EFFECT OF BENZYLADENINE, GIBBERELLIC ACID, AND ETHEPHON ON GROWTH AND FLOWER YIELD OF CHAMOMILE (*Matricaria chamomilla* L.) PLANTS

BY

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DECLARATION

This thesis is my original work and has not been presented for a degree in any University.

C.M.M. Mutunga — 14.10.1998 — Date

This thesis has been submitted for examination with our approval as University Supervisors.

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DEDICATION

This thesis is dedicated to

my beloved wife Beth Mueni

whose encouragement and support throughout

the course was an invaluable inspiration

and

to our children Kanini, Ndunge, Mutunga and Kitoo

who may, someday, choose to read it.

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ABSTRACT

An experiment at the University of Nairobi's Institute of Dryland Research Development and Utilization (IDRDU) in Kibwezi was conducted to study the effect of plant growth regulators benzyladenine, gibberellic acid, and ethephon on the growth and flower yield of chamomile plants. Plants were sprayed to run-off with various concentrations (0, 25, 50 and 75 mg/litre) of benzyladenine (BA) in two timings (4 and 6 weeks after transplanting - 4 and 6 WAT); gibberellic acid (GA) at rates of 0, 100, 200 and 300 mg/litre and ethephon at rates of 0, 50, 100 and 150 mg/litre at 6 WAT.

Benzyladenine applications significantly ($P = 0.05$) increased the vegetative growth and flower yield of chamomile plants. BA treatment at 4 WAT had no effect on dry matter production by the plants. Application of BA at 6 WAT increased dry matter production by the shoots and roots and also caused more partitioning of the dry matter to the roots than to the shoots. Generally, response to BA treatment was higher with the lower concentration of 25 mg/litre. Ethephon significantly ($P = 0.05$) increased plant spread, dry matter production by the shoots and roots with greater partitioning of the dry matter to the roots, and also increased flower yield. Plant response to increasing ethephon application rates was linear.

Spraying chamomile plants with GA reduced root growth and reduced flower yield at each harvest and consequently the total flower yield. The reduction on root growth and flower yield was linear and quadratic to increasing GA concentrations, respectively.

This study shows that BA and ethephon could be incorporated in the management practices of chamomile to enhance plant growth and improve the yield of dry flowers.

CHAPTER ONE

1.0 INTRODUCTION

1.1. Essential oils

Since ancient times, man has used plants in therapy, perfumes and as spices. Egyptian papyri dating back as far as 2000 B.C. record the common use in Egypt of plants like mustard, linseed, squill and myrrh (Trease and Evans, 1972; Balbaa, 1983).

Certain drugs are now obtained almost exclusively from cultivated plants such as cardamon, ginger, cinnamon, fennel, opium, linseed and chamomile (Trease and Evans, 1972). In other cases, both wild and cultivated plants are used. However, in most cases, it is advisable to cultivate medicinal plants because of the improved quality of the drugs (Balbaa, 1983; Franz 1983).

The medicinal property of most oils is attributed to their essential oils. These are volatile plant oils obtained by steam distillation of plant parts and which contribute to the odour of certain species.

Essential oils are complex mixtures of various compounds, therefore explaining the inherent variability exhibited by these oils regarding their medicinal properties. On the other hand, misuse of essential oils such as application in excessive doses or over long periods of time has been observed to result in undesired effects like allergy, necrosis, paralysis, abortion and sometimes cancer (Schilcher, 1985).

1.2. Chamomile, *Matricaria chamomilla L.*

Matricaria chamomilla L., the so called German, Hungarian or small chamomile is an annual plant from the family of Compositae. The plant tillers profusely to form a bushy herb ranging in height from 50 to 100 cm. The flowers are strongly scented and the leaves 1-3 times finely dissected. Each dried flower head is hemispherical and about 7-14 mm in diameter (Bailey, 1949; Kirk and Othmer, 1952; Masefield *et al.*, 1971). The flowers are smaller than those of *Anithemis nobillis* L., the so called Roman (English) chamomile whose flower diameter is about 12-20 mm (Trease and Evans, 1978).

The genus *Matricaria* has about 50 species native to Europe, Mediterranean region, Asia Minor, Egypt, Congo, Eastern and Southern African countries (Bailey, 1949; Watt and Breyer-Brandwijk, 1962; Masefield *et al.*, 1971). Some of the chamomile varieties are grown for ornamentals but most are grown for medicinal purposes. The medicinal product is highest in the flowers of chamomile but is also present in small and varying quantities in the leaves, stems and roots (Trease and Evans, 1972 and 1978).

Chamomile grows best in well drained, fertile soils of pH 5 (Eggens and Hilton, 1971) but can also grow well in partially salty soils and in soils with a pH range of 4-7 (Singh, 1970).

Chamomile is commonly propagated sexually. The seed is either directly sown in the field or seedlings are first raised in the nursery and then later transplanted to the field. In Kenya, transplanting is necessary due to the small size of the seeds and lack of appropriate precision planters. In the nursery, seeds are broadcasted on the soil surface and the nursery watered gently to maintain it moist. Germination occurs in four days and the seedlings are transplanted when they have reached 6-7 leaf stage i.e. at four weeks old (Vernar-Petri *et al.*, 1978).

Factors affecting chamomile growth and development include nutrition, growing site (micro-climate), photoperiodism, water availability, and varietal differences (Franz *et al*, 1978;

Penka 1978). Emongor (1988) working at Kabete found that application of 40 Kg $P_2 O_5$/ha during transplanting of chamomile seedlings and two weeks later top-dressing them with 50 Kg N/ha ensured good growth and development with high flower and essential oil yields. The essential oil was also of high quality. Chamomile has varieties falling in all the three categories of photoperiodic classes: short day, long day and day neutral (Saleh, *et al.*, 1978; Franz *et al.*, 1978).

Chamomile flowers are collected in dry weather. The first harvesting of the flower heads is done when the oldest flower heads of 50% of the plants have started to wilt (Trease and Evans, 1978). Subsequent harvests are done once every week and on average a total of 8 harvests per crop is possible. Chamomile grown on small farms is handpicked which permits removal of flowers without the stem. In large scale farming, flowers are collected by means of flower scoops or stripers. When using these implements, the harvesters must gather the flowers as carefully and with as little stem material and extraneous matter as possible. A single worker with a "flower comb" can collect from 60 to 100 Kg of fresh flowers per day while by hand he can gather only 8 to 10 Kg (Bailey, 1949; Masefield *et al.*, 1971; Trease and Evans, 1978). The harvested flowers are sifted in a suspended sieve (mesh diameter 7-12 mm) to separate the flower heads from the flowers with attached stems and from clinging bits of weed or grass.

The flowers are then spread out into thin layers of maximum 3 cm in fine-mesh screen trays and air dried under shade to equilibrium moisture content. The flowers are very delicate and prone to damage and once in the trays should not be turned or disturbed. Five kilograms of fresh flowers give 1 Kg of air-dried flowers.

After drying, grade one flowers (with a flower stalk less than 3 cm long) are used for herbal tea. The other grades are used for extraction of essential oil using steam distillation. The essential oil is light blue in colour when fresh and is highest in developed flower heads and maximum approximately one week after beginning of flowering (Franz, 1980; Holzl and Demuth,

1975). The oil content of chamomile flowers is in the range of 0.2% to 1.5% and is composed of chamazulene, bisabolol, bisabololoxides A, B and C, cis-spiroether, trans-spiroether, farnesene, matricine, and flavonoids (Trease and Evans, 1972; Isaac, 1974, 1980; Martindale, 1977; Franz, 1980).

The essential oil is used for the manufacture of drugs for the treatment of allergy, sleeplessness, stomach ulcers induced by stress, sore throat, rheumatic disease of the joints, baby teething powders and as an antiseptic (Martindale, 1977; Trease and Evans, 1978; Schilcher, 1985). The essential oil is further used for flavouring liquors and for making cosmetics (Bailey, 1949; Masefield *et al.*, 1971; Trease and Evans, 1972).

Chamomile flowers and essential oil are very expensive. The present Kenyan market price is in the range of Ksh 2500 to 3500 per kilogram dried flowers provided the essential oil is 0.5% and above (Anon, 1997). This makes chamomile a high value novel crop whose successful establishment countrywide as a cash crop would give local farmers profitable employment opportunities and a much needed source of income. It would also greatly help in the industrialisation process since the essential oil is mainly used by agrobased industries.

However, the cultivation of chamomile in Kenya is at present very limited. This is mainly because of farmers' lack of awareness of the existence of the crop, inadequate knowledge on the appropriate agronomic practises, and lack of processing facilities and organised marketing systems. Hence, there is need for research to add on to the scanty information currently available on this high value crop.

1.3 Justification and objectives of the study

Throughout the history of mankind, the expanding needs for food and other agricultural products of a growing world population have been met through a combination of increased efficiency in the use of currently inhabited land, and migration and expansion of populations into new virgin territory.

Until this century, the dominant contribution to expanding agricultural production to meet growing global food demands had been the expansion of area under cropping. However, the 20th century has seen a dramatic change in the rate of growth of world demand for food. This has resulted from improved nutrition and control of human diseases leading to a decline in mortality rates while birth rates remain high, giving a rapid increase in world population. By 1900 the world population was 1.5 billion, 2.5 billion in 1950, 4 billion in 1975, 5 billion in 1987 and 6 billion in 1999. According to the United Nations Population Fund's report on the state of the world population, the world population is currently growing by slightly more than 80 million a year and is projected to be 9.4 billion by 2050 (Lever, 1982; UNFPA, 1998).

To meet these rising demands, emphasis has to be put on improving yields per hectare. The scope to expand the cropped area in densely populated countries or fragile marginal areas is limited, and often resulting in environmental degradation: deforestation in the Himalayan foothills has been cited as a contributory cause of increased flooding of the Indus, Ganges and Brahmaputra rivers. Whereas, ecological stress on the Sahara desert fringe has resulted in a steady expansion of the desert in as much as 50 Km per annum in some places (Brown and Eckholm, 1974). Infact, many areas currently under agriculture should, for ecological reasons, be returned to forestry or other conservation use.

However, within a particular husbandry system, there is no incentive for a farmer to increase his/her output through increasing levels of input use once the economic optimum has been

reached, unless input costs fall or output prices rise. The escape from this impasse lies partly in reduced input costs and partly in technological change - the introduction of new or improved means of production which increase the productivity of the other inputs and lead to lower unit costs.

Technical changes that have been used to improve crop yields include improved genetic potential for yield of a wide range of crops achieved from intensive breeding programmes; improved crop husbandry creating artificial and more beneficial environment for crop growth such as establishment of irrigation systems thereby reducing the constrain to growth imposed by water stress, and fertilizer use reducing limitations imposed by a shortage of plant nutrients; protecting yield from the ravages of weeds, pests and diseases by use of herbicides and pesticides; and improved labour productivity by using modern machinery and equipment.

In many countries though, the use of fertilizers and pesticides has reached the economic optimum and any future refinement in use of these inputs is likely to be much slower and tend to plateau. Moreover, although plant breeding during the 20th century has significantly diverted the evolutionary trend to man's advantage, the rate of change possible is severely limited by the types and extend of existing genetic variation available for exploration. In addition, modifications of crop plant biochemistry, physiology and morphology have been relatively minor.

A new initiative is therefore required to generate new technologies in the late 20th century and into the next millennium in order to sustain or increase high crop yields to support the ever increasing human population.

One unexploited area of opportunity lies in the application of plant growth regulating chemicals to positively modify crop growth to economic advantage. This approach was the subject of this study. In this study, the potential for optimization of production for profit

maximisation using benzyladenine (BA), gibberellic acid (GA), and ethephon on chamomile flowers was investigated. The principal objectives were to:

1) Evaluate the effect of BA, GA and Ethephon at varying concentrations on the growth of plants and flower yield;

2) Assess the effect of the time of application of BA on the growth of plants and flower yield;

3) Examine any interaction between applied BA concentration and timing on plant growth and flower yield.

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 Plant growth regulators (PGRs)

Plant growth regulators are organic chemical substances which when added in small concentrations (≤1mM) promote, inhibit or qualitatively modify growth and development of plants. Those occurring naturally in plants are called hormones and are usually effective at internal concentrations of 1μM or less. The main groups of plant hormones are auxins, gibberellins, cytokinins, abscisic acid, ethylene and polyamines (Salisbury and Ross, 1985).

2.1.1 Auxins

In 1926, Frits Went working in Holland discovered that some unidentified compound caused curvature of oat coleoptiles towards light (Went, 1974). Went then coined the term auxin to refer to the compound which is now known to be indole -3- acetic acid (IAA). IAA and phenylacetic acid (PAA) are the most important auxin hormones in plants. Their most notable effect include cellular elongation, phototropism and geotropism, apical dominance, root initiation, parthenocarpy, callus formation and abscission due to auxin-induced ethylene synthesis (Booth *et al.*, 1962; Wareing, 1982).

The important synthetic auxins include 1 - naphthalene acetic acid (NAA) and indole-3-butyric acid (IBA) which are more active than IAA in inducing the rooting of cuttings as they are not deactivated by IAA oxidase and therefore persists longer. 2, 4 - dichlorophenoxyacetic acid (2, 4-D), 2,4,5-trichlorophenoxyacetic acid (2,4,5-T) and 2-methyl-4-chlorophenoxyacetic acid (MCPA) are synthetic auxins which at high concentrations are used as herbicides for controlling broad-leaf weeds (Nickell, 1982; Halman, 1990).

2.1.2 Gibberellins (GAs)

In the late 1890s, Japanese farmers observed a disease in rice seedling plants attacked by the fungus *Gibberella fujikuroi*. They called it "bakanae" (foolish seedling disease) because the infected seedlings grew excessively tall and often could not support themselves and eventually died. In 1926, Kurosawa produced the bakanae effect in rice and maize seedlings by treating them with cell-free culture medium in which the fungus *Gibberella fujikuroi* had been grown. This indicated that the fungus produced a "toxin" responsible for the disease (Kurosawa, 1926). Yabuta and Sumiki (1938) at the University of Tokyo isolated the active components from culture filtrates of the fungus and named them gibberellins.

More than 60 gibberellins have now been discovered in various fungi and plants, although no single species contains more than 15 (Radley, 1958; Phinney, 1983) and most species have only a few. They are abbreviated GA with a subscript, such as GA_1, GA_2, and GA_3, to distiquish them. All could properly be referred to as gibberellic acids, but GA_3 has been studied much more than the others because of its availability, so it is commonly referred to as gibberellic acid (GA).

The most prominent effect of GAs is shoot elongation in intact plants. This response is clearly observed when GAs are applied to young plants and is caused mainly by cell elongation and partially by cell division (Wareing *et al.*, 1960; Heden, 1983). GAs can overcome dormancy in seeds and buds, induce flowering in some species of long day plants under non-inductive conditions, and cause parthenocarpic fruit development in plants such as tomatoes, cucumbers, peaches, pears, apples and grapes (Witter, 1978; Halman, 1990). Further, GAs induce male sex expression in flower development of curcubits (Zeevaart, 1976 and 1983) and activate some hydrolyases like α-amylase in the seeds of grains such as barley (Jacobsen *et al.*, 1982).

GAs are used commercially to increase the size of produced seedless grape berries and the distance between them in order to reduce fungal infections (the less tightly packed bunches are less

susceptible to fungal infections) and to increase sugar-cane growth and sugar yields (Nickell, 1976; Halman, 1990). The ability of GAs to stimulate the mobilization of food and minerals in seed storage cells of cereal grains is used by breweries to increase the rate of malting of barley (Palmer, 1977). Certain growth retardants such as chlormequat and paclobutrazol inhibit stem elongation and cause overall stunting because they inhibit gibberellin synthesis (Caldicott and Lindley, 1964). In shortening plant internodes, growth retardants reduce lodging in cereals such as barley and wheat hence allowing increased use of nitrogen fertilizers.

2.1.3 Abscisic acid (ABA)

In 1963, F.T. Addicott and his co-workers in California (studying compounds responsible for abscission of cotton fruits) and a research group led by P. F. Wareing in Wales (studying causes of dormancy of woody plants) independently discovered and chemically characterised ABA (Addicott, 1982; Wareing, 1982).

ABA is a growth inhibitor whose main physiological role in plants is that of effecting response to environmental stress and particularly as an anti-transpirant through the induction of stomatal closure under drought conditions (Jones and Mansfield, 1970; Mansfield and Davies, 1981). ABA may be involved in leaf and fruit drop directly (Cooper and Horanic, 1973; Bangerth, 1975) or indirectly by stimulating ethylene production (Sexton *et al.*, 1985). An increased ABA concentration was found in fruits and/or seeds of apples after the application of naphthaleneacetamide (NAAm), carbaryl and ethephon (Ebert and Bangerth, 1981; Treharne *et al.*, 1985) but this ABA increase was not always correlated with abscission. A direct effect of ABA in the induced abscission process, therefore, seems doubtful (Zucconi and Bukovac, 1974; Martin and Nishijima, 1979).

2.1.4 Ethylene

Since the last century, ethylene has been known to affect plant growth and metabolism. The first reports on ethylene action date back to 1858 describing early shedding of street trees caused by illuminating gas. A Russian physiologist named Neljubow (1901) was evidently the first scientist to write about ethylene action on plants reporting the triple response of etiolated pea seedlings (inhibition of stem elongation, thickening of the subtropical region, and a horizontal orientation of stems to gravity) in the presence of illuminating gas and ethylene was identified as the inducing agent. This resulted in shorter and fatter shoots bending horizontally.

Towards the end of the 19th Century, a pineapple farmer experimenting with smoke fumes to kill insects in the greenhouse discovered an earlier flowering of the pineapple plants after this treatment (Grabham, 1903; Rodriquez, 1932). The method to induce flowering of pineapples in the greenhouse by smoke fumes, then became common in the following years making it probably the first application of a PGR in agriculture. Then in 1912, the induction of fruit ripening by ethylene was recognised when H.H. Cousins advised the Jamaican government that oranges and bananas should not be stored together on ships because some unidentified volatile agent would cause the bananas to ripen premature (Moore, 1979).

Although the discovery of the properties of ethylene precedes that of other plant hormones, progress in ethylene research was initially slow because of lack of techniques to analyse the relatively small amounts of the gas in plant tissues. However, this changed when Burg and Thimann (1959) began applying newly developed analytical techniques such as gas chromatography and radioisotope tracer. Since then, interest in ethylene research has spread throughout the world.

Ethylene as a gas can only be applied in practice under certain circumstances in more or less closed systems like glasshouses or incubation chambers, limiting the usefulness of the hormone itself to very few applications. However, modern applications are mainly based on the ethylene releasing compounds 2-chloroethylphosphonic acid (ethephon) and 1-aminocyclopropane-1-carboxylic acid (ACC) which is the immediate precursor of ethylene biosynthesis (Lurssen *et al.*, 1979; Konze and Kende, 1979; Adams and Yang, 1979).

Currently, ethylene is used for stimulation of ripening of various fruits and vegetables such as tomato, pepper, melon, papaya, peach, apple, and grapes when applied shortly before harvest (Rabinowitch, *et al.*, 1970; Sims, *et al.*, 1970; Martin *et al.*, 1969). Apart from ripening of fruits, ethylene can enhance ripening (senescence) of vegetative plant parts such as sugar cane. The harvested cane has a higher sugar content and higher juice purity (Anon, 1976). Ethylene is also used to induce flowering in mangoes and Bromeliads such as pineapple and ornamental plants like *Aechmea*, *Vriesea*, etc. (Rodriquez, 1932). In rubber plantations, ethylene is used to stimulate latex flow in *Hevea* trees (Abraham *et al.*, 1968; Abraham, 1970). In cucurbits, ethylene treatment stimulates early production of female flowers and often male flower formation is completely inhibited (McMurray and Miller, 1968; Iwahori *et al.*, 1969). Ethylene induces the abscission processes in plants (Seth and Wareing, 1967). This property is of practical use in agriculture: induction of ripe fruit abscission facilitates mechanical harvest of fruits grown on trees; abscission of young fruits prevents biennial cropping hence increasing fruit size and quality; defoliation of plants facilitates mechanical harvesting of cotton by preventing coloration of the fibre by leaf pigments and also facilitating mechanical as well as hand harvesting of grapes (Cooper and Henry, 1971; Knight, 1978; Lurssen, 1982).

2.1.5 Cytokinins

Cytokinins are substituted adenine compounds that promote cell division (cytokinesis) in tissues grown *in vitro*, such as cultures from tobacco pith, carrot phloem, or soybean stems (Salsibury and Ross, 1985). The sites of cytokinin synthesis are developing seeds, leaves and root tips (Miller, 1988).

The first cytokinin to be isolated was kinetin by Miller in 1954 and Skoog *et al.*, in 1955 by thermal decomposition of DNA during autoclaving of herring sperm. Although kinetin has not so far been found in plants, related cytokinins are present in most plants. Zeatin was the first cytokinin naturally occurring in plants to be isolated. It was first identified by Letham in 1963 and almost simultaneously by Miller, both of whom used the milky endosperm of corn, *Zea mays* (Miller and Skoog, 1957; Letham, 1963). The synthetic cytokinin, benzyladenine (BA), is closely related to kinetin and zeatin in carrying a side group at the 6-aminopurine position.

The predominant effects of cytokinins in plants are stimulation of cell division and enlargement, and the delaying of senescence (Mooney and Van Staden, 1986). In addition, cytokinins seem to play an important role in the regulation of plant growth under drought, in combination with ABA. Studies on the effects of the application of BA and moisture stress on the sponge gourd (*Luffa cylindrica*) indicated that BA was antagonistic to moisture stress (Virk *et al.*, 1985). In maize, stomatal closure caused by drought may be overcome by external application of cytokinins (Davies *et al.*, 1986). In rice cultivation, kinetin was found to ameliorate the injury of drought treatment when applied after re-irrigation (Moody, 1986).

Studies have shown that spraying genetic male flowers of certain plants with BA, the flowers become feminized and develop into phenotypically female flowers (Delaigue *et al.*, 1986).

Cytokinins are also important in the induction of greening and the initiation of the development of chloroplasts. BA has been shown to activate synthesis of two proteins of the chloroplasts - RUBP carboxylase and chlorophyll a/b protein complex (Funckees-Shippy and Levine, 1985).

Cytokinins have also been shown to have a senescence-retarding effect, exhibited by greater retention of chlorophyll and protein contents. This has a positive effect on cereal grain filling (Jung, 1984) and is useful in increasing flower longevity in cut-flowers (Van Staden and Joughin, 1988).

Cytokinins activate shoot induction in various plants. A recent important application of BA is for the propagation *in vitro* of apple rootstocks. Presence of BA in the medium (0.5-20 mg/L) causes a marked increase in the number of apple shoots (Dunstan *et al.*, 1985).

"Spray and pray" experiments provide considerable evidence that applied hormones, particularly cytokinins and ABA, can exert great effects on assimilate transport which, in some instances, can lead to enhanced crop productivity (Weaver and Johnson, 1985). This is mainly due to influencing of both loading and unloading of assimilates across the membrane boundaries of the vascular tissues (Hayes and Patrick, 1985; Clifford *et al.*, 1986). Foliar application of cytokinins on fruit trees before the bloom increases fruit size (Mauck *et al.*, 1986; Nickel, 1985). Particularly favourable results occur by simultaneous application of cytokinins and gibberellic acid. This increase in nutrient sink activity results in higher yields.

Studies with externally applied radioactive cytokinins have shown that cytokinins may be metabolised through any of the following ways: Irreversible degradation by the enzyme "cytokinin oxidase" which oxidizes the side chain double bond leading to loss of biological activity; irreversible conjugation with sugars or amino acids leading to loss of, or reduced biological activity; reversible conjugation leading to compounds which may in themselves posses biological activity or function as storage forms of active cytokinins.

2.2 The discovery and development of PGRs

The practical uses of synthetic PGRs emerged with the discovery in 1893 of the induction by smoke of pineapple plants to bloom out of season as a result of an accidental fire (Grabham, 1903). To the suprise of the grower, the plants burst into flower instead of being damaged. The ethylene in the smoke induced flower bud initiation and development. Subsequently an ethylene releasing chemical, ethephon (ethrel), was later marketed for use in commercial pineapple production.

The study of PGRs is to a large extend an offspring of herbicide research. By far the greatest use of any synthetic PGR was developed in the 1940s with the discovery of the auxin-type herbicides, 2, 4-D and MCPA (Blackman, 1945; Audus, 1972). The lethal effects of these compounds on many broad-leaved weeds when applied at high concentrations culminated in a major success story in weed control technology where they still enjoy widespread usage mainly in cereal crops (Audus, 1972; Kirby, 1980; Garrod, 1982).

PGRs have been used commercially since the 1950s to modify growth, enhance crop yield, alter harvest patterns and improve mechanical harvesting (Cibulsky and Crovetti, 1981). Although there is extensive use of PGRs in agronomic crops such as grains and sugar cane, the primary use of PGRs is in horticultural crops (Emongor, 1995). The first group of synthetic growth regulators to be used for horticultural purposes was derived from the discovery of the auxin - type plant hormones in the 1930s, synthetic IBA and NAA which were used to promote rooting of cuttings, prevent fruit drop in apples and control biennial bearing of fruit trees and fruit size through their effects on fruit thinning (Audus, 1972: Garrod, 1982; Halman, 1990).

Presently, major applications of growth regulators are in the sugar industry, in which PGRs are used on a commercial basis at almost every stage of development of the crop to increase the recoverable yield of sucrose in sugar cane (Nickell, 1983). Another interesting application is that

of substances causing pollen suppression (gametocides) used in hybrid breeding programmes to prevent self-fertilization in self-pollinating species (Jung, 1986). Growth regulators are also used to stimulate differentiation in the cloning of cells in tissue culture (Reynolds, 1987; Caruso, 1987; Griesbach, 1987).

2.3 World use of PGRs and their future potential

The six most widely used plant growth regulators, accounting for more than 90% of the total global use, are chlormequat, daminozide, maleic hydrazide, ethephon, gibberellic acid and glyphosine (Hoad, 1982).

Chlormequat/cycocel (CCC) is a growth retardant used commercially since 1960s in shortening internodes hence reducing or even eliminating lodging in cereals such as barley and wheat (Linser and Kuhn, 1962; Mayr *et al*, 1962; Caldicott and Lindley, 1964).

Daminozide is a growth retardant commonly used to control shape and growth rates of many ornamental species including *Chrysanthemum*, *Hydrangea* and *Poinsettia*. Daminozide was also used in preventing pre-harvest drop, promoting firmness and quality of pome fruits but was removed from the market in 1986 because of consumer awareness and dissatisfaction over the use of chemicals in food products (Miller, 1988).

Maleic hydrazide is a growth retardant, initially used as an herbicide, but later employed to control growth of certain trees and shrubs and to prevent sprouting of onions and potatoes in storage. Its most extensive use is in inhibiting the development of suckers on tobacco plants.

Ethephon is an ethylene-releasing compound. It is used in the ripening, senescence and abscission phenomena in various crops and also in promotion of flower initiation in pineapples, modification of sex expression in cucumbers and the stimulation of latex flow in rubber trees.

Gibberellins have been used to increase yield and quality of seedless table grapes, delaying ripening of citrus fruits on the trees, increasing fruit set in pears and by breweries to increase the rate of malting of barley (Palmer, 1977).

Glyphosine was first marketed in 1973 for use as a commercial sugar ripener. Sugar cane ripening is the maximising sucrose and minimising all other soluble solids at harvest (Nickel, 1977). This is achieved by restricting the growth of the cane in the few weeks prior to harvest and allowing the accumulation of sucrose in the storage tissue of the stem.

Many of the existing uses of PGRs are highly profitable to the grower. In Hawaiian sugar cane, maleic hydrazide reduce or prevent flowering, an effective suppression of tassel formation giving a 15% increase in yield. Gibberellic acid applied during the colder winter months of the growing season has been shown to give a gain of 0.6 to 1.5 t/ha sugar. Chemical ripeners such as glyphosine can add a further 10-15% to the sucrose yield (Nickel, 1976). In Western Europe and North America, cycocel (CCC) is widely used to shorten and stiffen wheat straw, allowing increased use of nitrogen fertilizers in intensive, high input high yield agricultural systems.

Plant growth regulators (PGRs) have been used to promote rooting and propagation of plants using plant parts; initiate or terminate the dormancy of seeds, buds and tubers; control of the development of lateral shoots; suppress unwanted vegetative growth, preventing lodging, dwarfing ornamental species, etc; control of the size, shape and colour of crops grown for the processed food industries; regulate the chemical composition of plants so enhancing quality; induce or retard senescence; promote, delay, or prevent flowering; control sex expression for breeding; gametocidal action as an aid to plant breeders in the development of hybrid cultivars; control fruit set, further development and ripening; defoliate and desiccate crops such as cotton to facilitate machine harvesting; induce abscission to facilitate picking of fruits such as citrus; prevent post-harvest spoilage; increase plant resistance to pests; enhance plant resistance to environmental stress

factors; regulate chemical composition of plants and colour of fruits; influence mineral uptake from soil; and change the timing of crop development (Nickel, 1982; Miller, 1988; Emongor, 1995). PGRs have also been used on various medicinal and spice plants such as *Origanum majorana*, *Catharanthus roseus*, *Matricaria chamomilla*, and sweet basil to increase vegetative growth, flower yield and improve quality of the medicinal and aromatic products of the plants (Abou-Zeid and El-Sherbeeny, 1970, 1974; El-Antably *et al.*, 1975; Mousa and El-Emary, 1983; Sadowska *et al.*, 1983; Meawad *et al.*, 1984).

Each of the above applications of PGRs can yield a profound economic advantage to the producers, packers and consumers of a crop. However, the use of PGRs in agriculture has lagged behind the widespread application of herbicides and pesticides (Audus, 1972; Nickell, 1982 and 1985). The main reasons for this are the differences in sensitivity of different plant species or even cultivars of the same crop to given PGR treatment so preventing easy predictions of the biological effects, high costs for screening plant growth regulating activities, and the problem of potential toxic residues on food crops which require lengthy and costly testing (Wareing, 1982; Halman, 1990). All these reasons have made agrochemical companies hesitant to invest large amounts of capital in the development of PGRs hence restricting their use to highly specialized, predominantly horticultural situations.

Nevertheless, it is anticipated that a generation of growth regulators with novel chemical and biological activity could achieve commercial success similar to that enjoyed by the early herbicides. The first chemicals with consistent yield-enhancing properties in major crops are still eagerly awaited and their success in world agriculture when they are released is assured.

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 Experimental site

The experiment was conducted at the University of Nairobi, Kibwezi irrigation farm for two seasons between February and December, 1996. The farm is located about 250 km South East of Nairobi at an altitude of 800 m above sea level on the intersection of latitude 02° 17'S and longitude 38° 02'E.

This is a dryland area with unreliable and often inadequate rainfall for rainfed agriculture. The mean monthly totals of precipitation shows a bimodal distribution of rainfall averaging 600 mm per annum with peaks occurring in March-April and November-December. Mean monthly maximum and minimum temperatures are 30°C and 17°C, respectively, with a range of 13°C (Appendix 1). The details of radiation, evaporation, rainfall, humidity, and temperature during the experimental period are presented in appendix 2a and 2b.

According to FAO-UNESCO (1988) soil classification, the soils in the farm are of two types: chromic luvisols and haplic lixisols. These are developed on basement rocks of sedimentary origin derived from the Mozambique belt system. The main rock type is the sandstone rich in ferromagnesian minerals (Saggerson, 1963; Walsh, 1963). The soils are well drained, deep to very deep, red to dark reddish brown, sandy clay to clay loam with rock outcrops. They are hard when dry, friable when moist and sticky and plastic when wet with patchy clay cutans on some ped faces. The relief is flat to very gently undulating with slope topography between 1% and 5%. Ekirapa and Muya (1991) in their detailed soil survey of the farm found that the soils are deficient in all plant nutrients and especially nitrogen, potassium, magnesium and manganese, but have marginally adequate supplies of phosphorus. The soil reaction is neutral to slightly alkaline with a

pH between 6.4 to 7.9. The soils have moderate to very high infiltration rates (4-40 cm/hr), low water holding capacity and a high bulky density and are best irrigated by sprinkler or drip irrigation systems.

The crop was grown using sprinkler irrigation and water from the Kibwezi river. The water is non-sodic but has medium salinity hazards (appendix 3). However, it can be successfully used for irrigation if accompanied by proper soil and water management practises like provision of good drainage and use of salt tolerant crops such as chamomile (Singh, 1970).

3.2 Experimental treatments and design

In the first season (February-June 1996), a 4 x 2 factorial experiment was laid down as a split-plot in a randomized complete block design with four replications. The treatments comprised of four rates (0, 25, 50, and 75 mg/litre) of benzyladenine (BA) applied at two different times (4 and 6 weeks after transplanting - WAT) during crop growth. In each block, the timing of application of BA was independently randomized to make the main plots. The treatments of BA concentration were then randomized within each main plot to make the sub-plots.

In the second season (August-December 1996), the experiment was repeated and the effect of two more PGRs (GA and ethephon) investigsted. A simple randomized complete block design with four replications was used. The treatments comprised four rates (0, 100, 200, 300 mg/litre) of GA and four rates (0, 50, 100, 150 mg/litre) of ethephon. The PGRs were applied 6 WAT.

The experimental plots measured 3 m x 4 m and all trials used whole chamomile plants with plant growth regulators (PGRs) sprayed to run-off. The chamomile variety Bohemia whose seed was procured from Yugoslavia was used in the experiment. The crop was grown at a spacing of 40 cm between rows and 30 cm between plants to give a population of 83,334 plants per hectare. Each plot had 10 rows and 10 plants per row with two guard rows on each side.

3.3 Crop husbandry

A nursery measuring 1 m x 18 m was prepared and appropriate amounts of heat-treated manure (20 t/ha), D.A.P. (40 kg P_2O_5/ha) and C.A.N. (35 kg N/ha) fertilizers, and a nematicide (Nemacur at 130 kg/ha) incorporated into the soil and the surface levelled. A shade was then made over the nursery and chamomile seeds sown by broadcasting on the surface. The nursery was watered using mist sprayers (microsprinklers) for 30 minutes twice a day.

Spraying was done twice every week against attack by insect pests and fungal diseases by using an alternating combination of one insecticide among Karate (1000 ml/ha), Brigade (500 ml/ha), and Dimethoate-40 (1500 ml/ha) and one fungicide among Benlate (500 g/ha), Ridomil (3000 g/ha) and Dithane (2000 g/ha) in 400 litres of water per hectare. Two weeks before transplanting, the shade was removed. One week later, the frequency and amount of watering were gradually reduced in order to harden the seedlings.

Two weeks before transplanting, the field was mowed, subsoiled and then ploughed. Harrowing and then rotavation to a fine tilth were done one week before transplanting.

The crop was transplanted after 4 weeks in the nursery. Just before transplanting, the field was irrigated for 4 hours. Transplanting was done late in the afternoon hence giving the seedlings favourable conditions over the night for initial adjustment to field conditions thereby reducing seedling mortality. Light irrigation for 1 hour was then done every day for the next 10 days and gapping done during this period.

Two days after transplanting, phosphorus was added (band application) as D.A.P. (18:46:0) at a rate of 40 kg P_2O_5/ha. Nitrogen was top dressed as C.A.N. (26% N) two weeks after transplanting at a rate of 17 kg N/ha and repeated four weeks later. In total, 50 kg N/ha was applied. BA was applied at 4 and 6 WAT while GA and ethephon were applied 6 WAT.

Spraying in the field was done as in the nursery at twice weekly intervals for the control of aphids, whiteflies, stem rot, and powdery mildews. The crop was kept weed free by hand cultivation (hoeing between rows and within rows) throughout the growing season.

3.4 Determination of crop growth

The growth variables measured were plant height, spreading line, number of tillers and the number of leaves. Sampling for these measurements begun 6 WAT and continued after every two weeks until 12 WAT. At the start of taking measurements, 10 plants per plot were randomly sampled and tagged. All subsequent measurements were taken on these same plants.

At 12 WAT, three plants from each plot were selected at random and harvested for the determination of dry matter production and biomass partitioning.

3.5 Determination of flower yield

The number of days from sowing to flowering (when 50% of the plants have at least one flower open) was noted and the first harvesting of flowers done then. Subsequent harvests were done once every week.

For each plot, flower yield determination was done in an area of 1 m x 1 m randomly selected and marked out. After every harvest, the average flower diameter was measured from 10 flowers randomly sampled from each plot. The fresh flower weight was also determined. The flowers were then air-dried under shade to constant moisture content and the dry flower weight determined.

The number of days from sowing to withering (when 50% of the plants had withered) was noted and used to indicate the end of harvesting.

3.6 Statistical analysis

Analysis of variance (ANOVA) was performed on each of the growth and yield variables using the general linear models (proc glm) procedure of the Statistical Analysis System (SAS) programme package. Linear and quadratic orthogonal polynomials were tested and appropriate multiple regression models used to examine the nature of the response to BA, GA and ethephon concentrations and timing of application (Snedecor and Cochran, 1989; Steel and Torie, 1980). Multiple comparisons among means was done using protected Least Significant Difference (LSD) test at P=0.05. Proc univariate procedure was carried out on residuals to support the assumptions of normality made by the researcher.

CHAPTER FOUR

4.0 RESULTS

4.1 Effect of benzyladenine (BA), gibberellic acid (GA) and ethephon on vegetative growth of chamomile plants

4.1.1 Plant height

Spraying chamomile plants with BA significantly increased their height compared to the plants which were not sprayed (Table 1). The different concentrations of BA (25, 50 and 75 mg/litre) increased plant height by similar margins at maturity. In both seasons, the response to increasing BA concentration was quadratic.

The timing of BA application had no effect on the height of the plants in both seasons. However, there was an interaction between BA concentrations and the timing of BA application in both seasons (Table 2). In season one, application of 75 mg/litre BA 6 WAT produced the tallest plants while in season two, application of 25 mg/litre BA 4 WAT produced the tallest plants.

Spraying GA significantly increased the height of the plants (Table 4) and the response was cubic. Plants treated with 100 mg/litre were the tallest. However, there were no differences between GA treatments in respect to plant height (Table 4). Ethephon application had no effect on the height of the plants (Table 5).

Table 1: Effect of benzyladenine on the height and spreading ability of chamomile plants at maturity (10 WAT)

Benzyladenine (mg/litre)	Plant height (cm)		Plant spread (cm)	
	Season 1	Season 2	Season 1	Season 2
0	51.35[a]	50.43[a]	72.42	71.75
25	54.93[b]	53.63[b]	72.57	72.93
50	55.47[b]	53.89[b]	71.95	73.10
75	55.18[b]	52.00[ab]	71.87	71.38
Significance	L***, Q***	L***, Q***	ns	ns
LSD (P=0.05)	2.08	1.90	3.26	2.13

The response was linear (L) or quadratic (Q)

***, ns, Significant within columns at P=0.005 or nonsignificant, respectively.

Means followed by the same letter(s) within columns are not significantly different according to the protected Least Significant Difference (LSD) test at P=0.05.

Table 2: Interaction between benzyladenine concentration and timing of application on the height of chamomile plants at maturity (10 WAT)

Benzyladenine (mg/litre)	Plant height (cm)			
	Season 1		Season 2	
	4 WAT	6 WAT	4 WAT	6 WAT
0	51.47[a]	52.23[ab]	51.50[bc]	49.36[a]
25	54.30[bc]	55.27[c]	55.30[c]	51.95[bc]
50	55.33[c]	55.00[c]	53.33[cd]	54.05[de]
75	52.93[ab]	57.43[d]	53.30[cd]	50.70[ab]
Significance	*	*	*	*

LSD for interaction is 2.08 and 1.90 for season 1 and 2, respectively
*, Significant within columns at P=0.05.
Means followed by the same letter(s) within columns are not significantly different according to the protected Least Significant Difference (LSD) test at P=0.05.

Table 3: Effect of time of application of benzyladenine on the spread of chamomile plants at maturity (10 WAT)

Time of application	Plant spread (cm)	
	Season 1	Season 2
Before flowering	71.22	73.09
During flowering	73.18	71.49
Significance	ns	ns

ns = nonsignificant within columns at P=0.05.

Table 4: Effect of gibberellic acid on the height and spreading ability of chamomile plants at maturity (10 WAT)

Gibberellic acid (mg/litre)	Plant height (cm)	Plant spread (cm)
0	51.25[a]	63.90
100	57.35[b]	65.25
200	56.25[b]	58.35
300	56.73[b]	55.90
Significance	L*, Q*, C*	ns
LSD (P=0.05)	3.35	7.57

The response was linear (L), quadratic (Q), or cubic (C).

*, ns, Significant within columns at P=0.05 or nonsignificant, respectively.

Means followed by the same letter(s) within columns are not significantly different according to the protected Least Significant Difference (LSD) test at P=0.05.

Table 5: Effect of ethephon on the height and spreading ability of chamomile plants at maturity (10 WAT)

Ethephon (mg/litre)	Plant height (cm)	Plant spread (cm)
0	46.40	59.00[a]
50	50.70	65.75[b]
100	52.25	69.40[b]
150	49.05	69.20[b]
Significance	ns	L***
LSD (P=0.05)	5.70	5.30

The response was linear (L).

***, ns, Significant within columns at P=0.005 or nonsignificant, respectively.

Means followed by the same letter(s) within columns are not significantly different according to the protected Least Significant Difference (LSD) test at P=0.05.

4.1.2 Spreading of plants

BA treatments and time of application had no effect on the spreading of the plants (Table 1 and 3). Similarly, GA application had no effect on plant spread, although higher GA concentrations (200 and 300 mg/litre) exhibited reduced plant spread (Table 4).

Ethephon spraying caused increased vegetative spread of the plants (Table 5). The response was linear to increasing ethephon concentration. There were no differences between the ethephon concentrations used (50, 100, 150 mg/litre).

4.1.3 Tillering ability

Spraying chamomile plants with BA increased the tillering ability measured as number of tillers per plant (Table 6). The response of chamomile plants to increasing BA concentration was quadratic in season 1 and cubic in season 2 (Table 6). In season 1, there were no BA differences in response to tillering ability. However, in season 2, plants sprayed with 25 mg/litre BA, produced higher number of tillers than those sprayed with 50 or 75 mg/litre BA. There was no difference between 50 and 75 mg/litre BA. The timing of BA application had no effect on the tillering ability of the plants (Table 7) and there was no interaction between BA concentrations and the timing of application in both seasons. Treating chamomile plants with GA or ethephon at various concentrations had no effect on their ability to produce tillers (Table 9, 10).

Table 6: Effect of benzyladenine on the tillering ability and leaves of chamomile plants at maturity (10 WAT)

Benzyladenine (mg/litre)	Tillers per plant		Leaves per plant	
	Season 1	Season 2	Season 1	Season 2
0	18.27[a]	18.94[a]	447.32[a]	388.75[a]
25	19.52[b]	21.88[c]	485.90[b]	430.50[b]
50	19.73[b]	20.13[b]	500.52[b]	418.80[ab]
75	19.48[b]	19.55[ab]	495.27[b]	407.58[ab]
Significance	L*,Q*	Q***, C***	L***,Q***	Q***
LSD (P=0.05)	1.13	0.47	27.26	36.17

The response was linear (L), quadratic (Q), or cubic (C).

*,***, Significant within columns at P=0.05, 0.005, respectively.

Means followed by the same letter(s) within columns are not significantly different according to the protected Least Significant Difference (LSD) test at P=0.05.

Table 7: Effect of time of application of benzyladenine on the tillering ability of chamomile plants at maturity (10 WAT)

Time of application	Tillers per plant	
	Season 1	Season 2
Before flowering	18.92	19.76
During flowering	19.57	20.49
Significance	ns	ns

ns = nonsignificant within columns at P=0.05.

Table 8: Interaction between benzyladenine concentration and timing of application on the leaves of chamomile plants at maturity (10 WAT)

Benzyladenine (mg/litre)	Number of leaves/plant			
	Season 1		Season 2	
	4 WAT	6 WAT	4 WAT	6 WAT
0	424.07^{a}	470.57^{bc}	368.00^{a}	409.50^{b}
25	456.53^{b}	515.27^{d}	424.60^{bc}	436.40^{bc}
50	483.37^{c}	517.17^{d}	407.55^{b}	430.05^{bc}
75	511.80^{d}	478.73^{bc}	366.40^{a}	448.75^{c}
Significance	**	**	*	*

LSD for interaction is 27.26 and 36.17 for season 1 and 2, respectively
*,**, Significant within columns at P=0.05 or P=0.01, respectively.
Means followed by the same letter(s) within columns are not significantly different according to the protected Least Significant Difference (LSD) test at P=0.05.

Table 9: Effect of gibberellic acid on the tillering ability and number of leaves of chamomile plants at maturity (10 WAT)

Gibberellic acid (mg/litre)	Tillers/plant	Leaves/plant
0	20.50	391.35
100	21.50	395.40
200	18.80	353.95
300	19.20	361.60
Significance	ns	ns

ns = nonsignificant within columns at P=0.05

Table 10: Effect of ethephon on the tillering ability and number of leaves of chamomile plants at maturity (10 WAT)

Ethephon (mg/litre)	Tillers/plant	Leaves/plant
0	21.10	356.55
50	20.10	358.65
100	21.35	391.45
150	21.00	392.40
Significance	ns	ns

ns = nonsignificant within columns at P=0.05

4.1.4 Number of leaves

Chamomile plants sprayed with BA had higher number of leaves per plant than those that were not sprayed (Table 6). For both seasons, the response to increasing BA concentration was quadratic. However, in all seasons there were no differences between BA concentrations. In season 2, only plants sprayed with 25 mg/litre had higher leaf number per plant than those untreated (Table 6).

Timing of BA application had no effect on the number of leaves per plant. However, there was an interaction between BA concentrations and the time of their application (Table 8). Application of either 25 or 50 mg/litre BA 6 WAT in season 1 increased the number of leaves per plant (Table 8). However, plants sprayed with 75 mg/litre BA were not different from the control in reference to number of leaves per plant (Table 8).

GA application had no effect on leaf number per plant, although, higher GA concentrations (200 or 300 mg/litre) tended to reduce leaf number (Table 9). Ethephon application did not influence the number of leaves per plant (Table 10).

4.1.5 Dry matter accumulation and partitioning

BA application significantly increased dry matter accumulation (shoot, root and shoot/root ratio) by the plants compared to the control plants (Table 11). The response to BA treatment was cubic. The timing of application had no effect on dry matter accumulation but there was an interaction between BA concentrations and the timing of their application (Table 12).

Table 11: Effect of benzyladenine on dry matter accumulation of chamomile plants at 12 WAT

Benzyladenine (mg/litre)	Dry matter accumulation (g/3 plants)		
	Shoot	Root	Shoot:Root ratio
0	180.15^{a}	18.65^{a}	9.87^{c}
25	214.65^{c}	22.65^{b}	10.18^{c}
50	197.80^{b}	22.90^{b}	8.69^{a}
75	204.65^{bc}	22.00^{b}	9.36^{b}
Significance	L**, Q***, C***	L**, Q***	L**, C***
LSD	13.79	2.20	0.50

The response was linear (L), quadratic (Q), or cubic (C).

,*, Significant within columns at P=0.01, 0.005, respectively.
Means followed by the same letter(s) within columns are not significantly different according to the protected Least Significant Difference (LSD) test at P=0.05.

Table 12: Interaction between benzyladenine concentration and timing of application on the dry matter accumulation of chamomile plants at 12 WAT.

Benzyladenine (mg/litre)	Dry matter accumulation (g/3 plants)					
	Shoot		Root		Shoot:Root ratio	
	4 WAT	6 WAT	4 WAT	6 WAT	4 WAT	6 WAT
0	210.00^{a}	150.30^{a}	22.50^{bc}	14.80^{a}	9.55^{b}	10.18^{c}
25	200.00^{a}	228.80^{c}	16.00^{a}	29.30^{c}	12.50^{c}	7.85^{a}
50	196.30^{a}	199.30^{b}	23.50^{c}	22.30^{b}	8.38^{a}	9.00^{b}
75	200.80^{a}	208.50^{b}	21.00^{b}	23.00^{b}	9.63^{b}	9.08^{b}
Significance	**	**	**	**	**	**

LSD for interaction is 13.79, 2.20 and 0.5 for the shoot, root and shoot:root ratio, respectively.
**, Significant within columns at P=0.01.
Means followed by the same letter(s) within columns are not significantly different according to the protected Least Significant Difference (LSD) test at P=0.05.

Spraying BA 6 WAT increased dry matter production by the shoots compared to the control plants, with the lowest concentration (25mg/litre) used giving the highest dry matter accumulation (Table 12). However, BA application 4 WAT had no effect on the plant dry matter accumulation (Table 12).

Spraying the plants with BA increased dry matter production by the roots (Table 11) and the response to increasing BA concentration was quadratic. There were no differences between BA treatments in respect to root dry matter accumulation. The timing of application had no effect on dry matter accumulation but there was an interaction between BA concentrations and the timing of their application (Table 12). Chamomile plants sprayed with BA 6 WAT had high dry matter accumulation in the roots, but plants sprayed with 25 mg/litre BA had the highest dry matter accumulation (Table 12). However, there were no differences between 50 and 75 mg/litre BA in response to root dry matter accumulation 6 WAT, but 25 mg/litre BA was significantly different from 50 and 75 mg/litre BA (Table 12).

Chamomile plants sprayed with 50 or 75 mg/litre BA partitioned more dry matter to the roots than to the shoots as indicated by their smaller shoot:root ratio compared to the untreated control plants (Table 11). 25 mg/litre BA increased dry matter partitioning to the shoots compared to the higher BA concentrations. The higher BA concentrations (50 and 75 mg/litre) reduced the shoot:root ratio (Figure 1). The timing of application had no effect on shoot:root dry matter accumulation but there was an interaction between BA concentrations and the timing of application (Table 12). Spraying BA 6 WAT increased the relative accumulation of dry matter into the roots than into the shoots (Table 12). Application of 25 mg/litre BA caused the highest relative partitioning of dry matter to the roots whereas 50 and 75 mg/litre were not significantly different from each other (Table 12). The effect of BA treatment 4 WAT on dry matter partitioning was not evident.

Table 13 shows that the plants sprayed with 50 mg/litre ethephon had a significant increase in dry matter production by the shoots and roots compared to the control. The higher ethephon concentrations (100 and 150 mg/litre) had no effect on shoot and root dry matter accumulation. Similarly, application of GA had no effect on the shoot dry matter accumulation by the plants (Table 14). However, GA significantly reduced dry matter accumulation to the roots (Table 14). GA reduced root dry matter accumulation in a linear fashion.

Plants sprayed with ethephon significantly reduced the shoot:root ratio and the reduction was cubic to increasing ethephon concentration (Table 13). 100 or 150 mg/litre ethephon were not different in their effect on reducing the shoot:root ratio. 50 mg/litre ethephon significantly reduced the shoot:root ratio compared to 100 or 150 mg/litre ethephon (Table 13).

Plants sprayed with 200 or 300 mg/litre GA increased the shoot:root ratio (Table 14). However, 100 mg/litre GA had no effect on the plant's shoot:root ratio (Table 14).

Table 13: Effect of ethephon on dry matter accumulation of chamomile plants at 12 WAT

Ethephon (mg/litre)	Dry matter accumulation (g/3 plants)		
	Shoot	Root	Shoot:root ratio
0	210.00[a]	22.00[a]	9.50[c]
50	255.75[b]	34.75[b]	7.51[a]
100	233.75[a]	26.25[a]	8.49[b]
150	199.75[a]	23.75[a]	8.22[b]
Significance	Q**	Q**, C**	L*,Q*,C
LSD (P=0.05)	35.91	5.88	0.87

The response was linear (L), quadratic (Q), or cubic (C).
*,**,***. Significant within columns at P=0.05, 0.01, 0.005, respectively.
Means followed by the same letter(s) within columns are not significantly different according to the protected Least Significant Difference (LSD) test at P=0.05.

Table 14: Effect of gibberellic acid on dry matter accumulation of chamomile plants at 12 WAT

Gibberellic acid (mg/litre)	Dry matter accumulation (g/3 plants)		
	Shoot	Root	Shoot:root ratio
0	194.00	23.75[c]	8.38[a]
100	162.75	18.50[b]	8.85[a]
200	203.75	17.00[ab]	12.06[b]
300	192.25	13.50[a]	14.34[b]
Significance	ns	L**	L***
LSD (P=0.05)	35.88	4.76	2.43

The response was linear (L).
,*, ns, Significant within columns at P=0.01, 0.005, or nonsignificant, respectively.
Means followed by the same letter(s) within columns are not significantly different according to the protected Least Significant Difference (LSD) test at P=0.05.

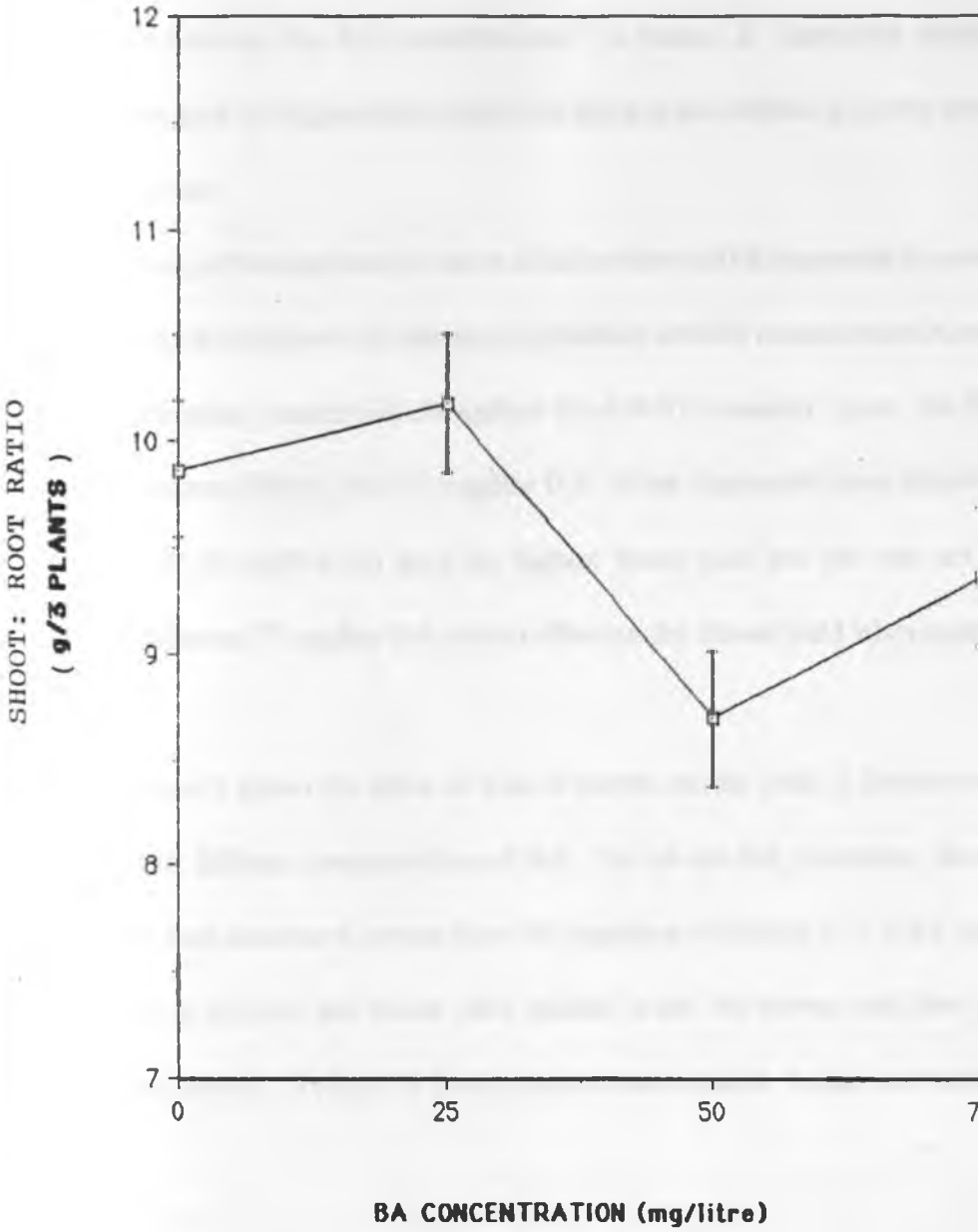

Figure 1: Effect of benzyladenine (BA) on the shoot:root ratio measured as drymatter at 12 weeks after transplanting

4.2 Effect of BA, GA and ethephon on dry flower yield and flower diameter of chamomile

BA application significantly increased the yield of chamomile flowers in both seasons of the experimental period compared to the untreated control plants (Table 15). A test for orthogonal polynomials showed that the response to BA treatment was cubic. In season 1, there were no differences between the BA concentrations. In season 2, chamomile plants treated with 25 mg/litre BA gave the highest flower yield, but this was not different from the yield of plants treated with 50 mg/litre.

Timing of BA application had no effect on the yield of chamomile flowers. However, there was an interaction between the timing of application and BA concentration in season 2 (Table 16). Chamomile plants treated with 50 mg/litre BA 6 WAT in season 2 gave the highest flower yield but this was not different from 25 mg/litre BA. In the chamomile plants treated with BA 4 WAT in season 2, 25 mg/litre BA gave the highest flower yield but this was not different from 50 mg/litre. Whereas 75 mg/litre BA had no effect on dry flower yield when applied 4 WAT (Table 16).

Figure 2 shows the effect of time of harvest on the yield of flowers of chamomile plants treated with different concentrations of BA. For all the BA treatments, flower yield increased steadily in each successive harvest from the beginning of picking at 7 WAT upto 10 WAT. The increase then reduced and flower yield peaked in the 5th harvest and then declined with each subsequent harvest. A total of 8 flower harvests were done at weekly intervals.

Table 15: Effect of benzyladenine on total dry flower yield of chamomile

Benzyladenine (mg/litre)	Dry flower yield (kg/ha)	
	Season 1	Season 2
0	2850.00[a]	3286.30[a]
25	3291.70[b]	4426.30[c]
50	3270.00[b]	4337.50[bc]
75	3223.30[b]	4010.00[b]
Significance	L**,Q***, C***	L**,Q***,C*
LSD (P=0.05)	110.60	345.80

The response was linear (L), quadratic (Q), or cubic (C).
*, **, ***, significant within columns at P=0.05, 0.01, 0.005, respectively.
Means followed by the same letter(s) within columns are not significantly different according to the protected Least Significant Difference (LSD) test at P=0.05.

Table 16: Interaction between benzyladenine concentration and time of application on total dry flower yield of chamomile

Benzyladenine (mg/litre)	Dry flower yield (kg/ha)			
	Season 1		Season 2	
	4 WAT	6 WAT	4 WAT	6 WAT
0	2796.70	2903.30	3722.50[a]	2850.00[a]
25	3320.00	3263.30	4422.50[c]	4430.00[bc]
50	3293.30	3246.70	4165.00[bc]	4510.00[c]
75	3190.00	3256.70	3860.00[ab]	4160.00[b]
Significance	ns	ns	**	**

LSD for interaction in season 2 was 345.80.
**, Significant within columns at P=0.01.
Means followed by the same letter(s) within columns are not significantly different according to the protected Least Significant Difference (LSD) test at P=0.05.

Plants treated with 100 and 150 mg/litre ethephon had significantly higher total flower yield than the untreated control plants (Table 17). The response to ethephon treatment was linear. 50 mg/litre ethephon had no effect on total dry flower yield. Figure 3 shows that for all the ethephon treatments, flower yield increased in each successive harvest from the beginning of picking, peaked at the 5th harvest, and subsequently declined upto the end of harvesting. A total of 8 flower harvests were done at weekly intervals.

Chamomile plants sprayed with GA significantly reduced the flower yield when compared to the untreated control plants (Table 18). The highest reduction in yield was in plants treated with 300 mg/litre GA but this was not different from 200 mg/litre GA. A test for orthogonal polynomials showed that the response to GA treatment was quadratic. Figure 4 shows that the yield of flowers increased in each successive harvest from the beginning of picking, peaked at the 4th harvest, and subsequently declined upto the end of harvesting. A total of 8 flower harvests were done at weekly intervals.

Spraying chamomile plants with BA, ethephon, or GA had no effect on flower diameter (Table 19, 20).

Table 17: Effect of ethephon on the total dry flower yield (kg/ha) of chamomile

Ethephon rate (mg/litre)	Dry flower yield (kg/ha)
0	2922.50[a]
50	3620.00[ab]
100	4152.50[b]
150	4265.00[b]
Significance	L***,
LSD (P=0.05)	701.10

The response was linear (L).
***, significant within column at P=0.005.
Means followed by the same letter(s) within columns are not significantly different according to the protected Least Significant Difference (LSD) test at P=0.05.

Table 18: Effect of gibberellic acid on the total dry flower yield (kg/ha) of chamomile

Gibberellic acid (mg/litre)	Dry flower yield (kg/ha)
0	3887.50[c]
100	2895.00[b]
200	2487.50[a]
300	2337.50[a]
Significance	L***,Q**
LSD (P=0.05)	379.30

The response was linear (L) or quadratic (Q).
, *, significant within columns at P=0.01, 0.005 respectively.
Means followed by the same letter(s) are not significantly different according to the protected Least Significant Difference (LSD) test at P=0.05.

Table 19: Effect of benzyladenine on the mean flower diameter of chamomile

Benzyladenine (mg/litre)	Mean flower diameter (mm)	
	Season 1	Season 2
0	9.11	8.89
25	9.28	9.04
50	9.41	8.93
75	9.29	9.04
Significance	ns	ns

ns = nonsignificant within columns at P=0.05

Table 20: Effect of ethephon and gibberellic acid on the mean flower diameter of chamomile

Ethephon (mg/litre)	Mean flower diameter (mm)	Gibberellic acid (mg/litre)	Mean flower diameter (mm)
0	8.48	0	8.54
50	8.68	100	8.31
100	8.78	200	8.52
150	8.70	300	8.46
Significance	ns		ns

ns = nonsignificant within columns at P=0.05.

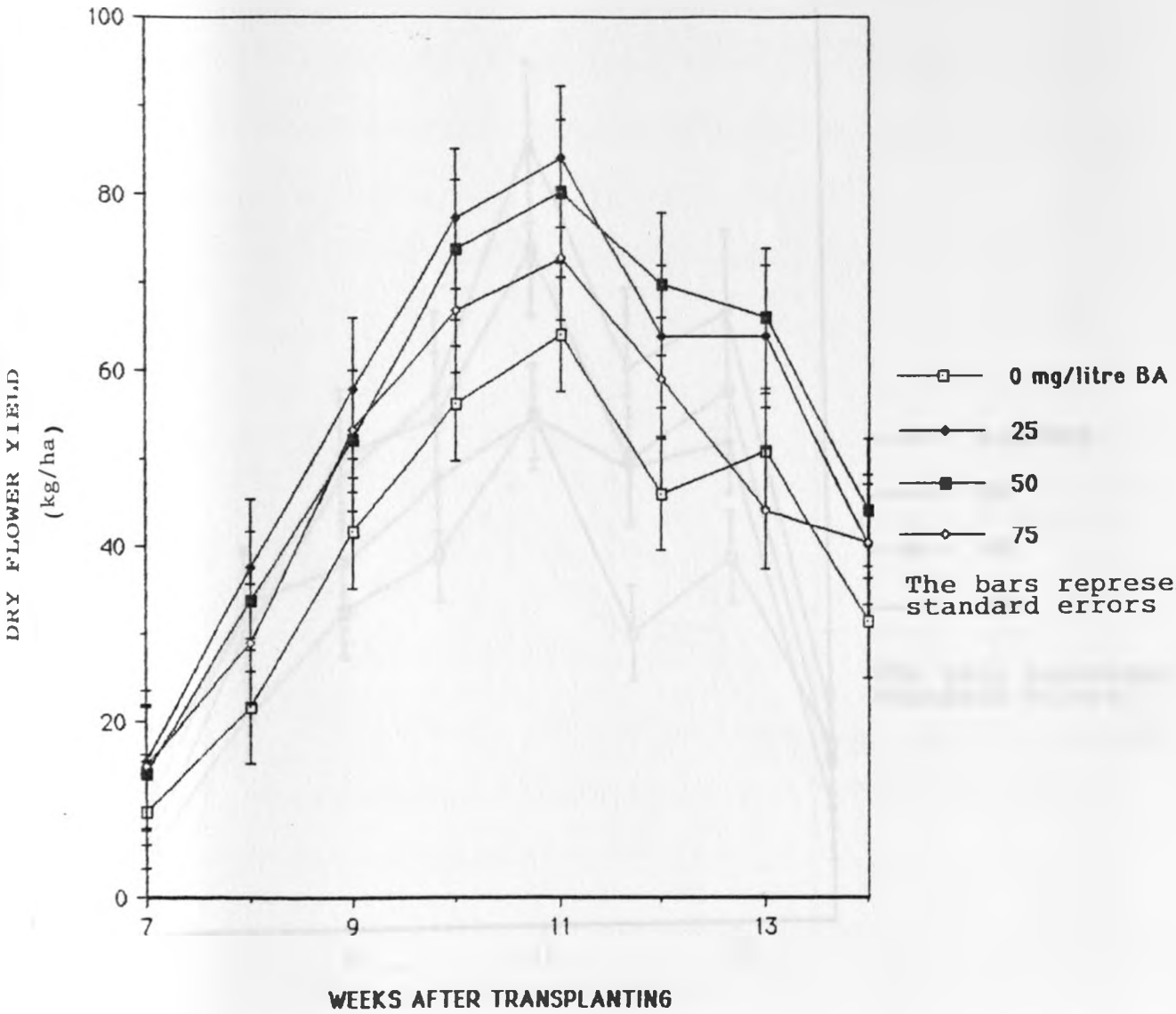

Figure 2: Effect of benzyladenine on the dry flower yield of chamomile

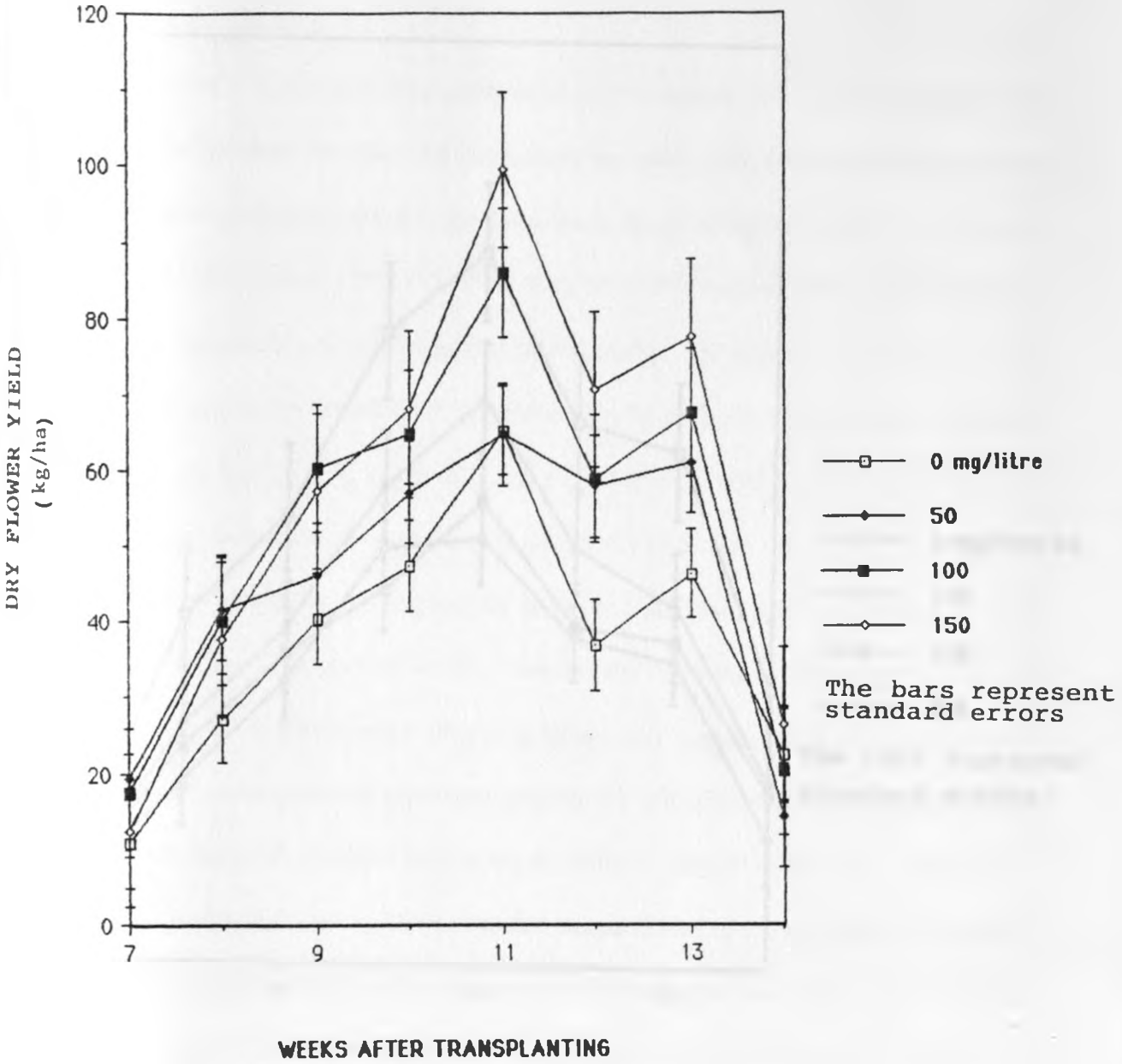

Figure 3: Effect of ethephon on the dry flower yield of chamomile

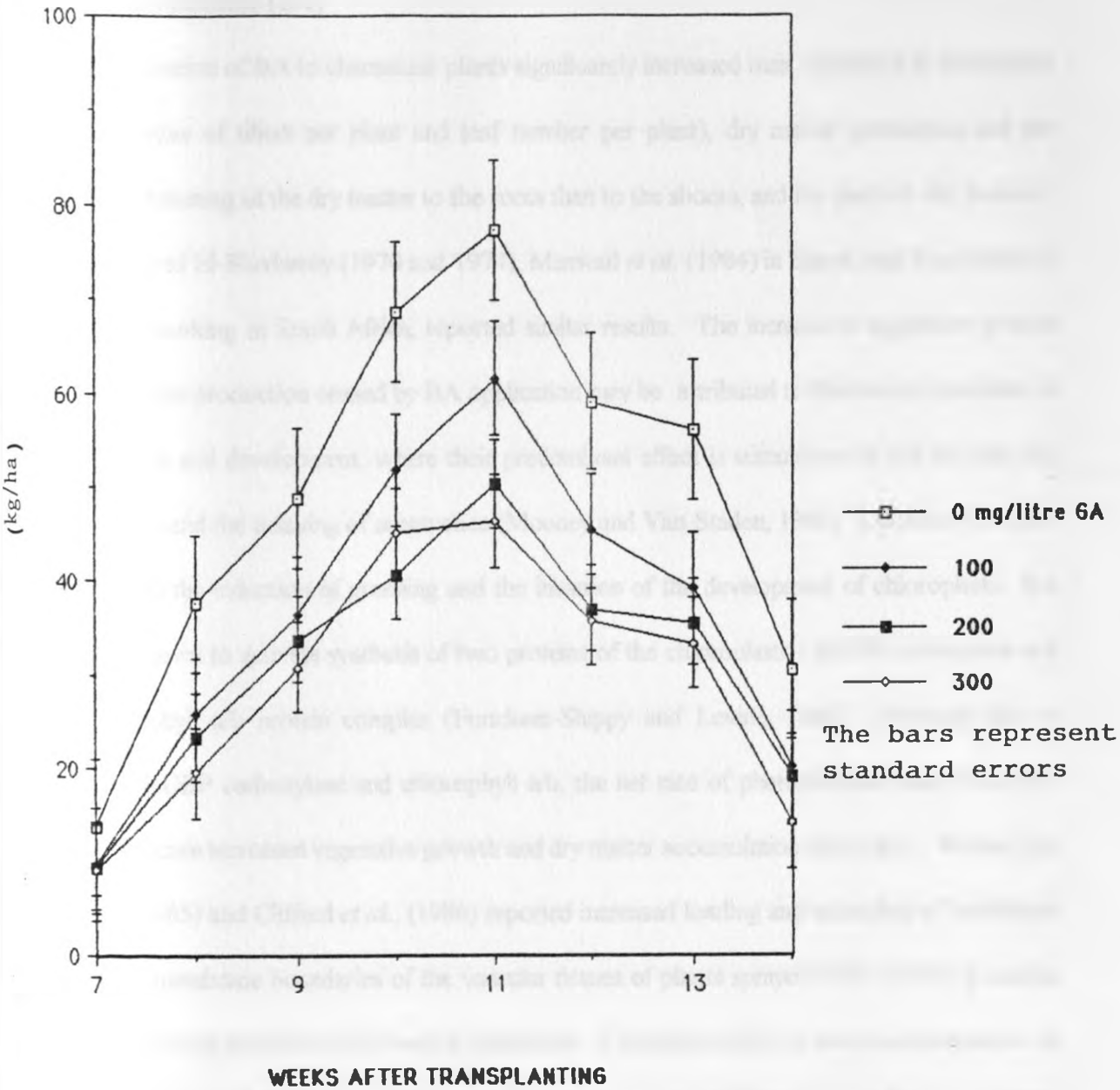

Figure 4: Effect of gibberellic acid on the dry flower yield of chamomile

5.0 DISCUSSION

5.1 Benzyladenine (BA)

Application of BA to chamomile plants significantly increased their vegetative growth (plant height, number of tillers per plant and leaf number per plant), dry matter production and the relative partitioning of the dry matter to the roots than to the shoots, and the yield of dry flowers. Abou-zeid and El-Sherbeeny (1970 and 1974), Meawad *et al.* (1984) in Egypt, and Van Staden *et al.* (1986) working in South Africa, reported similar results. The increase in vegetative growth and dry matter production caused by BA application may be attributed to the role of cytokinins in plant growth and development, where their predominant effect is stimulation of cell division and enlargement and the delaying of senescence (Mooney and Van Staden, 1986). Cytokinins are also important in the induction of greening and the initiation of the development of chloroplasts. BA has been shown to activate synthesis of two proteins of the chloroplasts - RUBP carboxylase and the chlorophyll a/b protein complex (Funckees-Shippy and Levine, 1985). Probably due to increased RUBP carboxylase and chlorophyll a/b, the net rate of photosynthesis may have been increased hence increased vegetative growth and dry matter accumulation due to BA. Weaver and Johnson (1985) and Clifford *et al.*, (1986) reported increased loading and unloading of assimilates across the membrane boundaries of the vascular tissues of plants sprayed with cytokinins leading to enhanced crop growth and dry matter production. Cytokinins and IAA have been implicated as antagonists in apical dominance and lateral branching, respectively. Wickson and Thimann (1958) demonstrated that kinetin applications can remove IAA inhibition, thereby increasing lateral branching in poinsettia (Carpenter and Becker., 1972; Milbroker, 1972) and inducing bottom

breaks of roses (Parups, 1971). Therefore, it is suggested that BA overcame apical dominance in chamomile plants hence the increase in tillering ability.

Timing of BA application had no effect on chamomile plants, but spraying 6 WAT tended to increase vegetative growth and yield of flowers compared to plants sprayed 4 WAT. These results compare with those of Katsumi (1962) and Jeffcoat (1977) who found that early application of BA to carnations and chrysanthemums increased the mass of carnation flowers and chrysanthemum inflorescence but did not increase floret number in chrysanthemum. The tendency of increased activity of BA sprayed during flowering (6 WAT) compared to spraying before flowering (4 WAT) may have been due to differences in the endogenous levels of cytokinins in the chamomile plants. This result would be in agreement with work by various workers who have reported that the endogenous levels of different plant hormones vary with the stage of development of the plant (Nickell, 1976; Wareing, 1982; Knee, 1985; Jung, 1986) and that external applications of PGRs have greatest response when endogenous hormone levels are limiting.

The increased flower production by plants sprayed with BA may be attributed to their increased vegetativeness. Taller plants with more branches and leaves are able to photosynthesize more and hence more assimilates are available for distribution to the various sinks such as the flowers and seeds. Moreover, BA application increased dry matter production by the plants and a relative partitioning of the dry matter to the roots. This apparent increase in root growth could have been important in yield development by ensuring adequate provision of water and nutrients to the plant thereby avoiding any limitations to photosynthesis. Since BA treatments had no effect on the flower size, then the observed increase in flower yield must have been caused by production of more flowers per plant as opposed to larger flowers. In addition to increased assimilate production resulting from more plant vegetativeness, a direct BA effect on flowering cannot be

ruled out. BA increased both vegetative growth and dry flower yield in this study, suggesting a direct BA effect on flowering and vegetative growth, negating the theory that plant growth regulators affect flowering indirectly by reducing vegetative growth (Miller, 1988). Cytokinins have been shown to promote carbohydrate metabolism and create new source-sink relationships (Mothes and Engelbretcht, 1961; Dyer *et al.*, 1990) thus leading to increased sink strength hence more dry matter accumulation in the sink (flower).

5.2 Ethephon

Application of ethephon to chamomile plants significantly increased plant spread and the yield of dry flowers. Increasing concentrations of ethephon tended to cause more spreading of chamomile plants and higher total flower yield. Plants sprayed with 50 mg/litre ethephon significantly increased dry matter production of shoots and roots. However, 100 or 150 mg/litre ethephon had no effect on dry matter production.

The increase in flower yield caused by ethephon application may be attributed indirectly to the increased vegetative growth resulting in increased photosynthesis as indicated by the increased dry matter production and consequently making more assimilates available for translocation to the various sinks (flowers, seeds, roots, etc.). Meawad (1981) and Meawad *et al.*, (1984) in Egypt working on gladiolus and chamomile, respectively found that ethephon increased carbohydrate synthesis as well as nitrogen content and consequently promoted vegetative growth and flowering. Ethephon may have enhanced flower yield by a direct effect of ethylene on the chamomile plants. Ethylene is known to promote flower bud initiation and flowering. For a long time ethylene has been known to induce flowering in pineapple (Mwaule, 1983; Lurssen and Konze, 1985; Knee, 1985) and other bromeliads like *Aechmea victoriana*, L. (De Greef, 1983). The direct effect of ethephon on chamomile flower formation is further supported by the observation that ethephon

had no effect on flower size hence indicating that the observed increase in flower yield must have been due to increased number of flowers per plant. The increase in flower yield occurred soon after application of ethephon between 7 WAT and 12 WAT but there was no effect towards the end of the growing season (13 WAT and 14 WAT). This implies that ethephon effects decreased over time from the date of application. The relative partitioning of dry matter to the roots than to the shoots ensured adequate supply of water and mineral nutrients to the plants hence preventing limitations to growth.

5.3 Gibberellic acid (GA)

GA treatment significantly increased the height of chamomile plants but had no effect on dry matter production by the shoots. The most prominent effect of GA in intact plants is shoot elongation. This response is clearly observed when GA is applied to young plants and is caused mainly by cell elongation and partially by cell division (Wareing *et al.*, 1960; Heden, 1983). The increased height of GA treated chamomile plants may have been caused by more stem elongation and succulence with no increased assimilate accumulation.

As opposed to BA and ethephon treatments, GA application significantly reduced dry matter production of the roots and the relative partitioning of the dry matter to the roots, and also reduced the yield of dry flowers. The reduction in root growth and flower yield, increased with increasing concentrations of gibberellic acid. Eid and Ahmed (1976) and Sadowska *et al.*, (1983) reported that GA lowered the dry weight of sweet basil (*Ocimum basilicum* L.) and *Catharanthus roseus* L. plants. Mousa and El-Emary (1983) working with sweet basil which is a very fragrant herb used as seasoning and making of liquors, found that each increase in GA concentration (50, 100 and 200 mg/litre) upto the high level considerably reduced plant height and number of branches per plant as well as the total yield of the herb. However, other authors have reported that

GA increased the yield or dry weight of various aromatic and ornamental plants such as *Origanum marjorana*, *Viola odorata*, *Ocimum sanctum*, and Phlox (Gulati *et al.*, 1974; El-Antably *et al.*, 1975; Mousa, 1979; and Mohamed *et al.*, 1983).

The reduction in yield of chamomile flowers caused by GA treatment may be attributed to the decreased vegetative growth and especially the inhibition of root growth observed in plants treated with GA. Gibberellins are known to increase hydrolysis of starch, fructans and sucrose, which are the principle components of dry weight (Salisbury and Ross, 1985). In agricultural research, the differences in response or sensitivity of different plant species or even cultivars to given PGR treatment has been widespread and this has prevented easy predictions of the biological effects of using various PGRs in crop production. The results of GA application reported in this study, which were largely unexpected, demonstrate this problem.

CONCLUSIONS AND RECOMMENDATIONS

The study showed that application of BA and ethephon 6 WAT enhanced plant growth and increased yield of dry flowers. Consequently, BA and ethephon can be used in the cultivation of chamomile with special emphasis to cost-effectiveness with respect to the purchase price, cost of transportation and ease of storage, cost of application, and availability of each.

Treatment of chamomile plants with GA inhibited root growth and decreased the yield of dry flowers. While GA increased plant height, there was no corresponding increase in shoot dry matter production. The reduction in yield increased with higher concentrations of GA upto the higher level used (100, 200 and 300 mg/litre).

Based on the results of this study, it is recommended that:

1. BA and ethephon can be incorporated into the management of chamomile plants to improve flower yield. The plants should be sprayed to run-off 6 WAT with 25 mg/litre BA and 100 mg/litre ethephon.
2. The study be repeated in several other locations in the country to refine and confirm the findings reported in this study.
3. The experimental scope be widened and further research carried out to establish the combined effect of multiple applications of different concentrations of BA and ethephon over the entire growing season and also find out whether these have any effect on the essential oil of chamomile.
4. The potential for GA application in chamomile production be investigated further using concentrations lower than 100 mg/litre GA.
5. A multidisciplinary collaboration between crop scientists, chemists and the industrial users be established to forge a united approach in harnessing the existing potential of chamomile production in Kenya. This mutually beneficial partnership would remove the current

marketing constraints where production is hampered by a limited demand of the flowers mainly as herbal tea by a few expatriate consumers.

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APPENDICES

Appendix 1: Mean climatic data for Kibwezi during 1991-1995:

Month	Total rainfall mm	Evaporation mm	Max. Temp. °C	Min Temp. °C	Radiation $MJ/m^2/day$	RH %	Daily windrun km
January	45	4.3	28.9	17.9	456.2	58	72.80
February	30	5.3	31.6	18.5	506.4	52	86.30
March	81	5.8	32.5	19.4	502.6	51	96.10
April	113	5.1	31.8	18.9	503.9	52	101.90
May	30	4.7	28.4	17.5	446.8	56	120.90
June	2	5.4	28.9	15.5	401.6	51	129.80
July	1	5.3	28.3	14.2	355.8	51	143.40
August	1	5.8	28.5	14.8	398.8	51	154.30
September	2	6.6	30.1	14.2	440.6	51	157.50
October	30	6.7	31.6	17.1	466.3	50	146.84
November	160	4.9	29.8	19.4	442.6	50	91.02
December	115	3.6	28.2	19.0	458.7	58	71.44
	50.8	5.3	29.9	17.2	448.4	52.6	114.36

Source: University of Nairobi, Meteorological Station, Kibwezi.

Appendix 2a: Mean climatic data for Kibwezi during 1996:
(Source: University of Nairobi's meteriological Station Kibwezi)

Month	Total rainfall mm	Max. Temp °C	Min. Temp °C	Wind run km/day	Radiation $MJ/M^2/day$
January	11.0	31.6	19.0	77.4	479.2
February	51.4	32.9	19.9	104.1	469.7
March	107.3	32.9	20.3	101.4	444.0
April	23.7	31.2	18.7	101.9	493.0
May	40.9	29.8	17.3	135.4	391.1
June	2.5	28.0	16.0	134.8	323.9
July	0.0	27.5	15.3	147.7	319.4
August	0.0	28.2	14.9	158.0	397.0
September	0.0	29.7	16.7	158.0	433.8
October	0.0	30.3	17.7	158.4	472.9
November	168.9	30.2	19.4	113.3	442.6
December	2.3	29.0	17.0	82.9	505.2
	34.0	30.1	17.7	122.8	430.9

Appendix 2b: Mean climatic data for Kibwezi during 1996:

(Source: Kibwezi Irrigation Project (K.I.P.)

Month	Total rainfal mm	Evaporation mm	Maximum temp. °C	Minimum temp. °C	R.H. %	Wind Speed Km/hr
January	2.0	5.9	29.8	8.8	-	2.7
February	59.2	6.4	25.7	7.1	70.3	2.6
March	109.9	4.9	24.0	8.0	73.6	2.3
April	39.0	4.7	24.3	7.2	63.2	2.1
May	63.0	4.3	20.7	5.8	70.5	2.3
June	1.5	3.7	20.0	4.5	51.6	2.1
July	0.0	3.7	19.6	3.6	49.4	2.2
August	0.0	4.6	18.1	3.0	41.4	2.5
September	0.0	5.6	21.0	4.3	43.3	3.2
October	0.0	6.4	21.9	4.9	38.6	3.1
November	193.0	4.3	16.3	5.0	37.3	1.8
December	0.0	5.3	22.0	3.5	56.5	2.2
	39.0	4.9	22.0	5.5	54.2	2.4

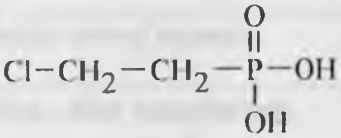
Appendix 3: Chemical characteristics of irrigation water from Kibwezi river

(Source: A.E. Ekirapa and E.M. Muya (1991).

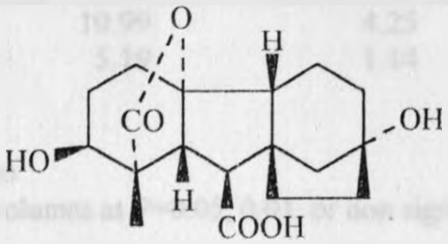
Component	Quantity	
pH	8.8	
EC	650	Micro Siemens/cm
Na^+	3.48	m.e./litre
K^+	1.18	"
Ca^{++}	0.42	"
Mg^{++}	1.83	"
CO_3^-	2.00	"
HCO_3-	0.07	"
Cl^-	0.10	"
SO_4^{2-}	0.46	"
SAR	3.30	

Appendix 4: Chemical structures of 2-chloroethylphosphonic acid (ethephon), gibberellic acid (GA_3) and benzyladenine (BA)

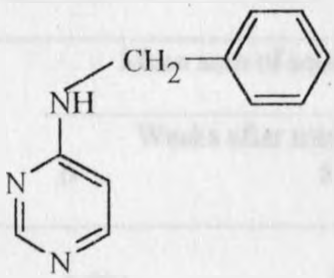
2-Chloroethylphosphonic acid (ethephon)
(Ethephon decomposes spontaneously in aqueous solution and in plant tissues to yield ethylene and phosphoric acid).



Gibberellic acid (GA_3)



Benzyladenine (BA)



Appendix 5: Effect of benzyladenine (BA) and its time of application (TA) on the height of chamomile plants (Season 1) - ANOVA table.

Source	df	Mean sum of squares		
		Weeks after transplanting		
		6	8	10
Block	2	74.117	2.154	21.791
TA	1	6.00^{ns}	1.084^{ns}	11.759^{ns}
Error a	2	65.274	17.577	22.239
BA	3	29.247**	25.047**	17.063**
TA.BA	3	2.247^{ns}	2.215**	7.737*
Error b	12	3.640	0.318	1.371
CV_a		10.99	4.25	4.34
CV_b		5.19	1.14	2.15

df = degrees of freedom
CV = % coefficient of variation
*,**, ns = Siginificant within columns at P=0.05, 0.01, or non significant, respectively.

Appendix 6: Effect of benzyladenine (BA) and its time of application (TA) on the height of chamomile plants (Season 2) - ANOVA table.

Source	df	Mean sum of squares		
		Weeks after transplanting		
		6	8	10
Block	3	1.721	2.162	5.506
TA	1	25.920^{ns}	6.125^{ns}	30.225^{ns}
Error a	3	41.287	33.504	32.614
BA	3	20.027**	11.211**	20.654**
TA.BA	3	4.758^{ns}	0.779^{ns}	5.066*
Error b	18	1.722	0.661	1.231
CV_a		9.37	6.38	5.44
CV_b		3.83	1.79	2.11

df = degrees of freedom.
CV = % coefficient of variation
*,**, ns = Significant within columns at P=0.05, 0.01, or non significant, respectively.

Appendix 7: Effect of ethephon on the height of chamomile plants - ANOVA table.

Source	df	Mean sum of squares			
		Weeks after transplanting			
		6	8	10	12
Block	3	11.483	15.857	0.647	12.500
Treatment	3	21.163^{ns}	6.537^{ns}	16.967^{ns}	25.033^{ns}
Error	9	6.121	13.548	12.302	12.680
CV		6.48	8.41	7.37	7.29

df = degrees of freedom.
CV = % coefficient of variation
ns = Significant at P=0.05.

Appendix 8: Effect of gibberellic acid on the height of chamomile plants - ANOVA table.

Source	df	Mean sum of squares			
		Weeks after transplanting			
		6	8	10	12
Block	3	49.137	0.719	0.752	6.524
Treatment	3	137.417**	146.632**	60.009*	31.337^{ns}
Error	9	18.517	8.347	11.440	4.385
CV		10.35	5.64	6.43	3.35

df = degrees of freedom.
CV = % coefficient of variation
*,**, ns = Significant within columns at P=0.05, 0.01, or non significant, respectively.

Appendix 9: Effect of benzyladenine (BA) and its time of application (TA) on the spread of chamomile plants (Season 1) - ANOVA table

Source	df	Mean sum of squares		
		Weeks after transplanting		
		6	8	10
Block	2	1.196	30.751	42.798
TA	1	0.921^{ns}	12.041^{ns}	23.207^{ns}
Error a	2	80.448	6.888	16.124
BA	3	2.492^{ns}	6.146^{ns}	0.710^{ns}
TA.BA	3	3.014^{ns}	4.028^{ns}	3.281^{ns}
Error b	12	1.023	1.964	3.366
CV_a		11.74	2.48	2.78
CV_b		2.65	2.23	2.54

df = degrees of freedom.
CV = % coefficient of variation
ns = Non significant at P=0.05.

Appendix 10: Effect of benzyladenine (BA) and its time of application (TA) on the spread of chamomile plants (Season 2) - ANOVA table.

Source	df	Mean sum of squares		
		Weeks after transplanting		
		6	8	10
Block	3	18.692	11.233	3.500
TA	1	22.445^{ns}	15.123^{ns}	20.475^{ns}
Error a	3	18.414	24.805	37.432
BA	3	9.784^{ns}	3.123^{ns}	5.833^{ns}
TA.BA	3	16.029^{ns}	14.908^{ns}	5.598^{ns}
Error b	18	7.299	2.907	2.055
CV_a		5.51	4.55	4.23
CV_b		6.94	3.11	1.98

df = degrees of freedom.
CV = % coefficient of variation
ns = Not significant at P=0.05.

Appendix 11: Effect of ethephon on the spread of chamomile plants - ANOVA table.

Source	df	Mean sum of squares			
		Weeks after transplanting			
		6	8	10	12
Block	3	2.246	6.449	3.660	1.462
Treatment	3	8.057*	48.069**	183.713*	94.342**
Error	9	1.712	4.270	31.187	10.965
CV		3.25	3.87	8.73	5.03

df = degrees of freedom.
CV = % coefficient of variation
*,** = Significant at P=0.05, 0.01, respectively.

Appendix 12: Effect of gibberellic acid on the spread of chamomile plants - ANOVA table.

Source	df	Mean sum of squares			
		Weeks after transplanting			
		6	8	10	12
Block	3	3.570	6.94	25.394	14.000
Treatment	3	15.902ns	46.820ns	100.421ns	31.337ns
Error	9	4.306	13.409	26.222	22.380
CVs		5.83	7.36	8.63	7.77

df = degrees of freedom.
CV = % coefficient of variation
ns = Not significant at P=0.05.

Appendix 13: Effect of benzyladenine (BA) and its time of application (TA) on the tillering ability of chamomile plants (Season 1) - ANOVA table.

Source	df	Mean sum of squares		
		Weeks after transplanting		
		6	8	10
Block	2	1.092	1.932	2.164
TA	1	3.840^{ns}	0.021^{ns}	2.407^{ns}
Error a	2	7.426	5.652	9.178
BA	3	0.806^{*}	1.744^{**}	2.652^{*}
TA.BA	3	0.387^{ns}	0.255^{ns}	0.083^{ns}
Error b	12	0.228	0.202	0.535
CV_a		9.61	6.53	7.87
CV_b		3.33	3.47	3.80

df = degrees of freedom.
CV = % coefficient of variation
*,**, ns = Significant at P=0.05, 0.01, or non significant, respectively.

Appendix 14: Effect of benzyladenine (BA) and its time of application (TA) on the tillering ability of chamomile plants (Season 2) - ANOVA table.

Source	df	Mean sum of squares		
		Weeks after transplanting		
		6	8	10
Block	3	4.161	2.912	2.570
TA	1	3.512^{ns}	1.488^{ns}	4.278^{ns}
Error a	3	5.261	1.832	16.459
BA	3	5.398^{**}	2.409^{**}	12.809^{*}
TA.BA	3	1.284^{*}	0.098^{ns}	0.391^{ns}
Error b	18	0.305	0.165	0.200
CV_a		7.95	4.06	10.08
CV_b		3.83	2.44	2.23

df = degrees of freedom.
CV = % coefficient of variation
*, **, ns = Significant at P=0.05, P=0.01 or non significant, respectively.

Appendix 15: Effect of ethephon on the tillering ability of chamomile plants - ANOVA table.

Source	df	Mean sum of squares			
		Weeks after transplanting			
		6	8	10	12
Block	3	0.900	1.002	2.209	2.169
Treatment	3	0.367^{ns}	3.289^{ns}	2.209^{ns}	1.189^{ns}
Error	9	1.613	3.103	1.900	3.049
CV		8.64	10.67	6.56	8.36

df = degrees of freedom.
CV = % coefficient of variation
ns = Not significant at P=0.05.

Appendix 16: Effect of gibberellic acid on the tillering ability of chamomile plants - ANOVA table.

Source	df	Mean sum of squares			
		Weeks after transplantin			
		6	8	10	12
Block	3	5.060	1.102	2.180	6.300
Treatment	3	4.567^{ns}	1.689^{ns}	0.460^{ns}	6.107^{ns}
Error	9	2.280	1.514	2.978	8.309
CV		10.60	7.83	9.46	14.41

df = degrees of freedom.
CV = % coefficient of variation
ns = Not significant at P=0.05.

Appendix 17: Effect of benzyladenine (BA) and its time of application (TA) on the leaves of chamomile plants (Season 1) - ANOVA table.

Source	df	Mean sum of squares		
		Weeks after transplanting		
		6	8	10
Block	2	635.095	6619.017	149.180
TA	1	3128.193^{ns}	220.853^{ns}	4171.203^{ns}
Error a	2	529.549	6767.295	6763.327
BA	3	558.696**	1857.951**	3473.530**
TA.BA	3	68.118^{ns}	79.089^{ns}	2516.672*
Error b	12	38.463	531.092	234.838
CV_a		5.07	8.16	9.85
CV_b		2.73	4.57	3.18

df = degrees of freedom.
CV = % coefficient of variation
*,**, ns = Significant at P=0.05, 0.01, or non significant, respectively.

Appendix 18: Effect of benzyladenine (BA) and its time of application (TA) on the leaves of chamomile plants (Season 2) - ANOVA table.

Source	df	Mean sum of squares		
		Weeks after transplanting		
		6	8	10
Block	3	874.630	7227.835	4682.865
TA	1	300.170^{ns}	3490.313^{ns}	12505.693^{ns}
Error a	3	6646.073	51284.917	17233.461
BA	3	1672.063**	12309.835**	2525.922*
TA.BA	3	1507.300**	5529.071*	1930.944^{ns}
Error b	18	152.935	1095.495	592.697
CV_a		16.85	27.82	15.95
CV_b		5.11	8.13	5.92

df = degrees of freedom.
CV = % coefficient of variation
*,**, ns = Significant at P=0.05, 0.01, or non significant, respectively.

Appendix 19: Effect of ethephon on the leaves of chamomile plants - ANOVA table.

Source	df	Mean sum of squares			
		Weeks after transplanting			
		6	8	10	12
Block	3	1104.053	1400.474	3244.100	371.890
Treatment	3	2239.233^{ns}	5831.314^{ns}	1749.380^{ns}	1574.483^{ns}
Error	9	746.617	3420.881	970.358	2076.009
CV		10.89	20.89	7.49	12.16

df = degrees of freedom.
CV = % coefficient of variation
ns = Non significant at P=0.05.

Appendix 20: Effect of gibberellic acid on the leaves of chamomile plants - ANOVA table.

Source	df	Mean sum of squares			
		Weeks after transplanting			
		6	8	10	12
Block	3	4543.991	25245.007	5880.161	250.000
Treatment	3	2074.694^{ns}	2458.773^{ns}	3981.892^{ns}	1748.200^{ns}
Error	9	1328.985	4930.797	6450.774	5454.283
CV		15.30	19.58	22.24	

df = degrees of freedom.
CV = % coefficient of variation
ns = Non significant at P=0.05.

Appendix 21: Effect of benzyladenine (BA) and its time of application (TA) on the dry matter production and partitioning in chamomile plants at 12 weeks after transplanting - ANOVA table.

Source	df	Mean sum of squares		
		Shoot	Root	Shoots:root ratio
Block	3	1098.458	11.031	1.035
TA	1	215.313^{ns}	19.532^{ns}	7.802^{ns}
Error a	3	2891.687	47.031	3.707
BA	3	1688.792**	31.115**	3.382**
TA.BA	3	2886.352**	154.281**	12.537**
Error b	18	77.960	2.115	0.125
CV_a		13.49	15.93	10.11
CV_b		4.43	6.75	3.71

df = degrees of freedom.

CV = % coefficient of variation

**, ns = Significant at P=0.01 or non significant, respectively.

Appendix 22: Effect of ethephon on the dry matter production of chamomile plants at 12 weeks after transplanting - ANOVA table.

Source	df	Mean sum of squares	
		Shoot	Root
Block	3	1623.230	46.879
Treatment	3	2512.730*	127.729**
Error	9	504.062	13.507
CV		9.99	13.77

df = degrees of freedom.
CV = % coefficient of variation
*,** = Significant at P=0.05, 0.01, respectively.

Appendix 23: Effect of gibberellic acid on the dry matter production of chamomile plants at 12 weeks after transplanting - ANOVA table.

Source	df	Mean sum of squares	
		Shoot	Root
Block	3	96.230	4.396
Treatment	3	1252.730ns	72.562**
Error	9	503.284	8.840
CV		11.92	16.35

df = degrees of freedom.
CV = % coefficient of variation
**, ns = Significant at P=0.01 or non significant, respectively.

Appendix 24: Effect of ethephon on dry matter partitioning of chamomile plants at 12 weeks after transplanting (Shoot:root ratio) - ANOVA table.

Source	df	SS	MSS	F
Block	3	5.224	1.741	
Treatment	3	8.142	2.714	9.138**
Error	9	2.677	0.297	
CV	6.46			

df = degrees of freedom.
CV = % coefficient of variation
** = Significant at P=0.01.
ss = Sum of squares
MSS = Mean of squares

Appendix 25: Effect of gibberellic acid on dry matter partitioning of chamomile plants at 12 weeks after transplanting (Shoot:root ratio) - ANOVA table.

Source	df	SS	MSS	F
Block	3	7.870	2.623	
Treatment	3	94.845	31.615	13.752**
Error	9	20.688	2.299	
CV	13.90			

df = degrees of freedom.
SS = Sum of squares
MSS = Mean sum of squares
CV = % coefficient of variation
** = Significant at P=0.01.

Appendix 26. Effect of benzyladenine (BA) and its time of application (TA) on the dry flower yield of chamomile (Season 1) ANOVA table

Source	df	Mean sum of squares						
		Weeks after transplanting						
		7	8	9	10	11	12	13
Block	2	4.875	60.875	21.500	6.500	24.500	23.292	26.000
TA	1	0.042^{ns}	7.042^{ns}	0.042^{ns}	10.667^{ns}	192.667^{ns}	126.042^{ns}	135.375^{ns}
Error a	2	16.792	130.792	112.667	32.667	88.667	59.542	39.500
BA	3	43.597^{**}	74.931^{**}	48.819^{**}	15.222^{**}	24.444^{**}	33.375^{**}	33.597^{**}
TA.BA	3	2.486^{*}	5.819^{ns}	3.153^{ns}	3.666^{ns}	5.777^{ns}	15.153^{**}	3.819^{ns}
Error b	12	0.500	6.333	3.195	1.361	1.862	1.472	2.750
CV_a		14.23	18.93	13.00	6.41	8.05	8.21	8.18
CV_b		4.25	7.22	3.79	2.27	2.02	2.24	3.74

df = degrees of freedom.
CV = % coefficient of variation
*,**, ns = Significant at P=0.05, P=0.01 or non significant, respectively.

Appendix 27: Effect of benzyladenine (BA) and its time of application (TA) on the dry flower yield of chamomile (Season 2) - ANOVA table.

Source	df	Mean sum of squares						
		Weeks after transplanting						
		7	8	9	10	11	12	13
Block	3	11.917	24.208	44.198	147.709	115.365	73.583	76.040
TA	1	12.500^{ns}	144.500^{ns}	108.782^{ns}	3.120^{ns}	11.283^{ns}	3.125^{ns}	180.495^{ns}
Error a	3	101.417	147.583	208.364	292.210	430.781	358.375	245.252
BA	3	53.000**	375.875**	381.865**	695.373**	625.115**	814.833**	377.040**
TA.BA	3	15.500*	19.083*	28.031^{ns}	96.043^{ns}	242.364**	316.792**	354.418**
Error b	18	3.333	5.285	12.671	9.820	19.573	54.535	13.174
CV_a		37.85	19.96	14.11	12.49	13.79	15.87	12.82
CV_b		13.52	7.55	6.96	4.58	5.88	12.38	5.94

df = degrees of freedom.
CV = % coefficient of variation
*,**, ns = Significant at P=0.05, P = 0.01, or non significant, respectively.

Appendix 28: Effect of ethephon on the dry flower yield of chamomile -ANOVA table

Source	df	Means sum of squares							
		Weeks after transplanting							
		7	8	9	10	11	12	13	14
Block	3	5.667	7.883	54.729	21.052	83.752	49.729	210.396	58.167
Treatment	3	64.833*	171.333*	343.229**	491.729**	1148.417**	424.396**	686.729ns	100.000ns
Error	9	9.611	35.056	17.007	62.340	91.250	14.007	257.963	30.278
CV		20.67	16.11	8.08	13.57	12.11	7.46	25.47	26.52

df = degree of freedom
CV = % coefficient of variation
*, **, ns = Significant at P=0.05, P=0.01, or non significant, respectively.

Appendix 29: Effect of gibberellic acid on the dry flower yield of chamomile -ANOVA table

Source	df	Means sum of squares							
		Weeks after transplanting							
		7	8	9	10	11	12	13	14
Black	3	1.062	7.000	9.562	38.896	31.562	41.833	42.229	12.417
Treatment	3	18.229*	241.500**	249.562**	603.062**	771.062**	457.833**	425.729**	192.250**
Error	9	3.563	23.833	30.729	26.007	5.841	31.111	14.563	4.194
CV		17.87	18.42	14.81	9.91	4.11	12.61	9.32	9.81

df = degrees of freedom
CV = % coefficient of variation
*, ** = Significant at P=0.05 or P=0.01, respectively.

Appendix 30: Effect of benzyladenine (BA) and its time of application (TA) on the total dry flower yield of chamomile (Season 1) - ANOVA table.

Source	df	SS	MSS	F
Block	2	1293.225	646.613	
TA	1	18.350	18.350	0.026^{ns}
Error a	2	1429.775	714.888	
BA	3	7772.433	2590.811	67.146^{**}
TA:BA	3	299.817	99.939	2.590^{ns}
Error b	12	463.025	38.285	

$CV_a = 4.88$

$CV_b = 1.97$

df = degrees of freedom

SS = Sum of squares

MSS = Mean sum of squares

CV = % coefficient of variation

**, ns = Significant at P = 0.01 or non significant, respectively.

Appendix 31: Effect of benzyladenine (BA) and its time of application (TA) on the total dry flower yield of chamomile (Season 2) - ANOVA table.

Source	df	SS	MSS	F
Block	3	3333.250	1111.080	
TA	1	242.00	242.00	0.042^{ns}
Error a	3	17268.252	5756.083	
BA	3	64338.730	21446.250	39.589^{**}
TA.BA	3	19164.750	6388.250	11.793^{**}
Error b	18	9751.000	541.722	

$CV_a = 9.40$

$CV_b = 5.80$

df = degrees of freedom.

SS = Sum of squares

MSS = Mean sum of squares

CV = % coefficient of variation

ns = Not significant at P=0.05

** = Significant at P=0.01.

Appendix 32: Effect of ethephon on the total dry flower yield of chamomile - ANOVA table.

Source	df	SS	MSS	F
Block	3	4636.50	1545.50	
Treatment	3	45139.50	15046.50	7.832**
Error	9	17290.00	1921.11	

CV = 11.72

df = degrees of freedom
SS = Sum of squares
MSS = mean sum of squares
CV = % coefficient of variation
** = Significant at P=0.01.

Appendix 33: Effect of gibberellic acid on the total dry flower yield of chamomile - ANOVA table.

Source	df	SS	MSS	F
Block	3	126.687	42.229	
Treatment	3	1277.187	425.729	29.234**
Error	9	131.064	14.563	

CV = 9.32

df = degrees of freedom.
SS = Sum of squares
MSS = Mean sum of squares
CV = % coefficient of variation
** = Significant at P=0.01.

Appendix 34: Effect of ethephon on the flower diameter of chamomile -ANOVA table

Source	df	Means sum of squares							
		Weeks after transplanting							
		7	8	9	10	11	12	13	14
Block	3	1.256	0.096	0.078	0.078	0.549	0.242	0.054	0.364
Treatment	3	0.009^{ns}	0.252^{ns}	0.182^{ns}	0.132^{ns}	0.026^{ns}	0.138^{ns}	0.356^{ns}	0.274^{ns}
Error	9	0.414	0.187	0.082	0.191	0.039	0.101	0.146	0.263
CV		7.20	4.76	3.16	5.35	2.16	3.73	4.56	6.56

df = degrees of freedom
CV = % coefficient of variation
ns = Non significant at P=0.05.

Appendix 35: Effect of gibberellic acid on the flower diameter of chamomile -ANOVA table

Source	df	Means sum of squares							
		Weeks after transplanting							
		7	8	9	10	11	12	13	14
Block	3	0.172	0.082	0.062	0.035	0.272	0.121	0.070	0.125
Treatment	3	0.364^{ns}	0.421^{ns}	0.185^{ns}	0.118^{ns}	0.157^{ns}	0.911^{ns}	0.135^{ns}	0.088^{ns}
Error	9	0.358	0.162	0.291	0.259	0.247	0.301	0.171	0.230
CV		7.20	4.56	6.13	6.63	5.64	6.44	4.91	6.17

df = degree of freedom
CV = % coefficient of variation
ns = not significant at P=0.05