even though the number of cuttings showed a trend of increment with increasing concentration of both retardants, no further rise was achieved with rates greater than 1 ml L^{-1} CCC and 2 g L^{-1} Alar.

Glady *et al.* (2004) reported increment in the number of cuttings in *Salvia nemorosa, Coreopsis, verticillata* and *Veronica spicata* using Cycocel as did Carpenter and Carlson (1972) in *Geranium*. The observed variation in the number of cuttings could be as the result of more branching response of the stock plants from Alar and CCC treatment. As the number of branches increase, a rise in the number of cuttings would be obvious.

Table 5. Effect of Cycocel and Alar on number of cuttings per stock plant

Factors and Levels	N	Total number of cuttings per stock plant
Alar		
0 g L^{-1}	12	69.4^{b}
1 g L^{-1}	12	69.9^{ab}
2 g L^{-1}	12	70.4^{a}
3 g L^{-1}	12	69.6^{b}
LSD		0.73
SE(±)		0.25
CV (%)		1.26
Cycocel		
0 ml L^{-1}	12	66.2^{c}
0.5 ml L^{-1}	12	68.7^{b}
1ml L^{-1}	12	75.0^{a}
1.5ml L^{-1}	12	69.2^{b}
LSD		0.73
SE(±)		0.25
CV (%)		1.26

Means followed by different letters are significantly different at the 5 % level of probability; ml=milliliter, g=gram; SE=Standard Error; CV=Coefficient of variation

4.2 Correlations

The present study unveiled that number of main branches was highly significant and positively correlated with number of cuttings with the correlation coefficient being (r=0.93**). This might be due to the fact that as number of branches increases, the number of shoots in the plants will also increase which then enhance the possibility of getting more number of cuttings. Mean while significant negative correlation was noticed with leaf area (r = -30*), stem length (r = -0.29*) and shoot dry weight (r = -0.31*). The negative association with stem length could be due to as plants have more pronounced vertical growth, formation of lateral branches is less stimulated for the reason of the dominance of terminal buds growth.

Canopy diameter was highly and positively correlated with internode length (r = 0.92**), stem length (r = 0.91**), average leaf area (r = 0.86**), shoot dry weight (r = 0.85**), shoot fresh weight (r = 0.82**), root fresh weight (r = 0.54**), and root dry weight (r = 0.58**). In contrary, it was highly significant and negatively correlated with stem diameter (r = -0.73**). The positive association with shoot dry weight might be attributed to as plants have wider

canopy size they would have more surface area too absorb solar radiation which can promote rate of production and distribution of assimilates to different part of the plant. The association with stem diameter could be result of more assimilate expenditure of the plants to have larger canopy which limits the amount assimilates to be kept in the stem.

Average leaf area found to have highly significant and positive correlation with internode length, stem length, shoot fresh weight, shoot dry weight, root fresh and dry weights with the correlation coefficient being 0.91**, 0.91**, 0.66**, 0.73**, 0.63**, and 0.69** respectively. In contrast, it has highly significant and negative correlation with stem diameter (r = -0.65**). Average leaf area observed having a significant and negative correlation with number of primary branches and number of cuttings with a correlation coefficient -0.30*, and -0.33*, respectively. This could be attributed to the fact that with increasing leaf area the vegetative or the upward growth of plants would be encouraged so there will be less stimulation of lateral branching.

Stem length has shown a highly significant and positive correlation with internode length (r = 0.96**), shoot fresh weight (r = 0.75**), shoot dry weight (r = 0.78**), root fresh weight (r = 0.66**), and root dry weight (r = 0.71**). Stem length had highly significant and negative correlation with stem diameter (r = -0.69**). Regarding number of cuttings, it had significant and negative correlation having a correlation coefficient of -0.34*.

Root dry weight had highly significant and positive correlation with internode length and root fresh weight with the correlation coefficient being 0.77** and 0.90** respectively. Root dry weight had significant and positive correlation with shoot fresh weight having a correlation coefficient of 0.34*. Root fresh weight had shown highly significant and positive correlation with internode length (r = 0.73**) and root dry weight (r = 0.90**). Significant positive correlation was noticed between root fresh weight and shoot fresh weight having a correlation coefficient of 0.30*.

Shoot dry weight had a highly significant and positive correlation with internode length (r = 0.73**), and shoot fresh weight (r = 0.73**). It has shown a highly significant and negative correlation with stem diameter (r = -0.84**).

Shoot fresh weight found to have highly significant and positive correlation with internode length with the correlation coefficient being 0.73**. A significant and positive correlation was noticed for root fresh weight (r = 0.34*) and root dry weight (r = 0.31*). Shoot fresh weight was highly and negatively correlated with stem diameter (r=-0.61**) and negatively correlated with number of cuttings (r = -0.32*).

The association of number of cuttings with number of primary branches and stem diameter was highly significant and positive with a correlation coefficient of 0.93** and 0.66**, respectively. Number of cuttings had a significant and negative correlation with shoot dry weight (r = -0.32**). The negative association with shoot dry could be due to the plants utilization more assimilates for formation of more number of shoots which can exhaust the plants dry matter content.

Table 6. Simple Correlation Coefficients among Response Variables

	CD	**IL**	**LA**	**NB**	**SD**	**SL**	**RDW**	**RFW**	**SDW**	**SFW**	**NC**
CD	-										
IL	0.92**	-									
LA	0.86**	0.91**	-								
NB	-0.19	-0.19	-0.30*	-							
SD	-0.73**	-0.66**	-0.65**	0.65**	-						
SL	0.91**	0.96**	0.91**	-0.29*	-0.69**	-					
RDW	0.58**	0.77**	0.69**	-0.03	-0.26	0.71**	-				
RFW	0.54**	0.73**	0.63**	-0.08	-0.26	0.66**	0.90**	-			
SDW	0.85**	0.75**	0.73**	-0.31*	-0.84**	0.78**	0.28	0.26	-		
SFW	0.82**	0.73**	0.66**	0.03	-0.61**	0.75**	0.34*	0.30*	0.90**	-	
NC	-0.24	-0.26	-0.33*	0.93**	0.66**	-0.34*	-0.08	-0.11	-0.32*	0.02	-

***,* = statistically significant difference at 0.1 % and 5 % probability level, respectively; CD=Canopy diameter (cm); IL=Internode length (cm); SD=Stem diameter (mm); SL=Stem length (mm); NB=Number of main branches; LA=Average Leaf area (cm^2); RDW=Root dry weight (g)RFW=Root fresh weight (g), SDW=Shoot dry weight (g); SFW=Shoot fresh weight (g); NC=number of cuttings per stock plant.*

4.3 Subsequent Rooting Performance of *Verbena hybrida* Cultivar Vegas Scarlet cuttings as affected by Cycocel and Alar Stock Plant Treatment

Based on the results presented in Table 7, there was a significant ($P<0.05$) interaction persistent effect among different concentrations of Alar and Cycocel on number of roots per cutting.

Table 7. P Values for ANOVA for number of roots per cutting, root fresh weight, root dry weight, root length, percentage of rooting, and root volume as influenced by Alar and Cycocel and their interaction

Source of Variation	DF	NR	RFW	RDW	RL	PR	RV
Alar	3	<.0001	0.0617	0.0684	0.8615	0.1011	0.1130
Cycocel	3	0.0113	0.0702	0.1480	0.7990	0.2594	0.3483
Alar * Cycocel	9	0.0465	0.3366	0.9595^{ns}	0.0786	0.1184^{ns}	0.1284
CV (%)		3.24	1.85	3.47	3.33	1.65	6.42

ns=non significantly different ;DF=Degree of Freedom; NR=Number of roots per cutting; RFW=Root fresh weight; RDW=Root dry weight; RL=Mean Root length; PR=Percentage of rooted cuttings; RV=Root volume

As shown Fig.9 (Table 8) the maximum number of roots per cutting (9.37) was obtained from cuttings that were collected from stock plants treated with 0.5ml L^{-1} CCC X 2g L^{-1} Alar which however was not significantly different from those treated with 0ml L^{-1} CCC X 3g L^{-1} Alar (9.13), 0ml L^{-1} CCC X 2g L^{-1} Alar (8.97), 0.5ml L^{-1} CCC X 3g L^{-1} Alar (9.13), 1ml L^{-1} CCC X 2g L^{-1} Alar (9.17) and 1.5ml L^{-1} CCC X 2g L^{-1} Alar (9.07). On the other hand, cuttings from stock plants with no application of Alar and Cycocel produced significantly the minimum number of roots per cutting (7.80) which however was at par with those collected from stock plants treated with 0ml L^{-1} CCC X 1g L^{-1} Alar (8.27). Even though there was a statistical difference among the treatments on the number of roots per cuttings, the difference between the maximum and minimum value was very small.

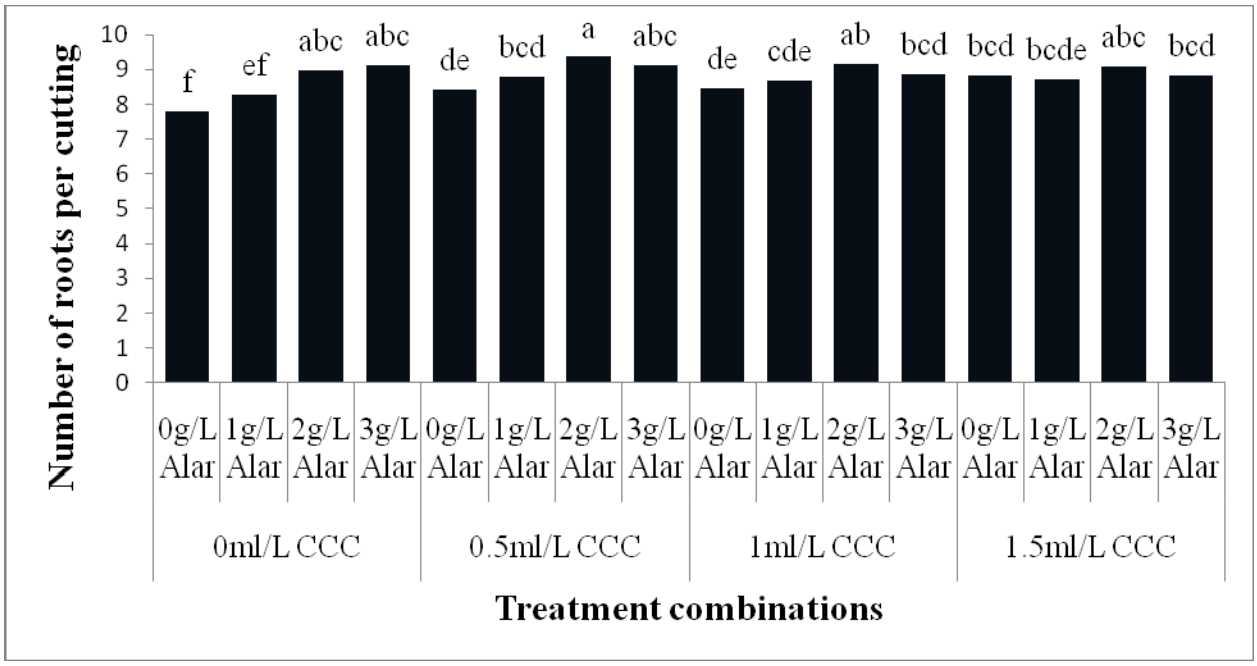

Fig. 9 Interaction effect of Cycocel and Alar on number of roots per cutting.

Similar effect on root number of cuttings was reported by Read and Hoyser (1971) from application of Alar on several species of ornamental plants. Such result could be due the persistent effect of plant growth retardants by altering the endogenous hormone levels like auxin and gibberellins which play a great role in adventitious root formation (Read and Yang, 1989). The modification in gibberellins levels could reduce their role in inhibiting rooting or reduce shoot growth which may compete with the base of cuttings for assimilates to the detriment of rooting. Such possible explanation was also made by Kefford (1973).

Table 7 depicts there was a non significant difference ($P>0.05$) among different concentrations of both Alar and Cycocel pertaining to the other rooting parameters except number of roots per cutting. Similarly, the interaction effect between Cycocel and Alar was found insignificant ($P>0.05$) for the rooting parameters. The result indicates that Cycocel and Alar applied on the stock plants have less persistent effect to influence the rooting performance of the cuttings collected. This finding is in line with the observation made by Beck and Sink (1974) who reported Alar had no effect on rooting of *Euphorbia spp*. This could be due the short life span nature of Alar and Cycocel that limits the duration to persist in the tissue of the cuttings. More persistent activity might have influenced the level of gibberellins and auxin in the cuttings that

will have an impact on root development. Besides, since the cuttings have uniform leaf size based on the farms cutting size specification, photosynthetic activity of the cuttings will be comparable because of uniformity and less persistent activity of Alar and Cycocel on regulation of leaf size. According to Dole and Gibson (2006) photosynthetic activity of the shoot system of cuttings is one of the basic factors for optimal rooting. In contrast to these finding, Read and Hoysler (1969) illustrated increased rooting ability of *Chrysanthemum*, *Dahlia* and poinsettia cuttings from stock plant application of Alar. Read and Yang (1989) also demonstrated better rooting performance of *Dahlia* from application of Alar and Cycocel on stock plants. Generally, such divergence results might be explained by variable responses from different plant species.

Table8. Interaction persistent effects of Cycocel and Alar on number of roots per cutting, root volume, percentage of rooted cuttings, root fresh weight, root dry weight, and root length.

Treatment combinations	MEAN					
	NR	PR (%)	RL(cm)	RV(cm^3)	RFW(g)	RDW(g)
0 ml^{-1} L CCC X 0 g^{-1} L Alar	7.80^{f}	92.80	7.20	0.317	1.550	0.039
0 ml^{-1} L CCC X 1 g^{-1} L Alar	8.27^{ef}	90.27	7.42	0.337	1.540	0.041
0 ml^{-1} L CCC X 2 g^{-1} L Alar	8.97^{abc}	93.33	7.56	0.333	1.557	0.042
0 ml^{-1} L CCC X 3 g^{-1} L Alar	9.13^{abc}	92.70	7.17	0.310	1.573	0.041
0.5 ml^{-1} L CCC X 0 g^{-1} L Alar	8.43^{de}	90.00	7.61	0.350	1.503	0.039
0.5 ml^{-1} L CCC X 1 g^{-1} L Alar	8.80^{bcd}	93.10	7.17	0.327	1.560	0.039
0.5 ml^{-1} L CCC X 2 g^{-1} L Alar	9.37^{a}	93.23	7.58	0.310	1.560	0.040
0.5 ml^{-1} L CCC X 3 g^{-1} L Alar	9.13^{abc}	92.00	7.31	0.357	1.580	0.040
1 ml^{-1} L CCC X 0 g^{-1} L Alar	8.47^{de}	92.10	7.43	0.343	1.560	0.040
1 ml^{-1} L CCC X 1 g^{-1} L Alar	8.67^{cde}	93.30	7.48	0.347	1.557	0.041
1 ml^{-1} L CCC X 2 g^{-1} L Alar	9.17^{ab}	94.13	7.17	0.317	1.597	0.041
1 ml^{-1} L CCC X 3 g^{-1} L Alar	8.87^{bcd}	93.33	7.62	0.347	1.573	0.041
1.5 ml^{-1} L CCC X 0 g^{-1} L Alar	8.83^{bcd}	92.67	7.28	0.353	1.583	0.040
1.5 ml^{-1} L CCC X 1 g^{-1} L Alar	8.73^{bcde}	90.83	7.44	0.320	1.567	0.041
1.5 ml^{-1} L CCC X 2 g^{-1} L Alar	9.07^{abc}	92.03	7.49	0.327	1.593	0.041
1.5 ml^{-1} L CCC X 3 g^{-1} L Alar	8.83^{bcd}	93.23	7.43	0.350	1.573	0.041
S.E±	0.16	0.88	0.14	0.01	0.02	0.001
LSD	0.175	ns	ns	ns	ns	ns

Means followed by different letters are significantly different at the 5 % level of probability; ml=milliliter; g=gram; SE=Standard Error; CV=Coefficient of variation; LSD=Least significant difference; NR=Number of roots per cutting; RFW=Root fresh weight; RDW=Root dry weight; RL=Mean Root length; PR=Percentage of rooted cuttings; RV=Root volume

5. SUMMARY AND CONCLUSION

The floriculture industry in Ethiopia has been rapidly growing for the last five years and is becoming a very promising business opportunity for economic development of the country. Even though cut flower production is the major category of this sector, some companies are producing rooted and unrooted cuttings of bedding and pot plants. Because of the vigorous and less branched growth of *Verbena hybrida*, chemical plant growth retardants are very useful tools for controlling plant height and creating more branched stock plants for maximizing cutting yield.

Identification of effective growth retarding chemicals and establishing their optimum application rate is a basic input for promoting production. These obviously will give growers the stamina for competition in the global market and encourage other investors. In this regard, a study was conducted to determine the appropriate type and concentration of chemical growth retardants that can produce increased number of cuttings with acceptable rooting capacity.

The results of the investigation which consisted of two types of growth retardants at four concentrations each depicted that Alar and Cycocel had apparent effect on the stock plant growth of *Verbena hybrida*. It was vividly observed that interaction of Alar and CCC considerably affected internode length, main stem length, number of main branches, canopy diameter, root fresh and dry weight, and shoot fresh and dry weight. And yet, the interaction of CCC and Alar failed to have significant effects on stem diameter, average leaf area and number of cuttings.

Combined application of 1.5ml L^{-1} Cycocel and 3g L^{-1} Alar resulted in the minimum internode length (3.88cm), stem length (10.15cm), canopy diameter (48.44cm), root dry weight (12.39g), root fresh weight (72.86g), shoot fresh weight (196.74g), and shoot dry weight (35.81g). On the hand, the maximum value for internode length (7.56cm), stem length (21.35cm), canopy diameter (60.33cm), shoot fresh weight (229.80g), and shoot dry weight (35.81g) were observed from the untreated stock plants (0ml L^{-1} CCC X 0g L^{-1} Alar). The maximum value for root fresh

weight (82.01g) was recorded from application of 1ml L^{-1} CCC X 0g L^{-1} Alar. In contrast, 1.5 ml L^{-1} CCC X 0g L^{-1} Alar exhibited the highest root dry weight (16.17g) as judged against to the other treatment combinations.

With respect to average leaf area, a declining trend was observed as the concentration of Alar increased from 0g L^{-1} (8.26cm^2) to 3g L^{-1} (6.97cm^2). Similarly, there was a decreasing tendency in average leaf area as the concentration of Cycocel increased from 0ml L^{-1} (8.16cm^2) to 1.5 ml L^{-1} (6.83cm^2). The reverse trend was revealed regarding stem diameter where in increasing concentrations of Alar and Cycocel increasing of stem diameter was observed. 1ml L^{-1} CCC resulted in the maximum stem diameter (1.45mm) and 0ml L^{-1} produced the minimum (1.27mm). In case of Alar, 3g L^{-1} resulted in the maximum stem diameter (1.40cm) where as 0g L^{-1} and 1g L^{-1} brought about the minimum stem diameter (1.37mm).

Regarding the number of main branches more number of main branches was obtained from the combined application of 1ml L^{-1} CCC X 2g L^{-1} Alar but no further increase was observed beyond this concentration. Pertaining to the number of cuttings per stock plant, which is the major concern of the growers, maximum number of cuttings (75.0) was recorded from application of 1ml L^{-1} CCC among concentrations of Cycocel and among concentrations of Alar 2g L^{-1} resulted in maximum cutting yield (70.4).

The experiment on rooting performance of cuttings taken from stock plants treated with chemical growth retardants depicted a significant variation only among the mean number of roots per cuttings. Subsequently, the maximum number of roots per cutting (9.37) was obtained from cuttings that were collected from stock plants treated with 0.5ml L^{-1} CCC X 2g L^{-1} Alar. In contrast, cuttings from non treated stock plants produced minimum number roots per cutting (7.80). Generally, the persistent or subsequent effect of the treatments on rooting performance of cuttings was not notable for consideration since there was a non significant difference for most of rooting parameters specially on percentage of rooted cuttings.

One of the main prerequisites for successful production of cuttings is the selection of quality cuttings which mainly focuses on the rooting ability. The capability of cuttings to generate

uniform roots within a short period of time is vital for successful propagation and establishment of a sustainable business. Cognizant with the findings of the study, Alar and Cycocel influence the growth of *Verbena hybrida* stock plants and cutting yield without causing a significant reduction on subsequent rooting performance of cuttings. Thus, our concern should be on the quantity of cuttings obtained from the treated stock plants. Based on this aspect application of $1ml^{-1}L$ Cycocel, which demonstrated positive influence on cutting production, can be recommended for use by commercial growers.

Irrespective of number of cuttings produced from stock plants, among the mixed applications, $1ml^{-1}$ L CCC and $2g^{-1}$ L Alar can be considered as better performing treatment from other combinations because it attained significant superiority in relation to number of main branches. But further research work should be implemented on how to improve the cutting yield with appropriate size as it has produced insignificant number of cuttings (fit for propagation) in comparison with other treatment combinations with less number of main branches.

In general, since the investigation was conducted only once in a controlled environment the outcome of this experiment can only be used as guidance for growing *Verbenas* under greenhouse condition. Moreover, the combined application of Alar and Cycocel which showed a potential influence should be comprehensively studied to come up with a pertinent recommendation by including other production factors like frequency and type of application of retardants, rooting media influences, and economic factors.

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7. APPENDICES

Appendix Table. 1. ANOVA Mean Square Values for Growth Parameters

Source	DF	CD	IL	LA	NB	SD	SL	RDW	RFW	SDW	SFW	NC
Alar	3	56.52**	7.39**	3.88**	1.24*	0.003*	54.10**	19.19**	135.39**	19.35**	100.24**	2.41*
Cycocel	3	108.24**	6.25**	4.15**	105.31**	0.08**	53.83**	0.61*	3.34*	656.12**	1301.83**	165.88**
Alar*Cycocel	9	1.59*	0.14**	0.13^{ns}	1.11**	0.0001^{ns}	1.97**	0.43*	3.82**	47.38**	20.78*	0.77^{ns}
Error	32	0.68	0.011	0.115	0.300	0.0006	0.21	0.17	0.94	1.10	7.09	0.776
CV (%)		1.50	4.50	5.65	1.86	2.96	4.78	2.91	1.27	2.24	1.24	1.26
R^2		0.96	0.99	0.87	0.97	0.92	0.98	0.92	0.94	0.98	0.95	0.95

**, **= statistically significant difference at 5 %, 1 % probability level, respectively; ^{ns}=non significantly different; DF=Degree of Freedom; IL=Internode length; SL=stem length; NB=number of main branches; LA=Average leaf area; CD=canopy diameter; RFW=root fresh weight; RDW=root dry weight; SFW=shoot fresh weight; SDW=shoot dry weight; NC=number of cuttings; CV=Coefficient of Variation*

Appendix Table 2. ANOVA Mean Square Values for Rooting Parameters

Source	DF	NR	RL	PR	RV	RFW	RDW
Alar	3	1.438**	0.015^{ns}	5.26^{ns}	0.001^{ns}	0.0023^{ns}	0.00001^{ns}
Cycocel	3	0.352*	0.02^{ns}	3.28^{ns}	0.0005^{ns}	0.0022^{ns}	0.000004^{ns}
Alar*Cycocel	9	0.18*	0.12^{ns}	4.11^{ns}	0.0008^{ns}	0.001^{ns}	0.000001^{ns}
Error	32	0.081	0.06	2.33	0.0005	0.001	0.000002
CV (%)		3.24	3.33	1.65	6.43	1.85	3.47
R^2		0.73	0.38	0.46	0.44	0.45	0.34

**, **= statistically significant difference at 5 %, 1 % probability level, respectively; ^{ns}=non significantly different; DF=Degree of Freedom; NR=Number of roots per cutting; RFW=Root fresh weight; RDW=Root dry weight; RL=Mean Root length; PR=Percentage of rooted cuttings; RV=Root volume; CV=Coefficient of Variation.*

Appendix Table 3. P values for Least Squares Means for effect t of Alar* Cycocel on Canopy diameter

i/j	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
2	0.175															
3	<.0001	0.0002														
4	<.0001	<.0001	0.0576													
5	0.104	0.7765	0.0005	<.0001												
6	<.0001	0.0019	0.4354	0.0095	0.0041											
7	<.0001	<.0001	0.0092	0.427	<.0001	0.0012										
8	<.0001	<.0001	<.0001	0.0146	<.0001	<.0001	0.0851									
9	<.0001	<.0001	0.618	0.1524	0.0001	0.2051	0.03	0.0003								
10	<.0001	0.0008	0.6318	0.0198	0.0018	0.7615	0.0027	<.0001	0.3309							
11	<.0001	<.0001	0.01	0.4469	<.0001	0.0013	0.9726	0.0794	0.0324	0.0029						
12	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	0.0001	0.0166	<.0001	<.0001	0.0001					
13	<.0001	<.0001	<.0001	0.0008	<.0001	<.0001	0.0064	0.2626	<.0001	<.0001	0.0059	0.175				
14	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	0.003	<.0001	<.0001	<.0001	0.4975	0.0463			
15	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	0.0003	<.0001	<.0001	<.0001	0.142	0.0068	0.4186		
16	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	0.0002	0.0015	

Appendix Table 4. P values for Least Squares Means for effect of Alar* Cycocel on Internode length

i/j	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
2	<.0001															
3	<.0001	<.0001														
4	<.0001	<.0001	0.0079													
5	0.0001	<.0001	<.0001	<.0001												
6	<.0001	<.0001	0.0024	<.0001	<.0001											
7	<.0001	<.0001	0.1717	0.1604	<.0001	<.0001										
8	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001									
9	<.0001	0.0006	0.0004	<.0001	<.0001	0.4896	<.0001	<.0001								
10	<.0001	<.0001	0.2374	0.0003	<.0001	0.044	0.0139	<.0001	0.0087							
11	<.0001	<.0001	<.0001	0.0001	<.0001	<.0001	<.0001	0.8172	<.0001	<.0001						
12	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001					
13	<.0001	<.0001	0.0478	<.0001	<.0001	0.223	0.0016	<.0001	0.061	0.3992	<.0001	<.0001				
14	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	0.6443	<.0001			
15	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	0.0008	<.0001	0.0002		
16	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	

Appendix Table 5. P values for Least Squares Means for effect of Alar* Cycocel on Number of primary branches

i/j	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
2	0.4313															
3	0.0018	0.014														
4	0.0562	0.245	0.1666													
5	<.0001	<.0001	0.0002	<.0001												
6	<.0001	<.0001	<.0001	<.0001	0.0179											
7	<.0001	<.0001	<.0001	<.0001	0.0049	0.6005										
8	<.0001	<.0001	<.0001	<.0001	0.5903	0.0598	0.0186									
9	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001								
10	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	0.3014							
11	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	0.0156	0.1422						
12	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	0.7508	0.18	0.0071					
13	<.0001	<.0001	<.0001	<.0001	0.1967	0.2479	0.0977	0.4442	<.0001	<.0001	<.0001	<.0001				
14	<.0001	<.0001	0.0007	<.0001	0.6315	0.0055	0.0014	0.3117	<.0001	<.0001	<.0001	<.0001	0.0809			
15	<.0001	<.0001	0.013	0.0003	0.1126	0.0002	<.0001	0.0371	<.0001	<.0001	<.0001	<.0001	0.0059	0.2598		
16	<.0001	<.0001	0.0024	<.0001	0.3401	0.0015	0.0004	0.1403	<.0001	<.0001	<.0001	<.0001	0.029	0.6315	0.5121	

Appendix Table 6. P values for Least Squares Means for effect of Alar* Cycocel on Stem length

i/j	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
2	<.0001															
3	<.0001	<.0001														
4	<.0001	<.0001	0.0546													
5	<.0001	0.0001	<.0001	<.0001												
6	<.0001	<.0001	0.5051	0.0118	<.0001											
7	<.0001	<.0001	0.0426	0.9077	<.0001	0.0089										
8	<.0001	<.0001	0.0003	0.0487	<.0001	<.0001	0.0622									
9	<.0001	<.0001	0.6755	0.0215	<.0001	0.8029	0.0163	<.0001								
10	<.0001	<.0001	0.2772	0.3802	<.0001	0.0846	0.3216	0.0061	0.1363							
11	<.0001	<.0001	0.0011	0.1194	<.0001	0.0002	0.1478	0.6562	0.0003	0.0182						
12	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	0.0008	<.0001	<.0001	0.0002					
13	<.0001	<.0001	0.3174	0.3346	<.0001	0.1008	0.281	0.0048	0.1601	0.9289	0.0147	<.0001				
14	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	0.0004	<.0001	<.0001	0.0001	0.8306	<.0001			
15	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	0.0401	<.0001	0.0634		
16	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	

Appendix Table 7. P values for Least Squares Means for effect of Alar* Cycocel on Root dry weight

i/j	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
2	<.0001															
3	<.0001	1.000														
4	<.0001	0.6471	0.6471													
5	0.8453	<.0001	<.0001	<.0001												
6	0.0001	0.1255	0.1255	0.0502	<.0001											
7	<.0001	0.7111	0.7111	0.93	<.0001	0.0604										
8	<.0001	0.121	0.121	0.2665	<.0001	0.0034	0.2316									
9	0.93	<.0001	<.0001	<.0001	0.7774	0.0002	<.0001	<.0001								
10	<.0001	0.3281	0.3281	0.1553	<.0001	0.5659	0.1812	0.0145	<.0001							
11	<.0001	0.6126	0.6126	0.9611	<.0001	0.0452	0.8914	0.2875	<.0001	0.1423						
12	<.0001	0.0027	0.0027	0.0088	<.0001	<.0001	0.007	0.1063	<.0001	0.0002	0.0099					
13	0.5085	<.0001	<.0001	<.0001	0.6401	<.0001	<.0001	<.0001	0.4545	<.0001	<.0001	<.0001				
14	<.0001	0.388	0.388	0.1906	<.0001	0.4901	0.2208	0.0191	<.0001	0.9068	0.1752	0.0002	<.0001			
15	<.0001	0.0072	0.0072	0.0219	<.0001	<.0001	0.0178	0.2103	<.0001	0.0005	0.0245	0.7039	<.0001	0.0007		
16	<.0001	<.0001	<.0001	0.0002	<.0001	<.0001	0.0002	0.0053	<.0001	<.0001	0.0003	0.1937	<.0001	<.0001	0.0968	

Appendix Table 8. P values for Least Squares Means for effect of Alar* Cycocel on Root fresh weight

i/j	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
2	0.0013															
3	0.0008	0.8509														
4	<.0001	0.0339	0.0511													
5	0.0435	<.0001	<.0001	<.0001												
6	0.0002	0.4464	0.5651	0.1582	<.0001											
7	<.0001	0.0215	0.033	0.841	<.0001	0.1093										
8	<.0001	<.0001	0.0001	0.0271	<.0001	0.0007	0.0423									
9	0.0037	<.0001	<.0001	<.0001	0.3117	<.0001	<.0001	<.0001								
10	<.0001	0.1805	0.2469	0.4034	<.0001	0.5539	0.3021	0.0034	<.0001							
11	<.0001	0.0006	0.001	0.1211	<.0001	0.0047	0.1741	0.474	<.0001	0.0205						
12	<.0001	<.0001	<.0001	0.0187	<.0001	0.0004	0.0298	0.8738	<.0001	0.0022	0.383					
13	0.0099	<.0001	<.0001	<.0001	0.5265	<.0001	<.0001	<.0001	0.7009	<.0001	<.0001	<.0001				
14	<.0001	0.0276	0.042	0.9267	<.0001	0.134	0.9135	0.0333	<.0001	0.3546	0.1435	0.0232	<.0001			
15	<.0001	0.0002	0.0003	0.0455	<.0001	0.0013	0.0694	0.815	<.0001	0.0062	0.6284	0.6948	<.0001	0.0554		
16	<.0001	<.0001	<.0001	0.0109	<.0001	0.0002	0.0176	0.7009	<.0001	0.0012	0.2744	0.8215	<.0001	0.0136	0.5374	

Appendix Table 9. P values for Least Squares Means for effect of Alar* Cycocel on Shoot dry weight

i/j	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
1																
2	0.0069															
3	0.0001	0.1474														
4	<.0001	0.0255	0.3968													
5	<.0001	<.0001	0.0009	0.0081												
6	<.0001	<.0001	<.0001	<.0001	0.0044											
7	<.0001	<.0001	<.0001	<.0001	<.0001	0.1558										
8	<.0001	<.0001	<.0001	<.0001	<.0001	0.1495	0.9815									
9	<.0001	<.0001	<.0001	<.0001	<.0001	0.0026	0.0801	0.0839								
10	<.0001	<.0001	<.0001	<.0001	0.0001	0.1894	0.911	0.8927	0.0638							
11	<.0001	<.0001	<.0001	<.0001	0.006	0.9079	0.1262	0.1209	0.0019	0.1547						
12	<.0001	<.0001	<.0001	<.0001	<.0001	0.0003	0.0132	0.014	0.4204	0.0101	0.0002					
13	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001				
14	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	0.0314			
15	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	0.0151	0.752		
16	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	0.0006	0.129	0.2241	

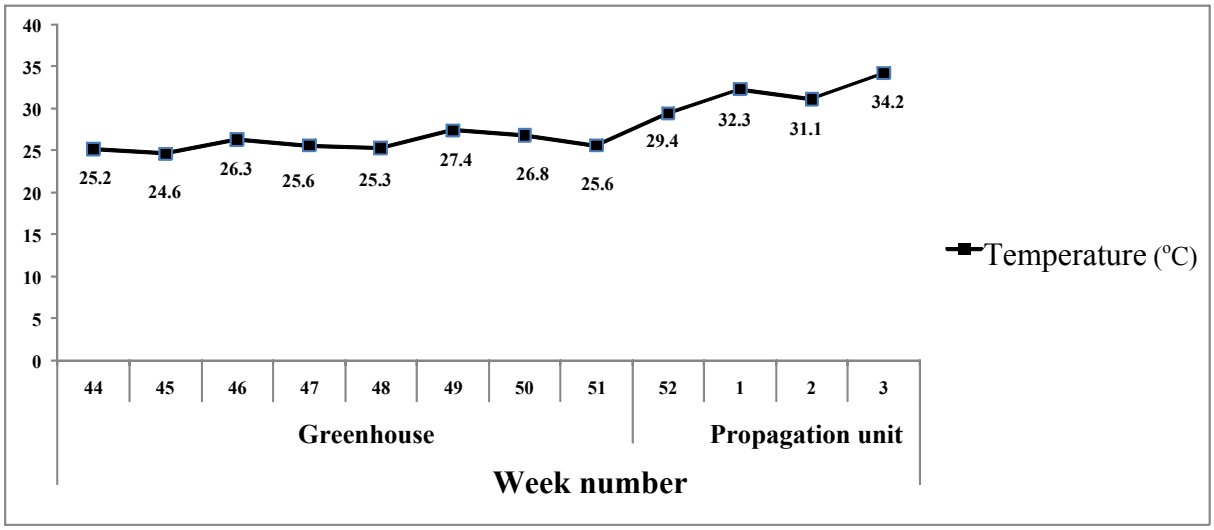
Appendix Table 10. P values for Least Squares Means for effect of Alar* Cycocel on Shoot fresh weight

i/j	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
2	0.0417															
3	0.0003	0.0547														
4	<.0001	0.0077	0.4021													
5	<.0001	0.0025	0.2072	0.6641												
6	<.0001	0.0001	0.0262	0.1481	0.3044											
7	<.0001	<.0001	0.0012	0.0108	0.0302	0.2297										
8	<.0001	<.0001	0.0004	0.0044	0.0131	0.1229	0.7211									
9	<.0001	0.0063	0.3607	0.9382	0.7211	0.17	0.0131	0.0053								
10	0.0003	0.0547	1	0.4021	0.2072	0.0262	0.0012	0.0004	0.3607							
11	0.0145	0.6466	0.1356	0.0234	0.0082	0.0005	<.0001	<.0001	0.0196	0.1356						
12	<.0001	<.0001	0.0114	0.0756	0.1718	0.7256	0.3905	0.2274	0.0883	0.0114	0.0002					
13	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	0.0002	0.0005	<.0001	<.0001	<.0001	<.0001				
14	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	0.1197			
15	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	0.0177	0.3742		
16	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	0.0024	0.0989	0.4305	

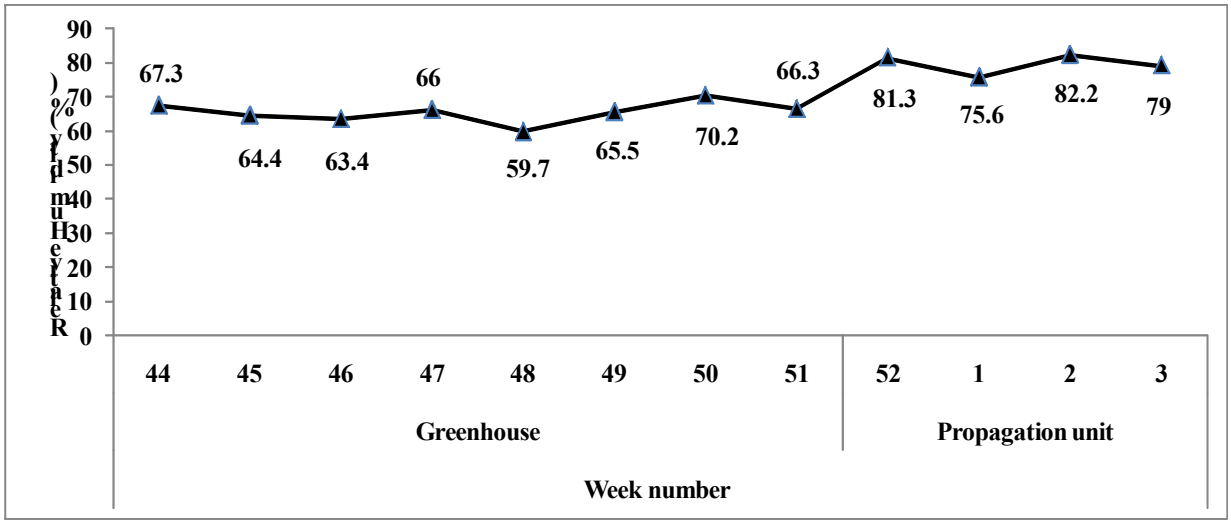
Appendix Table 11. Interaction effects of Cycocel and Alar on internode length, main stem length, number of main branches, canopy diameter, root fresh and dry weight, and shoot fresh and dry weight.

Treatment combinations	MEAN							
	IL	**SL**	**NB**	**CD**	**RFW**	**RDW**	**SFW**	**SDW**
0 ml^{-1} L CCC X 0 g^{-1} L Alar	7.56^{a}	21.35^{a}	5.73^{i}	60.33^{a}	79.53^{b}	15.94^{a}	229.80^{a}	57.84^{a}
0 ml^{-1} L CCC X 1 g^{-1} L Alar	6.80^{c}	18.02^{c}	6.09^{i}	59.39^{a}	76.75^{c}	13.94^{bcd}	225.18^{b}	55.36^{b}
0 ml^{-1} L CCC X 2 g^{-1} L Alar	6.13^{fg}	15.82^{de}	7.25^{h}	56.58^{bc}	76.60^{cd}	13.94^{bcd}	220.85^{bc}	54.09^{bc}
0 ml^{-1} L CCC X 3 g^{-1} L Alar	5.89^{h}	15.08^{efg}	6.62^{hi}	55.25^{cd}	75.00^{def}	13.78^{bcd}	219.00^{cd}	53.35^{c}
0.5 ml^{-1} L CCC X 0 g^{-1} L Alar	7.19^{b}	19.62^{b}	9.16^{efg}	59.20^{a}	81.20^{a}	16.01^{a}	218.05^{cd}	50.93^{d}
0.5 ml^{-1} L CCC X 1 g^{-1} L Alar	6.41^{d}	16.07^{d}	10.27^{cd}	57.11^{b}	76.14^{cde}	14.47^{b}	215.78^{de}	48.31^{e}
0.5 ml^{-1} L CCC X 2 g^{-1} L Alar	6.01^{gh}	15.03^{fgh}	10.51^{c}	54.71^{de}	74.84^{efg}	13.81^{bcd}	213.11^{e}	47.06^{ef}
0.5 ml^{-1} L CCC X 3 g^{-1} L Alar	5.50^{i}	14.32^{h}	9.40^{def}	53.51^{ef}	73.17^{h}	13.40^{de}	212.33^{e}	47.04^{ef}
1 ml^{-1} L CCC X 0 g^{-1} L Alar	6.47^{d}	15.97^{d}	13.21^{b}	56.24^{bc}	82.01^{a}	15.91^{a}	218.83^{cd}	45.51^{fg}
1 ml^{-1} L CCC X 1 g^{-1} L Alar	6.23^{ef}	15.41^{def}	13.68^{ab}	56.91^{b}	75.67^{cde}	14.28^{bc}	220.85^{bc}	47.16^{ef}
1 ml^{-1} L CCC X 2 g^{-1} L Alar	5.52^{i}	14.486^{gh}	14.35^{a}	54.73^{de}	73.74^{fgh}	13.77^{cd}	224.18^{b}	48.41^{e}
1 ml^{-1} L CCC X 3 g^{-1} L Alar	4.92^{j}	12.94^{i}	13.06^{b}	51.80^{gh}	73.04^{h}	12.84^{ef}	215.01^{de}	44.81^{g}
1.5 ml^{-1} L CCC X 0 g^{-1} L Alar	6.31^{de}	15.44^{def}	9.75^{cde}	52.74^{fg}	81.70^{a}	16.17^{a}	203.91^{f}	39.08^{h}
1.5 ml^{-1} L CCC X 1 g^{-1} L Alar	4.94^{j}	12.86^{ij}	8.94^{efg}	51.34^{h}	74.93^{efg}	14.24^{bc}	$200.44f^{g}$	37.15^{i}
1.5 ml^{-1} L CCC X 2 g^{-1} L Alar	4.60^{k}	12.14^{j}	8.43^{g}	50.78^{h}	73.35^{gh}	12.97^{ef}	198.48^{g}	36.87^{i}
1.5 ml^{-1} L CCC X 3 g^{-1} L Alar	3.88^{l}	10.15^{k}	8.72^{fg}	48.44^{i}	72.86^{h}	12.39^{f}	196.74^{g}	35.81^{i}
S.E±	0.06	0.26	0.32	0.48	0.56	0.24	1.54	0.61
LSD	0.175	0.755	0.912	1.38	1.61	0.69	4.43	1.75

Means followed by different letters are significantly different at the 5 % level of probability; ml=milliliter; g=gram; SE=Standard Error; CV=Coefficient of variation; LSD=Least significant difference; IL=Internode length; SL=stem length; NB=number of main branches; LA=Average leaf area; CD=canopy diameter; RFW=root fresh weight; RDW=root dry weight; SFW=shoot fresh weight; SDW=shoot dry weight; NC=number of cuttings per stock plant.



Appendix Fig 1. Greenhouse and Propagation unit Temperature Record during Growing Period



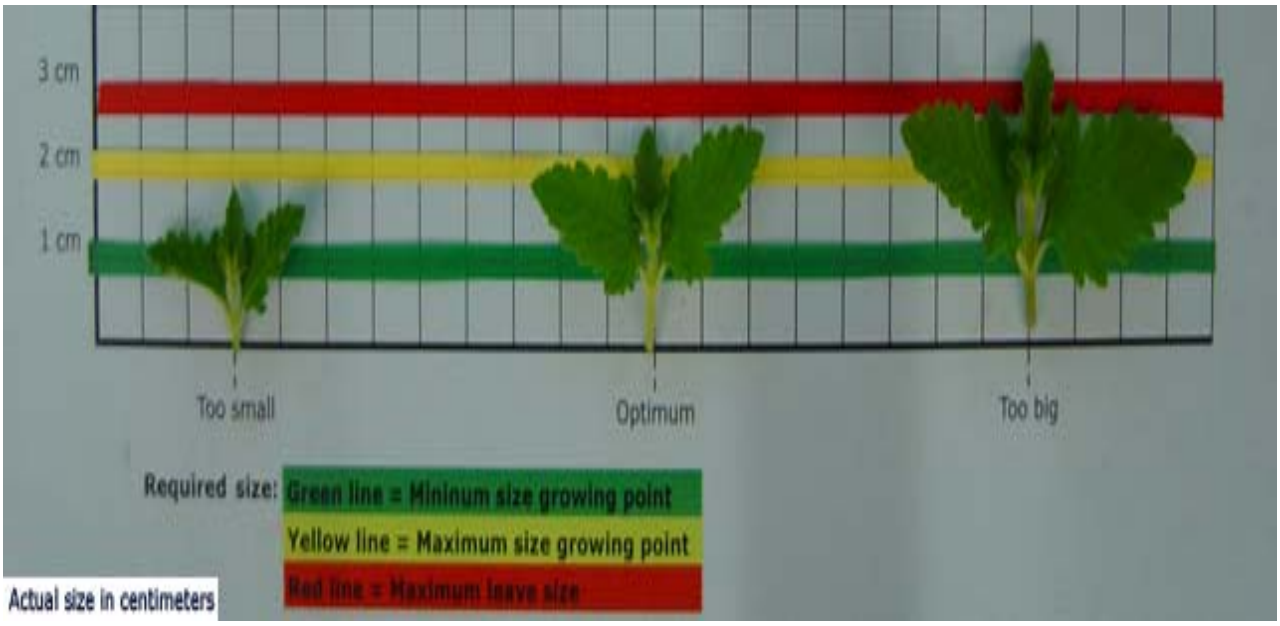
Appendix Fig 2. Greenhouse and Propagation unit Relative Humidity Record during Growing Period

Appendix Fig 3. Greenhouse for growing stock plants

Appendix Fig 4. Propagation unit for rooting the cuttings.

Appendix Fig 5. Verbena hybrida stock plants

Appendix Fig 6. Devices for environment control



Appendix Fig 7. Cutting size specification for Verbena hybrida cultivar Vegas Scarlet