EFFECT OF CALCIUM CHLORIDE, CYTOKININ AND ABSCISIC ACID ON GROWTH AND POSTHARVEST QUALITY OF ROSE CUT-FLOWER
(*Rosa hybrida*)

REGINAH WAKOMI MBURU

A Thesis Submitted to the Graduate School in Partial Fulfillment of the Requirements for the Award of the Degree of Master of Science in Horticulture of Chuka University

CHUKA UNIVERSITY

OCTOBER, 2024

DECLARATION AND RECOMMENDATION

Declaration

This thesis is my original work and has not been presented for an award of a diploma or conferment degree in any other institution.

Signature: ______________________ Date 11/10/2024

Mburu Reginah Wakomi
NM10/58075/22

Recommendation

This thesis has been examined, passed and submitted with our approval as University supervisors.

Signature: ______________________ Date 14/10/2024

Dr. Grace Opetu Oloo-Abucheli, PhD
Chuka University

Signature: ______________________ Date 11/10/2024

Prof. Geofrey Kingori Gathungu, PhD
Chuka University

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DEDICATION

This thesis is dedicated to the Almighty God, my parents Mr. Johnson Mburu and Mrs. Leah Mburu alongside my siblings for their love and support.

ACKNOWLEDGEMENT

This research and thesis would not have been possible, without the advice of, and support of several individuals and organizations. Above all, I give thanks to God Almighty for preserving my physical and mental health and giving me strength and grace throughout the process of collecting, analyzing, and interpreting data for my thesis. I am very grateful to my supervisors Dr. Grace Opetu Oloo-Abucheli and Prof. Geofrey Kingori Gathungu for their interest, advice, discussion, and assistance throughout the study period, proposal development, research implementation and thesis preparation. They diplomatically guided me across the chapters, with inspiration, good humor, and enthusiasm. Their methodical approach taught me a lot, and for this am indebted to them.

I sincerely thank Chuka University through the Department of Plant Science for providing the laboratory equipment required to run the research. Special regards to the directors Redlands Roses PLC, Mr. Aldric Spindler and Dr. Isabel Spindler, for allowing the research to be conducted on their farm. I thank Mr Jared Obure, Mr Samson Philip Kilonzo, Mrs. Lydia Ndung'u Kibet, and Ms. Farida Ochieng' for supporting the establishment and management of the experimental plot and data collection. I am grateful to Olij Kenya Propagator for the timely supply of quality planting materials to establish the experiment. I also thank Syngenta Research and Development Centre through Mr. Daniel Kisavi for allowing me to access and use their laboratory during the experiment. I am indebted to my parents Mr. Johnson Mburu and Mrs. Leah Mburu for their tireless financial and emotional support.

I sincerely thank my lecturers: Prof. Moses Muraya, Dr. James Kirimi, Dr. Martin Njogu, Prof. Njagi Nanua, Dr. Carol Anaye, Dr. Peris Nderitu, Dr. Haggai Ndukhu, and Prof. Shelmith Munyiri for their positive criticism on and improvement of my work. Special appreciation goes to technologist Ms. Lucy Gatwiri for her guidance on usage of laboratory equipment during the experiment. Many thanks goes to my colleagues: Joyce Manda, Elijah Barasa, Pauline Logova and Joyce Njihia for their support and encouragement.

ABSTRACT

The global floriculture industry has grown steadily over the last few years and is predicted to do so in the coming years. However, there has been an increase in losses both in quantity and quality which has significantly affected the production and return on investment for rose flowers. The highest loss reported has been at the retail level (39.82%), wholesalers (27.52%), producers (18.87%) and local traders (13.78%). These losses have been attributed to high respiration and rapid cell disintegration leading to accelerated aging and loss of aesthetic value of cut rose. Efforts to minimize these losses have been focused on manipulating the growing environment, using preservatives and cold treatment. Application of growth regulators can reduce preharvest and postharvest losses. Much as there is potential to reduce losses through growth regulators, there is limited knowledge on the effect of calcium chloride, cytokinin and abscisic acid on growth and postharvest quality of rose flower. This study therefore investigated the effect of calcium chloride, cytokinin, and abscisic acid on the growth and postharvest quality of tea hybrid rose, Rhodos variety. The study was cultivated over two flushes, August 2023- November 2023 and November 2023-January 2024 in Redlands Roses PLC, in Ruiru, Kiambu County. The field and the postharvest experiments were laid out in RCBD and CRD, respectively. The field experiment had 10 treatments including non-application (control); $CaCl_2$ (250 mg/L, 500 mg/L and 750 mg/L), CKs (150 mg/L, 250 mg/L and 350 mg/L), and ABA (5 mg/L, 10 mg/L and 15 mg/L). The postharvest experiment comprised of different sets of treatment. There were 28 treatments; the first set of ten treatments comprised of control (0); $CaCl_2$ (250 mg/L, 500 mg/L and 750 mg/L), CKs (150 mg/L, 250 mg/L and 350 mg/L), and ABA (5 mg/L, 10 mg/L and 15 mg/L). In the second set the cut flowers were subjected to the treatments in the field and during postharvest. The third set cut flowers were treated only at postharvest stage. Data was collected at seven days' interval throughout the growth period, starting from 3 weeks after initial bending in flush 1 and after pruning in flush 2. At postharvest, data was collected at two days' interval from the day of vasing until the termination of vase life of individual stem. Data obtained was analyzed using SAS version 9.4 and significant means were separated using the Least Significant Difference at $\propto=0.05$. The analysis of variance for flush 1 and 2 respectively, showed that CKs significantly ($p<0.05$) increased the number of shoots produced per plant (4.22 and 2.78 shoots), stem length (83.22 cm and 82.56 cm), leaf area (76.67 and 72.44 cm^2), number of suckers (17.83 and 15.67), flush days (51.72 and 53.67 days), and chlorophyll content (72.95 and 70.77 SPADS) in cut flowers. On the other hand, the shortest stem length was recorded on plots treated with ABA (68.56 and 68.78 cm), flush days (49.06 and 50.67 days), chlorophyll content (64.74 and 65.13 SPADS) and leaf area (61 and 54.11 cm^2). $CaCl_2$ reduced incidences of bent peduncles in flushes one and two (2.07 and 1.13). At postharvest in flush 1 and 2 respectively, $CaCl_2$ and CKs significantly ($p<0.05$) increased chlorophyll contents, improved the petal color, vase life (14.67 and 13 days; 14.33 and 12.67 days) and reduced weight loss. To improve on the growth qualities (number of shoots produced per plant, stem length, leaf area, flush days, and chlorophyll content) the growers should consider application of cytokinin at high concentrations (250 mg/L and 350 mg/L). However, application of cytokinin comes with a risk of increased number of suckers. To reduce incidences of bent peduncles, growers should always apply 750 mg/L of $CaCl_2$ before the formation of the flower bud. In order to improve cut flower quality after harvesting (chlorophyll contents, petal colour, vase life and reduced weight loss) growers should apply cytokinin and calcium chloride at preharvest or at preharvest and postharvest. To reduce the number of flush days in target of a specific market, growers can apply ABA (15 mg/L) at prebloom stage.

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LIST OF ABBREVIATIONS AND ACRONYMS

GAPs	Good Agricultural Practices
GDP	Gross Domestic Product
HCD	Horticultural Crop Directorate
PCPB	Pest Control Product Board
SAS	Statistical Analysis Software
PGRs	Plant Growth Regulators
ABA	Abscisic Acid
EU	European Union
$CaCl_2$	Calcium Chloride
CKs	Cytokinin

CHAPTER ONE

INTRODUCTION

1.1 Background to the Study

Rose (*Rosa hybrida*) is a popular and extensively cultivated cut flower amongst commodities in the floriculture industry. It is grown because of its beauty, fragrance and long-lasting blooming season (Desta *et al*., 2022). Cut Rose is a woody perennial flowering plant of the genus Rosa in the Rosaceae family. It is primarily native to Asia, with a few native to Europe, North America, and Northwest Africa (Leghari *et al.*, 2016). It presents as an erect or climbing shrub with stems that have sharp prickles, which are 1.5 feet to 6 feet tall. Rose may include floribunda, hybrid tea, Grandiflora, and miniature roses (Tkachenko, & Kapelian, 2021). The flower colour varies, some being scented and others unscented. According to Yu & Mingsan (2018), it may be grown for the production of cut flowers, for decoration purposes or as a raw material for the cosmetic and pharmaceutical industries.

Over the years, the Netherlands has been the leading global producer of floriculture crops, including roses. However, China has overtaken the Netherlands with a global production share of 19%, followed by the USA (12%), Netherlands (10%), Japan (8%) and Brazil (5%), respectively (Adebayo *et al*., 2020). The five largest importers have been the United States, Germany, Netherlands, United Kingdom, and Russia, having about a 60% market share (Guaita-Pradas *et al*., 2023). The worldwide market for roses (those that are grown for the primary purpose of being sold as cut flowers) is expected to grow up to 6.3% over the next five years, rising to $57.4 Billion in 2024, up from $42.4 Billion in 2019 (Guaita-Pradas *et al*., 2023). According to Mwase (2015) and Bartilol *et al*. (2019), Kenya is the top African country exporting flowers to the EU, with an export proportion of 38%. Other nations in order of ranking consist of Ethiopia (15%), Zimbabwe (5%), Uganda (3%), South Africa (2%), Zambia (2%), and Tanzania (1%) [Mèmonsso *et al*., 2023; Mwase, 2015]. This indicates potential for further growth in the sector and an opportunity for businesses to capitalize on the increasing demand of roses.

In Kenya, cut rose is grown in greenhouses. The major cultivation areas are around Lake Naivasha, Mt. Kenya, Nairobi, Thika, Kiambu, Athi-River, Kitale, Nakuru, Kericho, Nyandarua, Trans Nzoia, Uasin Gishu and Eastern Kenya. Further, Kenya has

over 200 registered flower farms, with the majority of the output being produced in big and medium-sized flower farms covering 20–100 hectares (HCD, 2021a). The most important area in Kenya for producing cut flowers is Lake Naivasha, where about 70 farms span more than 3,000 hectares of greenhouse space and generate about 8,000 metric tons of flowers every month, mostly roses.
(HCD, 2021b).

The floriculture industry in Kenya has been a major contributor to the economy, resulting in increased GDP, job creation, and improved livelihoods for millions of people. According to Kamer (2022), the country exported 210 thousand metric tons of cut flowers in 2022 and contributed to up to 33% of the Agricultural Gross Domestic Product (GDP) [Kogo *et al*., 2021]. The floriculture business supports about 500,000 people, comprising over 100,000 workers at flower farms, and affects more than 2 million people's daily lives (Wangechi & Kariuki, 2022). Studies on the performance of the horticultural subsector show that increased horticultural exports led to increased GDP in Kenya (HCD, 2021b). The horticulture sector is expected to continuously contribute to the country's economy.

Quality management at preharvest and postharvest stages of cut flowers is considered important and practical for supplying satisfying products to the market. As studied by Abdolmaleki *et al.* (2015), the quality of cut-roses after harvest is mainly affected by three major factors: Pre-harvest growth conditions; rapid respiration, which speeds up the aging of most flowers; and rapid cell disintegration, thus reducing the vase life of cut flowers. El-Beltagi *et al*. (2022) reported that continuous cold treatment and preservatives hardly improve the vase life and quality of roses as per the market requirements. This is due to the increased loss of cell wall integrity resulting from rapid respiration and depletion of food in the rose plant, thereby causing loss of commercial value of roses. Postharvest losses have significantly affected the production and return on investment for cut roses. Omar *et al*. (2014) reported that the highest loss occurred in ranked order at the retail level (39.82%), wholesalers (27.52%), producers (18.87%) and local traders (13.78%). As such, further study is needed to develop alternatives for improving cut roses' growth and postharvest quality.

Calcium chloride, Cytokinin and Abscisic Acid are inexpensive growth regulators and have great potential for commercial exploitation. These growth regulators are needed by plants for leaf emergence, healthy growth, flowering, fruiting and senescence (Chen & Zhang, 2018). They also help regulate the quantity of biomass generated, the plant's shape, and the capacity of the plant to resist environmental stress (Husen, 2022). In general, calcium chloride and the selected growth regulators can influence changes in the anatomical and metabolic processes associated with the growth and quality of plants at low concentrations.

Calcium chloride is an inorganic salt that functions as an endogenous signal molecule that controls plant growth factors (Maehara, 2020). It is among the most known soluble salt types of calcium. It triggers the production of hormones and enzymes that regulate processes, including leaf and shoot development, cell wall formation, and root development (Kumar & Pandey, 2017). Additionally, it enhances the proper development of cell walls and preserving their integrity aids in maintaining the quality of fresh produce (Barman *et al*., 2018). According to Kumar& Pandey (2017), it is involved in regulating calcium-dependent ion channels, which are involved in controlling cell membrane potential and the movement of ions across the cell membrane. This is important for proper plant growth, as it allows for the movement of photosynthates, nutrients and water in the plant. Consequently, Guo *et al.* (2023) observed that calcium chloride actively influences the horticultural crops' quality, maturity, repining, and senescence. The use of calcium chloride, therefore, promotes rose flower growth and quality by influencing physiological processes for shoot initiation, growth and maturity.

Previous research has indicated that calcium affects the quality and longevity of several vegetable products, including cucumber (Cid-Lopez *et al*., 2021) and strawberry (Amiri *et al*., 2021). Additionally, adding calcium chloride to fruits after they are harvested, increases their nutritional value, delays the ripening process, reduces rotting, and increases the amount of calcium in the treated fruit (Ben-Fadhel *et al*., 2018). The impacts of applying calcium chloride after harvest on some fruits and vegetables include reduced respiration, ripening development, and senescence (Mazumder *et al*., 2021). Thus, Calcium chloride application in roses may improve the postharvest quality of roses by slowing down the pace of degradation and metabolism, reducing losses and increasing the economic benefit.

Exogenous application of plant growth regulators such as Cytokinin and Abscisic acid also impacts the growth and quality of plants (Shah *et al*., 2021). They can alter or regulate plant physiological processes when used in small concentrations (Zahid *et al*., 2023). These physiological processes may include; cell division and elongation root and shoot growth, flowering, fruiting, and seed production (Zhang & Li, 2019). While metabolic processes give plants their energy and structural components, the growth regulator controls the rate at which each component grows and combines the many components to create the form that we identify as a plant. They also play a role in the defense of plants against diseases and pests (Muhammad *et al*., 2024). According to Sanaullah (2024) calcium chloride helps to trigger the production of defence compounds, such as phenolics and phytoalexins, which can aid in defending the plant from microbial infection and insect infestations. Therefore, losses arising due to quality issues in roses can be managed by using calcium chloride and growth regulators to cultivate roses.

Cytokinin is derived from the amino acid adenine. It consists of various molecular structures, including; kinetin, zeatin, and 6-benzyl amino purine. It is synthesized in several ways in plants, including the breakdown of ATP (adenosine triphosphate) (Argueso & Kieber, 2024) and the shikimate pathway (Li *et al*., 2021). It is important in regulating plant processes, such as root and shoot growth, bud formation, cell division and differentiation, leaf aging, and flowering (Tavakkoli& Rudell, 2016). Also, it helps create a balance between photosynthesis and respiration. It has been demonstrated to be crucial to postharvest quality by delaying the senescence of fruits and vegetables, thereby extending their shelf life (Li *et al*., 2024). Additionally, it has been shown to reduce the incidence of chilling injury, which can negatively affect the quality of harvested produce (Yadav & Prasad, 2015). Supply of rose flowers with cytokinin improves growth due to sufficient cell division and differentiation and also improves the vase life because of the delayed senescence.

Abscisic acid (ABA) is essential for the growth and maturation of plants. It is generated within the chloroplasts and then released in reaction to environmental conditions like extreme temperatures (Hao & Zhang, 2020). Plants use the carotenoid route, also referred to as "indirect pathway", to synthesise ABA (Chen *et al*., 2020). It is moved to other plant areas, attaches to receptors and starts several physiological processes. The processes may include seed dormancy and germination, stomata opening and closing, cuticle wax build-up, water uptake regulation, and stress response control (Hao & Zhang, 2020). Therefore, it can be used to regulate the growth and market value of roses because of its ability to delay leaf senescence, induce a response to stress and regulate water uptake.

Rapid senescence and loss of cell integrity are significant problems in cut roses. Further, roses frequently wilt because of continuous transpiration, rapid respiration and loss of cell wall integrity (Ahmed & Saleem, 2016). These physiological processes cause neck bending, wilting, and staining of petal sap resulting in the termination of the vase life and loss of ornamental value of cut flowers (Liu *et al*., 2024; Abdolmaleki *et al*., 2015). Growth regulators contribute to delayed ageing and the plant's capacity to react to both biotic and abiotic stressors, a requirement for quality growth and prolonged shelf life (Mazumder *et al*., 2021; Janowska & Andrzejak, 2022). They also regulate plant growth, flowering, fruiting, ripening and senescence (Chen & Zhang, 2018; Kudoyarova, 2024). However, their inadequate endogenous amounts may result in the limited ability of the plant to regulate physiological processes, therefore affecting growth and quality at pre and postharvest.

Postharvest quality is the biggest concern for the cut rose market and commercial growers (Zhang & Li, 2019). The postharvest quality of rose is affected by preharvest and postharvest activities, especially during growth and facing (Liu *et al.*, 2024). Maintaining the quality at preharvest and postharvest can help reduce losses, extend life of blooms in vase and increase the return on investment obtained from the sale of the flowers (Ahmed & Saleem, 2016). Cytokinin may contribute to reduced senescence rate (Tavakkoli& Rudell, 2016), Abscisic acid helps the plant respond to stress (Hao & Zhang, 2020), and calcium chloride contributes to cell wall integrity and uptake of other nutrients, which theoretically regulates the growth and quality of plants by their harmonious activity (Barman *et al*., 2018). They also regulate the timing of flower bud formation and flowering by controlling the rate at which cells divide and differentiate and helping the plant respond to biotic and abiotic stresses (Liu *et al*., 2023). Furthermore, it has been observed that growth regulators can be manipulated as strategies for improving the development and quality of roses. Thus, regulating flowering and the rate of senescence can be a good measure to improve the quality of growth and vase life of roses.

The use of growth regulators could help growers reduce losses across the supply chain. However, most rose growers do not supply growth regulators to the rose during growth and postharvest (Jain *et al*., 2016). Most efforts have been directed at manipulating the growing environment and using preservatives (Khan *et al*., 2018). Therefore, this study aims to contribute to and solve some of these challenges by finding appropriate concentrations of calcium chloride, cytokinin and abscisic acid for the growth and postharvest quality of roses in the future.

1.2 Statement of the Problem

There has been considerable growth in the global floriculture industry during the previous few years, and this is anticipated to continue. However, there are increased losses in the quantity and quality of cut roses that significantly affect the production and return on investment. The highest loss reported has been at the retail level (39.82%), wholesalers (27.52%), producers (18.87%) and local traders (13.78%). These losses have been attributed to high respiration and rapid cell disintegration resulting in rapid deteroriation and loss of aesthetic value in cut rose. Additionally, most of the rose growers do not apply growth regulators during growth and postharvest, and efforts to minimize losses have been focused on manipulating the growing environment, using preservatives and cold treatment. Application of growth regulators can reduce preharvest and postharvest losses. However, to the best of the author's knowledge, there is limited knowledge on the effects of concentrations of growth regulators like calcium chloride, cytokinin and abscisic acid on rose growth and postharvest quality. It is, therefore, necessary to determine suitable concentration levels that result in improved growth and postharvest quality of rose cut-flowers.

1.3 Objectives of the Study

1.3.1 General Objective

To evaluate the contribution of calcium chloride and plant growth regulators on the growth and quality of cut-roses.

1.3.2 Specific Objectives

i. To determine the effect of calcium chloride, cytokinin and abscisic acid application on the growth of rose cut-flower.
ii. To determine the effect of calcium chloride, cytokinin and abscisic acid application on the postharvest quality of rose cut-flower.

1.4 Hypotheses

The following hypotheses were tested;

H_{01}: There is no statistically significant effect of calcium chloride, cytokinin and abscisic acid application on the growth of rose cut-flower.

H_{02}: There is no statistically significant effect of calcium chloride, cytokinin and abscisic acid application on the postharvest quality of rose cut-flower.

1.5 Justification of the Study

The Kenyan economy is significantly influenced by the horticultural sector through increased GDP, job creation, and improved livelihoods for millions of people (Kogo *et al*., 2021). The floriculture business supports about 500,000 people, comprising over 100,000 workers at flower farms, and affects more than 2 million people's daily lives (Wangechi & Kariuki, 2022). Therefore, strategies must be explored to sustain and improve growth and postharvest quality of cut flowers. The application of growth regulators is an alternative for improving the growth and quality of rose. Calcium chloride, cytokinin and abscisic acid significantly affect flowers' growth and postharvest quality, but the exact concentrations required for roses are yet to be identified. Also, this area has not been researched widely and adequately documented; and could help enhance the growth and postharvest quality of roses due to increased cell wall integrity (El-Beltagi *et al.,* 2022), increased growth, reduced respiration, and senescence rate (Janowska & Andrzejak, 2022).

By understanding the appropriate concentrations of these growth regulators, the floriculture industry can optimize the growth and postharvest quality of roses and reduce the quantity and quality losses associated with the current production and postharvest processes. This is especially important because rose production and associated economic growth are key to the Kenyan economy. Therefore, this study provides invaluable information for the Kenyan rose industry, which is vital for the increased return on investment for producers and the country's economic growth. Additionally, it provides information on the appropriate use of agrochemicals that are environmentally friendly and therefore reduce the residue levels in cut roses and the pollution associated with the use of heavy chemicals used in preharvest and postharvest of cut rose flowers. In the long term, the study contributes to satisfying the rose market demand for quality and quantity.

CHAPTER TWO

LITERATURE REVIEW

2.1 Overview of Rose Flower Production

Rose flower (*Rosa hybrida*) is a popular species of flowering plants in the genus Rosa, within the family Rosaceae (Guan *et al*., 2024). This prickly, woody shrub is specifically employed as a peace and wartime emblem of beauty and love (Wang, 2023; Leghari *et a*l., 2016). It contains about 2000 cultivars and more than 300 species (Leghari *et al*., 2016). Many species are indigenous to Asia, but some are also found in Africa, North America, and Europe (Datta & Gupta, 2022). It is widely cultivated and highly prized for its attractive flowers in a variety of colours, fragrance and symbolism (Yu & Mingsan, 2018). Amongst the cut flowers including; carnation, chrysanthemum, lilies and alstroemeria, rose is the most popular cut-flower because it is widely utilized for creating bouquets and for floral arrangements (Bajaj, 2019).

Recently, rosess have been used in the floristry industry to decorate houses and events such as weddings and anniversaries (Horibe & Yamada, 2017). Additionally, some varieties of roses have recently been utilized as a supply of rose water, rose oil, and rose perfumes, which are produced by extraction with solvents and hydro-distillation (Baydar, 2016). Currently, about 300 volatile ingredients useful in cosmetology and pharmacology have been found in rose oil (Venkatesha *et al*., 2022). Rose oil has also been utilized in the culinary and pharmaceutical industries to flavor tobacco, liquors, and meals, as well as fragrances and cosmetics (Gorji-Chakespari *et al*., 2017). According to Gateva *et al*. (2020), Rose-derived natural compounds have been shown to have significant biological effects, including anti-tumor, anti-viral, anti-bacterial, and antioxidant. Therefore, this makes rose-flower important in secondary industries.

Rose production encompasses a broad spectrum of cultivation strategies, such as soilless, organic, greenhouse, open field, and protected methods (Koukounaras, 2020). Arguably the most intensive horticultural methods among them is greenhouse farming, which focuses on the production of high-value commodities like flowers. There are prospects for increased yields, earlyness, reliability in production, and better quality with the management of environmental variables (temperature and light), greater effectiveness of resource usage (water and fertilizers), and use of advanced technologies (hydroponic and automation) (Koukounaras, 2020). Consequently, to optimize rose flower productivity and quality, a variety of acceptable cultivars should be chosen, along with numerous cultural techniques.

Production between open fields and greenhouse cultivation differs greatly, in open fields, the ideal rose yield is 25–50 stems/M^2 per year; in greenhouses, and it is 50–200 stems / M^2/year (Meshram *et al*., 2022). Tsanakas *et al*. (2017), reported that greenhouse roses develop three classes of shoots: i) the flower shoots, which bear one or more terminal flowers and are the plant's most important part from a commercial standpoint; ii) the canopy shoots, which form the plant's framework and for the photosynthetic purpose; these shoots, which provide the plant's expanding sections with carbohydrates, arise from axillary buds on the main shoot; and iii) the non-flowering shoots, which are flower branches that lost their terminal bloom early in their development. This, therefore, makes greenhouse cultivation more productive due to the growth structure compared to open-field cultivation.

Quality and quantity are two crucial elements that determine cut-flower roses' value (Ketter, 2015). Thus, it is critical to comprehend how pre- and post-harvest factors affect quality and quantity (Abdolmaleki *et al*., 2015). Quality attributes such as stem length, days of freshness and absence of disorder influence the economic value of the crop (Verdonk *et al*., 2023). Generally, flowers are categorized according to their length in increments of 10 cm. If the stem length increases, the produce advances to the next grade above it (Kwon *et al*., 2022; Ahmad, 2009). Additionally, long days of freshness for flowers satisfy the market requirement and are made up by the amount that can be made from the flowers' sale (Janowska & Andrzejak, 2022).

2.1.1 Crop Management

Rose plants are planted in rows to allow for light, air circulation, and effective implementation of crop management practices. Leus *et al*. (2018) report that practices such as choosing the right varieties for a particular altitude, media preparation and fertilization, irrigation, pest and disease control, training, desuckering, disbudding, sanitation harvesting, postharvest handling are key to production of quality greenhouse cut roses. According to Singh (2006), the specific timing of each practice depends on the individual needs of the crop and the local climate (Singh, 2006). However, these practices should be incorporated into the cultivation program of roses from planting or pruning to harvesting.

Shoot-bending has become a standard cultural practice in cut flower rose (Rosa hybrida L.) production. It has been reported that bending of the primary shoot promotes the formation of lateral shoots by breaking apical dominance (Meshram *et al*., 2022). It is the most important operation for developing a good number of lower or basal shoots for the build-up of a strong framework of the plant (Raghatate, 2021). After four weeks of planting the mother shoot is bent on the second leaf or close to the crown region (Raghatate, 2021). After the initial bending, the bending of shoots is done all year round, towards the outside of the bed and these shoots form the photosynthetic canopy of the plant; while the stronger basal shoots arising subsequently from the crown is harvested later as cut flowers Gonzales-Real *et al*. (2007). The main characteristic of the bending system is that the blind, weak or early flowering shoots are bent low to the ground (Tangkas *et al*., 2017).

During the vegetative growth phase, rose flower shoots grow upwards and auxiliary shoots develop originating from the leaves (Singh, 2006). Auxiliary shoots suck photosynthates, water and nutrients from the flower shoot and therefore they are referred to as suckers. To ensures production of healthy flower heads and reduce competition for growth resources the suckers must be removed (Singh, 2006). The removal of these auxiliary shoots is referred to as desuckering. According to Singh (2006), the optimal time to desucker rose plants is during the early stages of growth when the sucker has not exceeded 2 cm in length. The sucker should be removed in the morning hours when the plant is turgid using manual techniques, as this allows for the selective removal of specific suckers, while still providing good coverage of the entire plant (Singh, 2006).

Disbudding operation is also an important factor in the maintenance of high-quality products (Meshram *et al*., 2022). This practice is reported to be a standard operation in the cultivation of roses, carnations, chrysanthemums and celosias (Norman, 2004). It involves the removal of green buds on the bend plants before they develop flowers to promote better growth and healthier flower shoots (Meshram *et al*., 2022). When allowed to grow, these buds consume the photosynthates stored by the plant in the bending therefore diverting nutrients away from the foliage and leading to an unbalanced, unhealthy rose crop (Singh, 2006). Sanitation efforts such as picking up fallen leaves and prunings can contribute to reducing disease incidence in spring (Karlik & Golino, 2017). Many insects can be hosed off with hand-picking of plant debris or pruned out while weeds can be managed with mulches and hand-weeding to reduce competition and provide a healthy crop (Karlik & Golino, 2017).

Rose is a heavy feeder, thus requiring regular fertilizer input with a balance in the macro and micronutrients (Leghari *et al*., 2016). Its growth and quality are compromised if not provided with the correct nutrients and growth regulator (Martin, 2015). To ensure efficiency use of nutrients fertilizer application should be done at the appropriate time when the plant requires them most and in the right method (Singh, 2006). Nutrients such as nitrogen, phosphorus and potassium are required in large quantities due to their function of building up the plant architecture and hence, affect plant growth more as compared to other mineral nutrients (Singh, 2006). In addition to these, secondary nutrients like calcium, sulphur and magnesium which influence metabolic functions and tissue building are also needed in fairer quantities than the other essential elements such as iron, manganese, zinc, copper, boron, molybdenum and chlorine (De Hoog, 2001).

Optimal irrigation is very important because it influences the plant's physiological processes which in turn affects plant growth and consequently crop production and quality (Lykas, 2005). Drought stress leads to defoliation and sunburn of canes and may contribute to spider mite problems. However, overwatering or poorly drained soils may lead to root disease and nutritional deficiencies (Karlik & Golino, 2017). The frequency and duration of irrigation depend on weather conditions and soil texture. Roses do best when 50 percent of available water is depleted between irrigations (Karlik & Golino, 2017). For greenhouse crops such as roses, climate sensors (temperature and humidity) are put inside the greenhouse to monitor plant water requirements (Lykas, 2005).

Rose plant is affected by several pests and diseases which attack different parts of rose plants. Commonly found and regular pests are thrips, aphids, mealybugs, whiteflies, caterpillars, and mites (Jayalaxmi *et al*., 2020). Several of these pests are found throughout the year damaging the rose plants and affecting the flower yield and quality. Insect and mite pests on rose can cause 28-95% damage individually or in groups (Jayalaxmi *et al*., 2020). Diseases such as botrytis and downey mildew and prevalent during cold seasons while powdery mildew is prevalent during the hot and windy seasons (Windham & Windham, 2019). However, the prevalence and significance of these pests and diseases in a crop can depend on the susceptibility of the variety. According to Windham & Windham (2019) pests and diseases can be managed using an IPM strategy which involves a combination of tactics such as pest monitoring, cultural, physical, biological and chemical methods.

The right stage, method and time of flower harvesting are of considerable importance to ensure their long vase life (Gupta & Dubey, 2018). Therefore, Flowers should always be harvested at an optimum stage of maturity early in the morning or in the evening when the temperatures are not high (Da Costa *et al*., 2021). According to Larson (2012), too immature buds do not open whereas over mature ones wither quickly. The stems should be given a slant cut using sharp secateurs on the first 5-leaflet leaf of the flowering stem to expose the maximum surface area to ensure rapid water absorption (Da Costa *et al*., 2021). The harvested flowers should be wrapped with a net, put in clean water and taken to the cold room within 45 minutes of harvesting for precooling (Larson, 2012). Precooling temperatures should be 1 to 2 ˚C to 5 ˚C for 4 hours before processing of the flowers at room temperature (Gupta & Dubey, 2018). Flowers for shipping perform better when stored at 0 ˚C to 2 ˚C temperature after grading otherwise, they can be put in vases with a postharvest solution for decoration purposes temperature (Larson, 2012; Gupta & Dubey, 2018).

2.1.2 Challenges in Rose Flower Production

Cut rose producers have suffered shortcomings at the postharvest due to inadequate storage facilities and high temperatures which have caused rapid wilting and deterioration of flowers (Nagaraju, 2020). Poor postharvest management and handling techniques have led to physical damage to the flowers reducing their quality and marketability resulting in low profits (Bakker & Vermeiren, 2018). The producers have also suffered short vaselife of their cut roses due to the high perishability of roses and poor-quality postharvest solutions used in treatment of roses after harvest (Ouedraogo & Bationo, 2018). According to Pouri *et al*. (2017), roses are prone to damage during transportation and handling due to their delicate and fragile nature, which can reduce the vase life even further causing quantity and quality losses along the supply chain.

2.2 Application of Growth Regulators

Growth regulators have been widely used to control the growth of plants including roses. Example of growth regulators that have been used for roses include; Spermine and Spermidine for growth, flowering and postharvest quality of roses (Tatte *et al.,* 2015; Tatte *et al.,* 2016) , gibberellins (GA3) for manipulating growth and flowering (Kumar *et al*., 2021), cytokinin (benzyl adenine, kinetin and zeatin) for delaying aging of leaves and increasing roses longevity (Yadav & Prasad, 2015), auxins (NAA, IAA and IBA) for shoot and root generation (Kumar *et al*., 2021), abscisic acid for response of roses to stress (Hao & Zhang, 2020), methyl jasmonate for defence against biotic and abiotic stresses (Kumar *et al*., 2021), salicylic acid for improving vase life (Abdolmaleki *et al*., 2015). Therefore, different growth regulators have different impacts on different crops at different concentrations.

According to Kumar *et al*. (2021), the possible effects of plant growth regulators depend on their method of application due to the difference in their mode of absorption by the plant, as some growth regulators are absorbed only through root, leaves or stem, and some are absorbed through all mentioned organs having an advantage to apply in either way, as cytokinin, gibberellins, salicylic acid, abscisic acid, methyl jasmonate, spermine and spermidine are absorbed through the roots, stem and also leaves by foliar sprays (Kumar *et al*., 2021) while auxin is majorly absorbed through roots by dipping and media solution (Asadi *et al.,* 2009). Some like the abscisic acid, spermine and spermidine may be administered through holding solutions (Tatte *et al.,* 2016). According to Al-Khassawneh *et al*. (2006) and Lee & Rho (2000) foliar application and soil drenching are the most common methods being used by commercial growers and relatively higher concentrations are used in the case of foliar sprays.

Foliar application is more effective if applied at the right stage of growth for controlling specific characters and it requires information about the phenology of the target plant (Stover & Greene, 2005). Another advantage of foliar spray is the repetition of application as many times as required can be made to attain certain goals. The plant response to foliar application also depends on the absorption rate and absorption is driven by the environmental conditions, temperature and humidity are the most important (Sajjad *et al*., 2017). Further, Stover & Greene (2005) reported that slightly high temperature, high humidity and longer drying time increase the absorption of growth regulators in plants.

Soil drenching is an efficient method and growth regulators are used in relatively lower doses but residual effects of growth regulators are retained in pots which sometimes harm the plant (Sajjad *et al*., 2015). Drenching has an advantage over foliar sprays because it ensures the uniformity of treatment as each plant receives the measured amount of growth regulators and absorption occurs through the root zone (Sajjad *et al*., 2017). This method is suitable for growth regulators having efficient absorption through root medium. Preplant soaking of plant material in PGRs is reported by Sajjad *et al*. (2015) to be an efficient method but its use is relatively less common on commercial scale.

2.3 Effect of Calcium Chloride, Cytokinin and Abscisic Acid Application on Plant Growth

2.3.1 Calcium Chloride and Plant Growth

Calcium is important in improving the qualitative characteristics of rose-cut flowers (Banijamali *et al*., 2018). Insufficient calcium levels lead to cell membrane deterioration; the cells become leaky, resulting in the loss of cell compounds and, eventually, the death of the cell and plant tissue (Patterson, 2013). Calcium also plays a role similar to a growth regulator regulating various plant cell functions. One such function is regulating the protein pump that regulates the uptake and movement of nutrients into the root and throughout cells within the plant. At the root level, calcium stimulates the protein channels that take up nutrients.

Studies by Abdolmaleki *et al*. (2015) showed that calcium chloride increases growth parameters in Plants of the “Dolce Vita” cultivar compared to untreated plants. Fresh weight and dry weight of shoot and flower bud and length of stem significantly increased along with augmentation of the Calcium chloride concentrations. Yildirim *et al.* (2009) also reported that the application of calcium salts significantly improved chlorophyll content helped strawberry plants avoid sodium toxicity and improved cell membrane stability and nutrient uptake under salinity stress. Additionally, a study conducted by Mortazavi *et al*. (2015) showed that pre-harvest treatment of lilies with 600 mg/L calcium chloride increased florets' diameter, relative water content and chlorophyll *b* content as compared to other concentrations and different salicylic concentrations.

Adequate availability of calcium at the root surface is required for this process to work effectively (Patterson, 2013). For most crops, net photosynthesis declines as temperatures increase beyond 34 °C. Heat stress increases stem length while reducing leaf size and flower head. Calcium can mitigate heat stress by improving stomatal function and other cell processes (Chen *et al*., 2020; Patterson, 2013). Moreover, it is involved in the regulatory mechanisms that plants activate to adjust to adverse environmental conditions of drought, heat, cold, salt, and heavy metal (Heidaria *et al*., 2022). It is often referred to as the plant's first line of plant nutrition defence. Many organisms infect plants by penetrating the cell tissue with enzymes known as pectinase, which dissolves pectin. The higher the calcium content in plants, the higher the concentration of pectin holding cells together and the greater the ability to withstand these enzymes (Banijamali *et al*., 2018; Patterson, 2013).

2.3.2 Cytokinin and Plant Growth

Temperature beyond the physiological optimum for growth induces heat stress in plants causing detrimental and irreversible damage to plant development, growth, as well as productivity (Li *et al*., 2021). Therefore, Plants have evolved adaptive mechanisms in response to heat stress. The classical plant hormones, such as auxin, abscisic acid, brassinosteroids, cytokinin, salicylic acid, jasmonate, and ethylene, integrate environmental stimuli and endogenous signals to regulate plant defensive response to various abiotic stresses, including heat. Exogenous applications of those hormones prior or parallel to heat stress render plants more thermos tolerant (Li *et al*., 2021). In particular, extreme seasonal heat caused by global warming substantially disturbs normal crop growth and yield around the world, which further exacerbates food insecurity and malnutrition. It is estimated that a 1 °C increase in seasonal temperature may directly cause 2.5–16% staple crop yield losses in tropical and subtropical regions (Battisti & Naylor, 2009).

Cytokinin, a group of plant growth regulators derived from adenine, consists of various molecular structures including kinetin, zeatin, and 6-benzyl amino purine (Li *et al*., 2021). Since their discovery, cytokinin has been implicated in almost all aspects of plant growth and development, including cell division and differentiation, shoot initiation and growth, leaf senescence, apical dominance, sink/source relationships, nutrient uptake, photomorphogenic development and increase the movement of sugars, amino acids, and trace elements to developing organs and generate protein synthesis (Argueso *et al*., 2012; Hwang *et al*., 2012). Argueso *et al*. (2012) further showed that cytokinins play important roles in interacting with biotic and abiotic factors.

Various abiotic stresses, like drought, and high and low temperatures, can severely affect the physiological metabolism of plants and cause a decline in crop yield. To survive, plants must respond quickly to external stresses and activate effective defensive responses (Wang *et al*., 2016). Increasing evidence suggests that cytokinins are involved in stress responses. Cytokinins can enhance resistance to adverse environmental factors, achieved predominantly through changes in the concentration of endogenous cytokinins or exogenous cytokinins application (Cortleven *et al*., 2019). Hot ambient temperatures cause pre-anthesis abortion in flower primordia of passion fruit during summers (Sobol *et al*., 2014). Cytokinin application showed an increased resistance in response to hot ambient temperatures. This result suggests that cytokinin has a protective role in developing flowers exposed to heat stress and may have important implications in future field applications to enhance crop production. Cytokinin applications can alleviate heat stress injury on creeping bentgrass (Wang *et al*., 2012).

Aguilar-Ayala & Herrera-Rojas (2023) when conducting a study on the inductor effect of cytokinins on flowering and fruit setting in an *Annona muricata* reported that the concentration of 1.5 ml/L cytokinin accelerated flower production and fruit formation compared to the control. This was because cytokinins increased the number of inflorescences, decreased flower drop compared to the other treatments, increased fruit size in length and diameter, and increased fruit production. Therefore, when applied during the growth of roses, the cytokinin can accelerate flower production, increase flower bud size and reduce flower abortion.

2.3.3 Abscisic Acid and Plant Growth

Abscisic acid (ABA) frequently defined as a "stress hormone" regulates biotic and abiotic stress responses (Vishwakarma *et al*., 2017). In guard cells, abscisic acid signalling activates the efflux of anion and potassium ions via membrane proteins. This decreases guard cell turgor pressure and volume, leading to stomatal closure, which prevents water loss (Kim *et al*., 2010), but simultaneously reduces the influx and assimilation of CO_2. In vegetative tissues under osmotic stress conditions, abscisic acid signalling pathways function cooperatively to induce a large number of genes involved in signal transduction and stress tolerance (Yoshida *et al*., 2014), diverting resources away from growth to enhance stress tolerance.

Abscisic acid also serves as a thermo-priming hormone that enables plants to respond more rapidly and efficiently to heat stress. It improves drought acclimation in plants. Exogenous application of abscisic acid confers *Arabidopsis* resistance more rapidly and effectively to drought-triggered dehydration stress by priming a transcriptional memory (Virlouvet *et al*., 2014). ABA modulates levels of carbohydrates and energy status through accelerated transport and enhanced metabolism of sucrose to strengthen plant thermal tolerance (Rezaul *et al*., 2019). Exogenous abscisic acid application alleviates heat-induced detrimental effects and enhances heat tolerance of tall fescue (Wang *et al*., 2017)

Additionally, abscisic acid plays an important role in both the induction and maintenance of dormancy and inhibition of germination of non-dormant seeds in many species (Duclos *et al*., 2014). The biological functions of abscisic acid have mostly been studied in the context of osmotic stress and seed dormancy. However, abscisic acid also modulates growth and development under non-stress conditions (Yoshida *et al*., 2019). Endogenous abscisic acid is associated with the maintenance of shoot growth and higher leaf emergence in Arabidopsis and tomato (Sharp *et al*., 2000; Le Noble *et al*., 2004; Yoshida *et al*., 2019).

2.4 Effect of Calcium Chloride, Cytokinins and Abscisic Acid Application on the Postharvest Quality

According to da Costa *et al*. (2021), cut-roses have a limited lifespan due to their short-lived nature and exposure to various stresses at postharvest. Reduced water uptake, exhaustion of stored carbohydrates, elevation in respiratory activity, and heightened ethylene production are some of the physiological processes that speed up withering and senescence at postharvest (da Costa *et al*., 2021). Finger *et al*. (2016) observed that Potential postharvest storage and respiration are inversely correlated, particularly in flowers where organic respiratory reserves are low. Addtionally, Tinebra *et al*., (202) reported that infection by bacteria and fungus at postharvest also shortens the vase life of cut flowers by obstructing the xylem vessels, which prevents water from being transported.

At postharvest, withering can be brought on by vascular blockage by bacteria or fungi, or it might be caused by excessive water loss linked to a higher transpiration rate as a result of natural senescence (Sun *et al*., 2001). According to Gómez-Merino *et al*. (2020), mechanical damage resulting from incorrect handling during flower postharvest causes an increase in respiration rate and shortens the flowers' functional life. Long-term exposure to inappropriate temperatures also shortens the vase life of cut flowers (Çelikel and Reid, 2002). Their shelf life is further reduced by the quality of the postharvest water, the presence of impurities like bacteria or fungus, or by the high concentration of salts, particularly chlorine.

Additionally, Postharvest quality characteristics have the potential to change during handling, transportation, and market operations (Gupta & Dubey, 2018). The major components of quality at the time of purchase include appearance, chemicals and anatomical (Salunkhe *et al*., 2012). Appearance includes size, shape, surface cleanliness, colour, freshness and absence of damage (Salunkhe *et al*., 2012). Of all the components of appearance; colour, freshness and absence of damage significantly influence customer acceptance of cut flowers (Salunkhe *et al*., 2012). According to Janowska, & Andrzejak (2022), Growth regulators can have a positive or negative impact on a flower's qualitative traits, which are indicated in terms of the stem's length as well as the size and weight of the flower and the leaf. Further, the use of growth regulators has a positive effect on leaf quality and freshness in calla lilies, due to a recorded higher greening index as well as higher protein and sugar contents (Janowska, & Andrzejak, 2022).

2.4.1 Calcium Chloride and Postharvest Quality

Several studies have shown the role of calcium in improving the vase life of rose-cut flowers (Banijamali *et al*., 2018; Javan *et al*., 2015). Moreover, applying calcium chloride after harvest delays the progress of ripening reduces decay, and increases the calcium level in treated plants, resulting in long days of freshness (Ben-Fadhel *et al*., 2018). Reducing respiration, reduced flower opening, and senescence are some effects observed in some horticultural crops from calcium chloride application after harvest (Mazumder *et al*., 2021).

Calcium chloride helps to maintain the postharvest quality of horticultural products by maintaining the integrity of plant cells' walls (Barman *et al*., 2018). Previous research has shown that calcium plays a role in the quality of several vegetable crops, including cucumber (Cid-Lopez *et al*., 2021) and strawberry (Amiri *et al*., 2021). Moreover, Aghdam *et al*. (2013) found that antioxidant capacity, anthocyanin, ascorbic acid, and phenolic compounds were conserved in cornelian cherry fruits by the postharvest application of calcium chloride.

In vase solutions, calcium chloride has been reported to extend the shelf-life of cut flowers (Perik *et al.,* 2014). It also stabilizes the cell wall due to its association with pectin, increasing rigidity and firmness and providing mechanical support to plants (Hawkesford *et al*., 2012; Li *et al*., 2012). Further, the presence of calcium chloride reduces the polygalacturonase enzyme activity and mediates the degradation of middle-lamella (García-González *et al*., 2022). These facts highlight the importance of adding

Calcium in vase solutions of rose cut flowers. Calcium is usually applied through solutions such as calcium chloride or lactate and has been shown to modify vase life in cut flowers (Combrink, 2018). García-González *et al*. (2022) reported that calcium is very important in extending the days of freshness and avoiding the loss of visual quality in cut flowers.

Foliar spray treatments of calcium chloride together with salicylic acid have been reported to reduce weight loss, improve the antioxidant system, delay chlorophyll degradation, and extend the shelf life of broccoli heads during storage (Rastegar *et al*., 2022). When treated with Ca (2%) alone and in combination with SA (0.01%) the broccoli heads significantly maintained the chlorophyll concentration at the end of storage (Rastegar *et al*., 2022). Additionally, the total phenols, flavonoid, and antioxidant capacity of the Ca (2%) + SA (0.01%) treated samples were noted to be higher. When used in the postharvest of roses, calcium chloride helps the rose flower retain freshness by maintaining chlorophyll concentration

2.4.2 Cytokinin and Postharvest Quality

It has been reported that plant growth regulators such as cytokinins enhance the chlorophyll concentration and CO_2 assimilation rate, thereby increasing the assimilated supply to the flower head and leaves (Guardiola & García-Luis, 2000) and therefore enhancing long vase life. Cytokinin application has been reported to exert good postharvest effects on bananas and broccoli, such as inhibited respiration rate, natural browning, and delayed senescence (Li *et al*., 2016). Preharvest application of Cytokinins delayed the ripening of grapes, pineapple, cherries and kiwi at postharvest (Schaller *et al*., 2015).

Cytokinins have shown an increase in flower formation and head size after preharvest treatment and reduced deterioration to maintain flower quality during postharvest storage (Aloni, 2010). Pre-bloom application of cytokinins increases the flower setting, whereas post-bloom application affects the flower head size, colour, maturity and storage quality in grapes (Schaller *et al*., 2015). The external application of cytokinins to the plant causes a decrease in chlorophyll degradation causing the plant to retain freshness at postharvest in broccoli (Clarke *et al*., 1994).

Lukaszewska *et al*. (1994) reported that feeding flowers with exogenous cytokinin might compensate for the reduction in the supply of endogenous compounds, and in carnations, senescence was delayed and average flower vase life prolonged. In another study, Liu *et al* (2023) demonstrated that treatment with cytokinin enhanced the disease protection of rose petals to *B. cinerea.* However, the mechanisms underlying the role of exogenous cytokinins in the vase life performance of ornamental roses still need to be better understood (Mayak & Halevy, 1974).

2.4.3 Abscisic Acid and Postharvest Quality

In roses, water deficit stress often leads to a bent neck disorder and, thereby, termination of the vase life because of compromised vase life (van Doorn & Perik, 1990). Abscisic acid supplied in the vase solution can induce stomatal closure in the leaves of cut flowers (Pompodakis *et al*., 2004). This effect is beneficial in reducing water deficit stress and increasing flower longevity and postharvest quality (Pompodakis *et al*., 2004). Shimizu & Ichimura. (2009) reported that cut flowers pulse treated with abscisic acid or sucrose + abscisic acid can retain water in their leaves for longer, which may be attributable to the suppression of transpiration by abscisic acid.

In Geraldton waxflower (*Chamelaucium uncinatum*), continuous treatment with abscisic acid was found to reduce solution uptake and extend foliage vase life (Joyce & Jones, 1992). Markhart & Harper (1995) reported that a pulse treatment with abscisic acid, a plant growth regulator that closes stomata, prevented "leaf crisping" in cut roses held in a vase solution containing sucrose. When applied in zucchini, ABA reduced the occurrence of chilling injury through induction of antioxidant metabolism during the first day of exposure to low temperatures and therefore preventing the formation of reactive oxygen species which causes cell damage, resulting in low postharvest quality (Castro-Cegrí *et al*., 2023). By increasing the flower’s resistance to water loss, and helping protect the petals from desiccation and other environmental stresses ABA can be used to improve the postharvest life of cut roses.

CHAPTER THREE

MATERIALS AND METHODS

3.1 Study Site

The study was conducted in Redlands Roses Public Limited Company. Redlands Roses Public Limited Company is situated at an altitude of 1564 m, a latitude of 1.145279° S and a longitude of 36.914652° E in Ruiru, Kiambu County, Kenya (Appendix 1). The climate is humid highland, characterised by seasonal dry and wet periods. Ruiru receives 845mm of rainfall annually, with long rains between March and May and short rains between October and December (Jaetzold & Schmidt, 1983). Temperatures are highest in January to mid-March before the long rainy season and lowest in June and August with a range of 13.0 °C to 24.9 °C (Nyakundi *et al*., 2017). The soils are red to dark brown friable clays suited for cash crops like coffee, maize, Irish potatoes, beans, tea, avocado, bananas, pyrethrum and livestock such as pigs, poultry and dairy cows (International Centre for Research in Agroforestry, 2020).

The field experiment was conducted in a greenhouse Delta installed with a 200-micron thick UV block and light diffusing polythene, automated ventilation, misters and fans, and nets. The first flush commenced in August 2023 and ended in November 2023. The second flush commenced in November 2023 and ended in January 2024. The postharvest experiment was conducted in Redlands Roses vase life testing room which was well-ventilated, with a temperature between 18-22 °C and humidity levels between 60-70% (Appendix 7). The room had adequate space and tables for conducting experiments and for data collection.

3.2 Experimental Design

In the field the study was laid out in a Randomized complete block design (RCBD) and replicated three times. In the postharvest the study was laid out in a completely randomized design (CRD) and replicated three times.

3.3 Treatments and Experimental Layout

In the field, the plot size was 2 m by 0.3 m and a spacing of 0.5 m between treatments and 1 m between blocks (Figure 1). The growth regulators were sourced from Precise Lab Limited. They were measured based on weights of different treatments using an analytical scale D475730236 model number TX4202L from Japan in the Research and Development Laboratory in Syngenta. The first treatment application was done at preplant. The first treatment was done 2 months after initial bending. It was done through foliar spraying of the treatment on a sunny day after dehydrating the crop for 4 hours to improve the uptake. The second treatment application was done during the pre-bloom stage when the crop was at the pea size stage through foliar application.

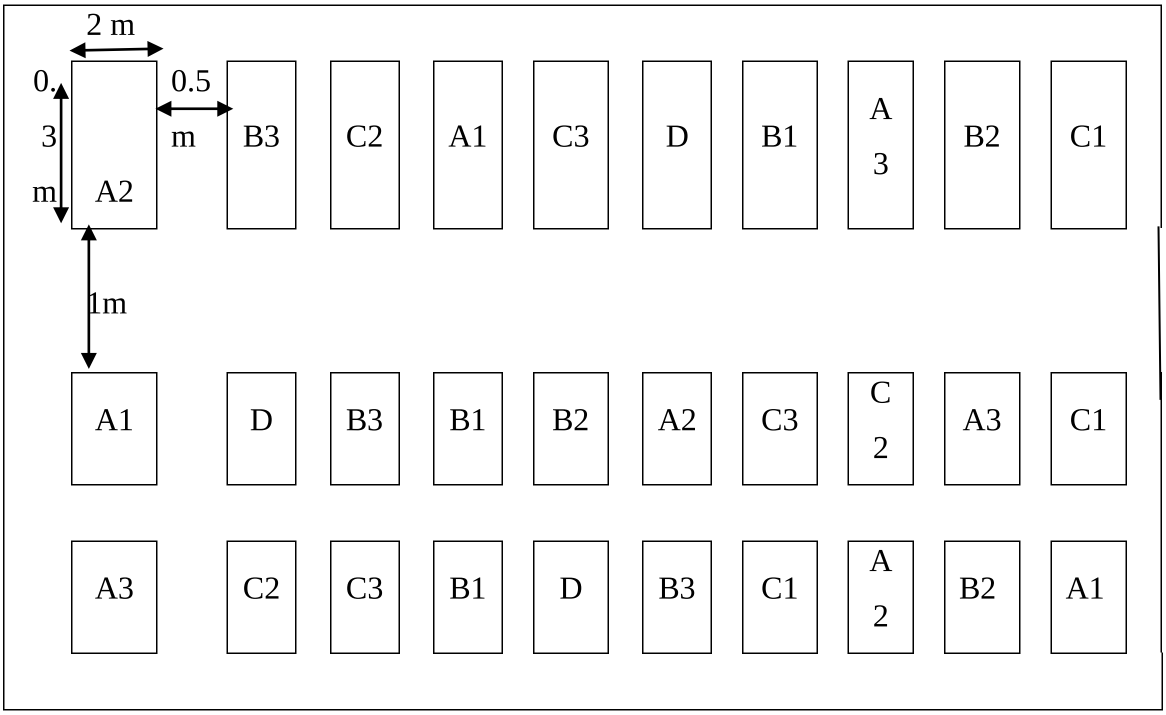

Figure 1: Field Layout

Key:

A1, A2, A3: 250 mg/L, 500 mg/L and 750 mg/L calcium chloride

B1, B2, B3: 150 mg/L, 250 mg/L and 350 mg/L cytokinin

C1, C2, C3: 5 mg/L, 10 mg/L and 20 mg/L of abscisic acid

D: Control

In postharvest, (Figure 2) the vases were disinfected with 300 ppm sodium hypochlorite solution and filled with plain water. The overnight precooled flowers were taken to room condition for grading and sorting before vasing. They were shaken to remove excess postharvest solution and checked under light for pest and disease. Hand defoliation of 60 cm and 70 cm was done at 25 cm, and 80 cm and above was done at 30 cm. The stems were trimmed at 20 mm to prevent blockage of xylem vessels and allow for uptake of water. They were sprayed with the different treatments, put in vases and placed in the vaselife room for observation and data collection. The vasing water was changed after 2 days and cleaning of the vases done simultaneously.

A2	B1	B8	D	C6	A3	A6	A4	B4	C2
A4	B8	C9	A7	B3	A6	B2	A3	A5	C1
A8	A1	C1	C5	C5	C2	A6	B7	B5	A2
D	C4	C3	A5	A9	B9	B2	B3	C2	C5
C1	B9	C6	B4	C8	B6	A2	B1	B9	C6
A3	B7	C8	B7	C7	C9	B2	A1	A1	B6
C8	A5	B5	C9	A9	C3	D	C4	C7	A4
C4	A8	C3	A7	B3	A9	C7	B8	A8	B4
A7	B6	B1	B5						

Figure 2: Postharvest Layout

Key:

A1, A2, and A3: 250 mg/L, 500 mg/L and 750 mg/L calcium chloride treated only at the field.

A4, A5, and A6: 250 mg/L, 500 mg/L and 750 mg/L calcium chloride treated both at the field and postharvest.

A7, A8, and A9: 250 mg/L, 500 mg/L and 750 mg/L calcium chloride treated only at the postharvest.

B1, B2, and B3: 150 mg/L, 250 mg/L and 350 mg/L cytokinin treated only at the field.

B4, B5, and B6: 150 mg/L, 250 mg/L and 350 mg/L cytokinin treated both at the field and postharvest.

B7, B8, and B9: 150 mg/L, 250 mg/L and 350 mg/L cytokinin treated only at the postharvest.

C1, C2, and C3: 5 mg/L, 10 mg/L and 20 mg/L abscisic acid treated only at the field.

C4, C5, and C6: 5 mg/L, 10 mg/L and 20 mg/L abscisic acid treated both at the field and postharvest.

C7, C8, and C9: 5 mg/L, 10 mg/L and 20 mg/L abscisic acid treated only at the postharvest.

D: control.

3.4 Land Preparation, Propagation, Crop Establishment and Management

3.4.1 Land Preparation

The beds were raised from the ground using building stones to a 1% gradient (20 cm height from the beginning of the bed and 15 cm at the end of the bed) (Plate 1A). The troughs were laid in the dimensions of 50 m × 0.3 m × 0.3 m and a spacing of 1 m between beds. Drainage holes were made at a spacing of 10 cm apart on the left side of the bed. Disinfection of the planting troughs using 0.05 ml/l hydrogen peroxide was done. U-shaped metal was placed to keep the troughs vertical and to provide a support system to prevent collapsing of the bed (Plate 1B). The metal was also used to lock the drainage trough at the base of the bed. The beds were filled with pumice 4.5 m^3 in 50 m bed as planting media. Cleaning of the pumice with plain water to remove impurities was done. The media was disinfected using 0.05 ml/l hydrogen peroxide to kill any existing pathogens (Plate 1C). The media was drenched with 0.04 ml/l phosphoric acid to attain a pH of 5.5 to 6.5 (Plate 1D). The driplines were laid 5cm from the end of the bed and 20 cm from each other (Figure 1). The driplines were tested for dripper capacity discharge to ensure all drips were 1.6 L/hr.

Plate 1: Raising of Hydroponics Planting Beds (A), Reinforcing the Planting Troughs (B), Sterilizing the Media with Hydrogen Peroxide (C) and Stabilizing Planting Media pH with Phosphoric Acid (D).

3.4.2 Propagation of Planting Material

The propagules were established through top-grafting. Clean stems of Red Rhodos variety were selected from the mother stock and disbudded at colour break stage when the sepals were detaching from the petals. The disbudded stems were left standing in the field for 7 days to allow for the formation of the buds. The stems were harvested using secateurs, put in plain water to avoid devitalisation and stored at 4 ˚C overnight to reduce respiration rate. They were transported to the propagation unit after 1 day and 2000 plants grafted the same day through top grafting. The scion was obtained from the Red Rhodos bud woods and the rootstalk was from natal briar bud woods. Propagation media was prepared from a mixture of cocoa peat and polyesteryne (Plate 2A). The media was put in propagation papers in trays, mounted on propagation table and the grafts planted (Plate 2B). After 21 days the roots (Plate 3A) and the shoots (Figure 5B) were fully established.

After 28 days the plants were taken to the hardening bay. On the 36th day plants were ready for transplanting and were dispatched in crates using a refrigerated vehicle to Redlands roses for planting (Plate 4). During Propagation the plants were scouted for pests and diseases every day. Based on the scouting records the plants were treated against the identified issue (Appendix 4). The fertilizer application was done from the third week of propagation using the flooding method. The fertilizer application involved farm based program (Appendix 5). The fertilizer mix was supplied in 3 cycles in a day through the fertigation system with an EC of 1.8 and a pH of 5.5-6.5. During the cold period, steaming was done to help induce rooting.

Plate 2: Planting Media at Propagation (A) and Mounted Top grafts (B)

Plate 3: Root Development (A) and Shoot Development (A) after 21 days

Plate 4: Top grafts Ready for Transplanting after 36 days

3.4.3 Crop Establishment

The quality of the grafted plants was assessed before planting by checking on diebacks, agrobacterium, pests and diseases. Only disease and pest-free plants were selected for planting. The media was irrigated with plain water to help keep the beds wet during planting. Planting was done early in the morning alternately in a spacing of 18.5 cm intra-row and 9.25 cm inter-row and 5 cm from the edge of the planting trough to avoid interlocking of roots. Planting was done with the shoots facing inside of the bed to increase surface area of exposure to light after first bending for maximum shooting. Planting was done in 3 beds with each bed having 540 plants.

3.4.4 Mulching

A black and white mulch paper of 25 microns' thickness was placed on the surface of the beds. The mulch was spread between the plants and holes punched in the middle of the mulching paper to ensure air circulation in the media (Plate 5).

Plate 5: Installation of the Plastic Mulch

3.4.5 Watering

Further, watering was done based on the weather conditions and the stage of growth. Light watering of the young plants was done in the first week of planting in the morning and in the afternoon to increase humidity and prevent wilting. Depending on the greenhouse temperatures and humidity watering was also done on the paths between the beds on daily basis.

3.4.6 Gapping

Gapping was done 7 days after planting and it involved replacement of the plants that failed to establish after transplanting.

3.4.5 Debudding, Deshooting and Desuckering

Debudding was done after every 2 days from 14 days after planting up to 56 days when initial bending was done. After initial bending, it was done after 2 weeks during the selective bending on the bending materials. It involved hand removal of the flower buds at green bud stage. De-shooting was done after every 2 days from 28 days after planting up to the the 56th day (Figure 8A). Deshooting involved removal of the maroon shoots on main crop to suppress vertical growth and increase foliage mass for bedding crop. Desuckering was done daily when the plants were at maroon stage.

3.4.6 Initial Bending

Initial bending was done eight weeks after planting. Three days before bending, the rooting system was compacted in the media and pipes with 13mm diameter laid horizontally on the media and adjacent to the base of the plant to reinforce the crop during and after bending. Leaves around the crown were also removed to expose the knuckles to light for effective shooting after initial bending (Plate 6A). Bending was done in a sunny afternoon. It involved bending the mother shoot and an addition of 2 new shoots to give enough bedding material for food reserve (Figure 8B). Initial was done 2-5 cm from the ground using both hands to encourage shooting of healthy and strong stems. One hand held the base of the plant, just above the grafting union, and the other hand held the entire upper foliage and the shoots were gently twisted to bend them at 45° from the ground. The shoots were bend towards the outside of the bed to form the bedding canopy.

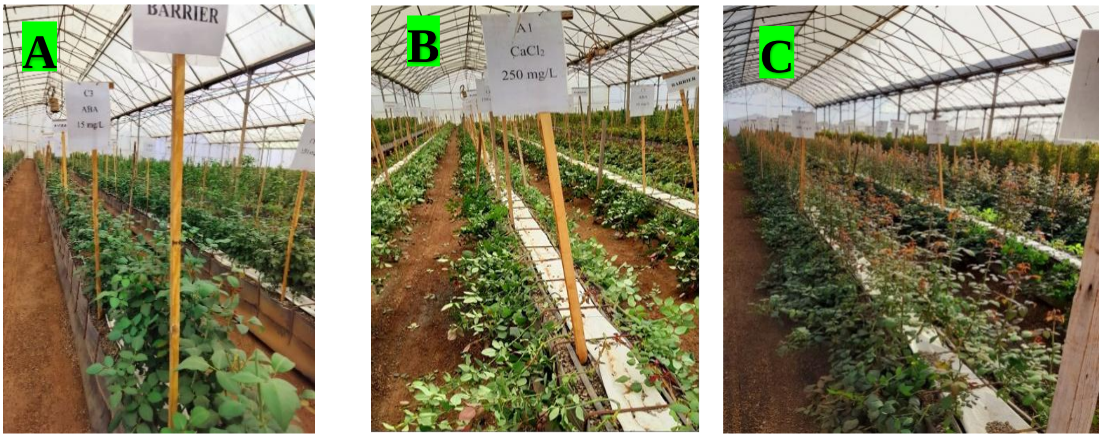

Plate 6: Before First Bending (A), After First Bending (B), and Selective Bending and Sanitation Done in the Greenhouse (C)

3.4.7 Selective Bending

Selective bending was done at an interval of 14 days after the initial bending to keep the bedding crop rejuvenated. Weak, blind and short stems were selected and used as bedding plants (Plate 6C). The stems were disbudded and bend in sunny weather when the turgidity of the plant was low to prevent breakage. The bending plants were maintained at three to five stems.

3.4.8 Sanitation

Sanitation involved weekly removal of any unwanted materials that could affect the overall survival and productivity of the plant through sweeping and weeding to keep the greenhouse clean (Figure 8C). The unwanted materials involved yellow leaves, dry leaves and stems from the bending material.

3.4.9 Fertigation and Crop Protection

Fertigation was through the closed hydroponic system. The mixing tank was filled with half the water. Agitation was done while adding phosphoric acid, mono potassium phosphate, mono ammonium phosphate, potassium Nitrate, magnesium nitrate, potassium sulphate, boric acid, trace elements (copper sulphate, zinc sulphate, borax, manganese sulphate and sodium molybdate) and magnesium sulphate, respectively according to the farm based fertilizer recipe. Calcium nitrate and iron were put in a different tank to prevent formation of the insoluble Calcium Phosphate and the unavailability of Iron. The tanks were filled up with plain water to the required volume of solution and target EC supply. They were agitated for 2 to 3 hours to prevent sedimentation and then supplied to the field in 8 cycles of 3 minutes each in hot weather and 6 cycles of 3 minutes each in cold weather at a pH of 5.5 to 6.5 with an EC of 2.0.

To avoid blockage of the driplines, cleaning with hydrogen peroxide and nitric acid was done fortnightly. A 50% concentration of hydrogen peroxide was injected and left to remain in the line for 30 minutes. Water was then flushed through the dripline to remove dirt. Nitric acid solution at a rate of 2L/3L of water was injected in the dripline and left to remain in the system for 4 hours before flushing. After 4 hours the drip lines endings were opened and flushed out with plain water. Dripper capacity was done immediately to confirm the efficiency of the cleaning.

Major pest involved two-spotted red spider mites (*Tetranychus urticae*), Thrips (*Frankliniella occidentalis*), green peach aphid (*Myzus persicae*) and caterpillar (*Helicoverpa armigera* and *Thaumatotibia leucotreta*). The diseases involved Downey mildew (*Peronospora sparsa*), Powdery mildew (*Podosphaera pannosa*) and Grey mould (*Botrytis cinerea*). Scouting for pests and diseases was done three times a week. It involved visual checks for live pests, honey dew and pest damage on the crop. Monitoring for diseases involved checking for spores using a magnifying lens. Idenfied Pest and diseases were managed through the Intergrated Pest Management approach (Appendix 6, Appendix 7). Copper-based fungicide was applied immediately after planting as a preventive control against fungal diseases.

3.4.10 Greenhouse Climate Management

Weather monitoring was done using an EL-USB-5 data logger of Serial Number 010180845 from Indonesia. The data logger recorded greenhouse humidity, temperature and dew point using a remote sensor and data collected daily (Appendix 6). An automatic fogging system was installed in the greenhouse to manage in-house humidity. Fans were also installed in the greenhouse for enhancing air circulation. Greenhouse temperature was maintained between 23 ˚C - 30 ˚C during the day and 12 ˚C – 16 ˚C during the night. Humidity was maintained between 70 % and 80%. When greenhouse humidity was 65% and below, fogging was done for 6 seconds at an interval of 3 minutes until the appropriate greenhouse humidity was achieved. During hot days, humidification was done through pouring water on the pathways between the greenhouse paths using horse pipes. When it was extremely hot and cold, the adjustable vents were opened to 80% to allow for air circulation. In rainy season the vents were closed to 5% to prevent rains from getting in the greenhouse. When the temperature was within the range, the vents and the side curtains were maintained at 50% to control temperature and air circulation.

3.4.11 Harvesting

In the first trial, harvesting was done 110 days after planting and 55 days after cutting back in the second trial. The flowers were harvested in the morning and in the evening when the temperature was low and atmospheric humidity high. Using a sharp secateur, the flowers were cut when at harvest stage 3 (the petals have started forming rings and showing bright colour) [Plate 7A]. The cut was made on the first true leaf above the point of origin of the flowering stem. During harvesting, the flowers were held in an upright position with the flower heads slightly above the shoulder to prevent mechanical damages. The secateurs were disinfected with 300 ppm sodium hypochlorite after every cut before moving to the next crop. After every 20 stems harvested, the flowers were taken to the shaded harvesting table and the quality examined. Conforming flowers were wrapped with nets in a conical shape and put in clean farm formulated postharvest solution in an egg tray bucket.

The postharvest solution had a temperature of 8˚C and a pH of 4.8. The harvested flowers were transported to the cold room for precooling using the flower transport line (Plate 7B). As part of the cold chain management, harvested flowers took at most 25 minutes in the greenhouse to mitigate the reduction of degree-hours and accumulation of heat in the harvesting solution. The flowers were held overnight in the cold storage 4 ± 2 ºC to remove the field heat before grading and vasing. The cold room was installed with UV air purifiers to prevent contamination of harvested flowers by botrytis.

Plate 7: Harvested Flowers for Postharvest Experiment (A) and Transportation of Flowers Using the Flower Transport Line (B).

3.5 Data Collection

After initial bending 3 stems in each experimental plot were tagged for data collection. The impacts of the growth regulators on growth of cut roses was assessed through counting the number shoots and petals produced, measuring the stem length, stem diameter, flower bud size, leaf area, chlorophyll content, and number of days to flower maturation. The impacts of the growth regulators on postharvest quality of cut roses was assessed through chlorophyll content, petal colour, Weight loss and vase life. The flowers for postharvest data collection were tagged at preharvest before harvesting (Plate 8).

Plate 8: Tagged Plants for Data Collection at Postharvest

3.5.1 Number of Shoots

The number of shoots produced were obtained through counting of emerging lateral shoots after initial bending in the first experiment and after pruning in the second experiment (Plate 9). The counting was done at an interval of 2 days from day 5 to day 12 after first bending in the first experiment and after pruning in the second experiment.

Plate 9: Lateral Shoots

3.5.2 Stem Length

The length of the stems was measured from the point of attachment with the main crop to the tip of the flower using a tape measure (Plate 10A). The measurement was taken on the tagged plant before harvesting and recorded in cm.

3.5.3 Stem Diameter

Stem diameter was measured 5cm from the point of branching with the mother crop from one side of the stem to the other side for each tagged plant using a Vernier calliper (Plate 10A). The measurement was recorded in mm.

3.5.4 Flower Bud Size

The flower head size was measured at harvesting stage using a Vernier calliper. The height measurement was taken from the bottom of the sepal to the tip of the petal (Plate 10C). Measurement was recorded in cm.

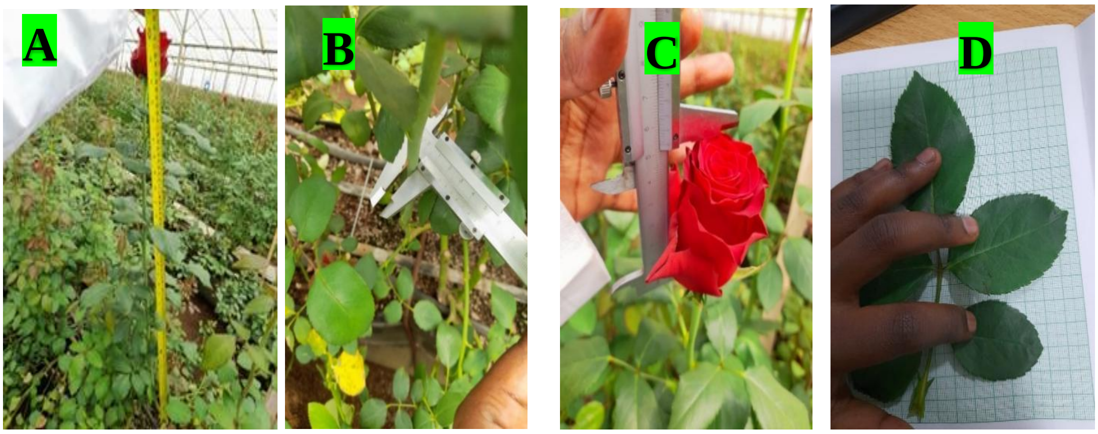

Plate 10: Measurement of Stem Length (A), Stem Thickness (B), Flower Head Size (C), and Leaf Area (D)

3.5.5 Leaf Area

To measure the leaf area, the plants were divided into 3 sections; the upper, middle and lower part. The leaves were plucked from each section during harvesting and placed on the graph paper, the leaves were outlined with a pen (Plate 10D). The area of the leaf was calculated by counting the number of squares within the outline of the leaf and multiplying the number by the area of each square. The measurement was recorded be in cm^2.

3.5.6 Bent peduncles

It involved counting the number of stems with bent peduncles whenever they occurred in each treatment. The counted stems were recorded and discarded at the field level.

3.5.7 Number of Suckers

It involved counting the number of auxiliary shoots desuckered on the tagged plants. The data was collected daily.

3.5.8 Chlorophyll Content

Chlorophyll content of the fully grown leaves (fourth alternate leaves from the top) was measured using a chlorophyll meter. The Chlorophyll Meter measured chlorophyll content through remote sensing without destruction of the leaf tissue. In the field, the first measurement was done two weeks after planting. Subsequent measurement was done at an interval of 7 days. In postharvest, the measurement was done before vasing and during the termination of the vase life. It was recorded in SPADS.

3.5.9 Flush Days

Data on flush days was obtained through counting the number of days the tagged plant took to reach the open flower harvesting stage. In the first experiment, the days were counted from the time of first bending to the time of harvesting stage. In the second experiment, the days were counted from the day of pruning to the harvesting day.

3.5.10 Number of Petals

It was done during the termination of vase for individual stems per treatment. Petals were plucked from the flower buds. Counting and recording the number of petals was done. (Plate 11).

Plate 11: Counting Number of Petals

3.5.11 Petal Colour

The petal colour was measured using a Chroma meter. The Chroma meter was placed on top of the petals to measure the intensity of the light reflecting off the petals in three different wavelengths (L*, a* and b*). The wavelengths were used to generate numerical values for the colour intensity of the petals. The mean of the numerical value per treatment was used to measure the petal colour variation at 2 days' interval until when the aesthetic value of the flower was lost.

3.5.12 Weight Loss

The initial weight of the freshly harvested produce of cut rose was measured immediately after harvesting on the tagged plants (Plate 12). The percentage of weight loss of each treatment was determined at an interval of two days. The weight difference percentage was calculated by differences between initial and final weights divided by the initial weights of cut stems. The weight loss (%) was calculated as;

$$\text{Weight loss\%} = \frac{\text{Initial weight} - \text{final weight}}{\text{Initial weight}} \times 100$$

Plate 12: Collection of Weight Loss Data

3.5.13 Vaselife

Vase life involved the period from the first day when cut roses was placed in vase solutions until they lost their ornamental value (Plate 13). The loss of ornamental value was defined by drooping, rotting, petal falling, leaf yellowing, complete leaf abscission, maximum opening and changing of petal colour. The data was collected daily and vase life recorded in days. Vase life days was calculated as V=S1D1 + S2D2 +…. + SnDn. Where S was stems in fraction of the total stems vased and D was the number of days' individual stem (s) had retained their ornamental value.

Plate 13: Vaselife Test for Flowers

3.6 Data Analysis

The collected data was analysed using analysis of variance (ANOVA) using SAS version 9.4. Significant means were separated using Least Significance Difference (LSD) at α=0.05.

The statistical model for the field experiment was;

$Y_{ijkl}= \mu + R_l + A_i + B_j + C_k + e_{ijkl}$

where,

i = A1, A2, A3 j = B1, B2, B3 k = C1, C2, C3 l = 1, 2, 3

μ = Overall mean

R_l = Block effect

A_i = Effect of the i^{th} treatment (Calcium chloride)

B_j = Effect of the j^{th} treatment (Cytokinin)

C_k = Effect of the k^{th} treatment (Abscisic acid)

e_{ijkl} = Random error

The statistical model for the postharvest experiment was;

$Y_{ijk} = \mu + A_i + B_j + C_k + e_{ijk}$

where,

i = A1, A2, A3, A4, A5, A6, A7, A8, A9 j = B1, B2, B3, B4, B5, B6, B7, B8, B9

k = C1, C2, C3, C4, C5, C6, C7, C8, C9

μ = Overall mean

A_i = Effect of the i^{th} treatment (Calcium chloride)

B_j = Effect of the j^{th} treatment (Cytokinin)

C_k = Effect of the k^{th} treatment (Abscisic acid)

e_{ijk} = Random error

3.7 Ethical Consideration

The study was done ethically by ensuring the confidentiality of the collected data. A clearance form was obtained from the Chuka University ethics and research committee before data collection by submitting a proposal for approval (Appendix 2). The research permit was acquired from National Commission for Science Technology and

Innovation (NACOSTI) before commencing the research (Appendix 3). Data collection and analysis procedures upheld a high level of integrity. The principles involved in research ethics were followed. Plagiarism was avoided in the entire research process by acknowledging the sources of all information obtained elsewhere through citations.

CHAPTER FOUR

RESULTS AND DISCUSSION

4.1 Effect of Calcium Chloride, Cytokinin and Abscisic Acid Application on Growth of Rose Cut-Flower

The effect of Calcium Chloride, Cytokinin and Abscisic acid on growth of cut rose's cv. Red Rhodos was assessed at different rates. The analysis of variance showed that number of shoots, stem length, stem diameter, flower bud size, leaf area, chlorophyll content, number of suckers, physiological disorders, flush days, and number of petals were significantly affected by the different rates of application of Calcium Chloride, Cytokinin and Abscisic acid. Moreover, Flush 1 had better results compared to flush 2. This is because, flowers in flush 1 emerged from the basal shoots which are always more vigorous while flowers in flush 2 emerged from lateral shoots which are always less vigorous compared to basal shoots.

Additionally, the greenhouse average temperature ranged between 23 ˚C and 27 °C. Relative humidity was maintained at an average of 70% to 80% (Table 1). The different growing temperatures and humidity caused varying morphological characteristics and harvest dates. Lower temperatures and higher humidity in the first flush (Table 1) yielded flowers with longer lengths (Table 3), bigger stem diameter, bigger flower bud size (Table 4), larger leaves (Table 5), more chlorophyll content (Table 6), and more flush days (Table 9) compared to flush 2. It is possible that lower temperatures slowed the growth causing the plant to consume less energy and store more sugar therefore improving the plant morphological traits. Similar outcomes were reported by Younis *et al.* (2013) who observed that lower temperature than the recommended causes stems to elongate and flush intervals to lengthen.

Table 1: Greenhouse Environment Weather Condition in Flush 1 and Flush 2.

Flush	Month	Temperature (^{0} C)			Relative Humidity (%)		
		Minimum	Max	Mean	Minimum	Max	Mean
Flush 1	August	13.0	29.6	23.43	48	93.9	81.8
	September	14.3	33.2	24.66	43	89.2	77.6
	October	13.8	32.9	23.2	45	91	79.3
Flush 2	November	14.9	33.4	25.67	41	90	80.4
	December	16.9	35.8	26.54	39	81	73.5
	January	17.1	36.4	27.31	36	84	71

4.1.1 Effect on Number of Shoots

The analysis of the treatment effect showed that the application of Calcium Chloride (250, 500, and 750mg/L) had no significant ($p < 0.05$) effect on the number of shoots at different rates both in flush 1 and 2 (Appendix 10). Calcium chloride is a salt. When in excess, salts cause detrimental effects in plant growth. It could be that the amounts of calcium chloride applied in the current experiment were too low to affect the number of shoots produced in rose flower. It is also possible that at concentrations higher than 750 mg/L Calcium Chloride has a potential of causing detrimental impacts on shoot growth. According to Haque *et al.* (2017), salt treatment has a potential to decrease shoot emergence. Additionally, Pawłowska and Bach (2010) observed that rising concentrations of calcium chloride had a detrimental impact on rose shoot regrowth and proliferation. El-Enany (1997) also discovered that a high salt level prevented the regrowth of shoots from cotyledons and hypocotyls in tomatoes.

The study also showed that application of Cytokinin (150, 250, and 350mg/L) had a significant ($p < 0.05$) effect on the number of shoots at different rates both in Flush 1 and 2 compared to the control (Table 2; Appendix 10). At 10 days after initial bending in flush 1 and pruning in the flush 2 respectively, an average number of 4.22 and 2.78 shoots, 4.11 and 2.67 shoots, and 3.89 and 2.56 shoots was recorded when Cytokinin was applied at 350, 250, and 150 mg/L. On the other hand, 3.22 and 1.77 shoots were recorded in the control experiment both in flush 1 and flush 2, respectively (Table 2). It could be that the application of Cytokinin in Rhodos variety accelerated cell divison in the meristem encouraging development of more shoots. It has been reported that Cytokinin enhances growth of more shoots because it encourages cell division and the development of plant tissues especially in meristematic parts responsible for new shoot emergence (Cammarata *et al*., 2019).

The findings of this study were in agreement with Roy *et al*. (2017) who did research on the effect of pre-plant soaking of corms in cytokinin on sprouting, vegetative growth and corm formation in gladiolus (*Gladiolus grandiflorus* L.). They observed that at 300 ppm cytokinin increased the most shoots per corm in variety Jessica. Priyanka *et al.* (2018) also found out that 300 ppm of cytokinin application in gladious had the highest spike production per plant. Additionally, Azizi *et al*. (2015) in the study of auxin and cytokinin in rice reported that, Cytokinin encouraged the division of cells and played an integral role in several aspects of plant growth, such as the activity and generation of shoot meristems.

The application of Abscisic acid (5, 10, and 15 mg/L) had no significant ($p < 0.05$) effect on the number of shoots at different rates both in flush 1 and 2 (Appendix 8). Abscisic Acid is a growth retardant but at low concentrations, it may have no impact on plant growth. Therefore, it is likely that the amounts of Abscisic Acid applied in the current experiment were too low to affect the number of shoots in rose flower, and at concentrations higher than 15 mg/L Abscisic Acid has a potential of causing detrimental impacts on shoot growth. According to Vysotskaya *et al*. (2018), ABA inhibits growth. This observation was supported by Brookbank *et al.* (2021) who reported that large concentrations of exogenously administered ABA restricted growth and that endogenous ABA built up under a range of stressful circumstances, simultaneously causing the stressed plant to grow less.

Table 2: Effect of Calcium Chloride, Cytokinin and Abscisic Acid on the number of lateral shoots in flush 1 and 2.

Treatment	Rate of Concentration in mg/L	Number of lateral shoots in Flush 1	Number of lateral shoots in Flush 2
$CaCl_2$	250	3.22b	1.78b
$CaCl_2$	500	3.22b	1.89b
$CaCl_2$	750	3.11b	1.78b
Cytokinin	150	3.89a	2.56a
Cytokinin	250	4.11a	2.67a
Cytokinin	350	4.22a	2.78a
Abscisic Acid	5	2.89b	1.67b
Abscisic Acid	10	2.89b	1.33b
Abscisic Acid	15	2.78b	1.33b
Control	0	3.22b	1.78b
	LSD	0.85	0.61
	CV	27.50	33.15

Means followed by the same letter(s) along the column are significantly different at 5% probability level.

4.1.2 Effect on Stem Length

Calcium chloride application at 250, 500, and 750 mg/L had no significant effect on stem length ($p < 0.05$) (Appendix 11; Table 3). The impact of Calcium Chloride on the growth and elongation of a plant can be affected by multiple physiological, and environmental variables. It could be that the uptake of exogenously applied Calcium was affected by the lower temperatures observed during the vegetative growth. This therefore, affected physiological activity of Calcium Chloride causing it to have no significant effect on stem length. Moreover, Aldon *et al.* (2018) explained that the amount of calcium that plants require varies depending on the stage of development and the species of the plant. Therefore, if a plant is not lacking in calcium, then it could be unresponsive to more of it. This also explains why the application of Calcium chloride had no effect on the stem length of the Rhodos variety in the current study. Contrary to the current study, Khalaj *et al*. (2023) reported that application of calcium on Gladiolus increased the length of the flower and the spike. They observed that the length increased with the increasing concentrations of calcium. Additionally, Mehdi *et al.* (2015) observed that the stem length of cut rose cv. “Dolce Vita” increased with exogenous application of Calcium chloride. However, at high concentration of calcium chloride the stem length reduced.

This study revealed that applying Cytokinin at 150, 250, and 350 mg/L positively affected the lengthening of stems compared to control at 0 mg/L. The longest stems were observed in 350 mg/L with 83.22 cm and 82.56 cm in flush 1 and 2 respectively (Appendix 11; Table 3). It may be that the active role of cytokinin in cell division and elongation led to longer stems of cut rose flowers in this study. Jacqmard *et al*. (2019) also reported that application of cytokinin caused the cells in the stem tissues to divide and elongate more in plants. Gabrel *et al*. (2018) reported a similar pattern of findings using the Chrysanthemum plant. They observed that Chrysanthemum morifolium cv. "Zambla White" reached the highest plant height when exposed to high concentrations of cytokinin (200 ppm).

Where Abscisic acid was applied at 5, 10, and 15 mg/L it significantly reduced the length of the stems ($p < 0.05$). The shortest stems at 68.56 and 68.78 cm were noted with application of ABA at 15 mg/L in flush 1 and 2, respectively (Appendix 11; Table 3). Based on the findings of this study, Abscisic acid probably reduced the stem length of Rhodos variety by inhibiting stem elongation because of its ability to retard growth. According to Chen *et al*. (2020), Abscisic acid stimulates stem cell dormancy and inhibition of differentiation in the principal root meristem. Kishor *et al*. (2022)

indicated that in stressful conditions, elevated levels of Abscisic acid leads to restricted stem development as the plants adapted to preserve resources and water. This is achieved by Abscisic acid through controlling the expression of genes related to cell division and elongation (Sun *et al*., 2018). Similar to the current study, Vu *et al*. (2020) reported that the height of coffee seedlings reduced with increasing concentration of Abscisic acid. It is very likely that application of ABA reduced cell division or elongation which expressed in terms of short stems. Vu *et al*. (2015), also demonstrated that the use of Abscisic acid affected plant growth, significantly reducing the height of tomato seedlings.

Table 3: Effect of Calcium Chloride, Cytokinin and Abscisic Acid on stem length produced in flush 1 and 2.

Treatment	Rate of Concentration in mg/L	Stem length in Flush 1 (cm)	Stem length in Flush 2 (cm)
$CaCl_2$	250	79.22b	78.33b
$CaCl_2$	500	79.67b	78.44b
$CaCl_2$	750	77.89b	76.11b
Cytokinin	150	82.44a	81.44a
Cytokinin	250	82.67a	82.44a
Cytokinin	350	83.22a	82.56a
Abscisic Acid	5	69.22c	69.33c
Abscisic Acid	10	68.89c	69.00c
Abscisic Acid	15	68.56c	68.78c
Control	0	78.67b	77.33b
	LSD	3.93	4.21
	CV	5.43	6.14

Means followed by the same letter(s) along the column are not significantly different at 5% probability level within each flush.

4.1.3 Effect on the Stem Diameter and Flower Bud Size

Analysis of treatment effect revealed that the application of Calcium chloride at 250, 500, and 750 mg/L had no significant effect ($p < 0.05$) on the diameter of the stem and the flower bud size compared to the control at 0 mg/L (Appendix 12; Appendix 13; Table 4). These observations imply that, in this specific variety, there is no direct relationship between the administration of calcium chloride and stem and flower diameter. The findings of the study on the effect of Calcium chloride on stem and flower diameter was similar to the findings of Sabah *et al*. (2019) in snapdragon plants (Antirrhinum majus cv. butterfly). They reported that when applied independently calcium chloride had no discernible effect; however, when combined with humic acid at a higher concentration, the stem morphology and flower production of Snapdragon significantly enhanced. Similarly, on a research in Rosa hybrida cultivars and calcium application, Oloo-Abucheli (2018) reported that field treatment of cut roses with calcium had no effect on stem and flower diameter. According to Ali and Abd Asal (2023) calcium is necessary for many physiological functions in plants, such as the creation and integrity of cell walls, however calcium chloride by itself usually has no direct impact on stem diameter.

Cytokinin at 150, 250, and 350 mg/L had no significant effect ($p < 0.05$) on the diameter of the stem and the flower bud size compared to the control at 0 mg/L (Appendix 12; Appendix 13; Table 4). Cytokinins mainly promote the formation of shoots by stimulating cell division in the meristem. Even though more cell division can lengthen shoots overall, this does not always translate into a corresponding stem or flower diameter increase. Aside from cytokinin regulation, additional variables that affect stem diameter include; physiological factors (development of vascular bundles), environmental factors (temperature and humidity), and cultural factors (feeding, and crop balancing) [Xiang *et al*., 2019].

Abscisic Acid at 5, 10, and 15 mg/L had no significant effect ($p < 0.05$) on the diameter of the stem and the flower bud size compared to the control at 0 mg/L (Appendix 12; Appendix 13; Table 4). It is very likely that in this specific variety, there is no direct relationship between the administration of Abscisic acid and stem and flower diameter. It is also possible that the effect of the varying concentrations of Abscisic acid were overshadowed by the activity of other hormones in the plant. This is because Abscisic acid is most effective in stressful conditions but during the growth of Rhodos variety the growing conditions were favourable. In research on Short-and long-term responses of pepper seedlings to ABA exposure Ban *et al.* (2017) observed that while stem diameter increased in pepper, it was not influenced by Abscisic Acid. Contrary to the past studies by Al-Deeb *et al.* (2023), Abscisic Acid was unable to significantly affect stem diameter. This is probably because, stem and flower diameter are influenced by multiple factors including genetic and environmental. According to Skubacz & Daszkowska-Golec (2017), a complicated interaction between variables exists where the effects of Abscisic Acid are obscured by other physiological functions or outside stimuli, underscoring the complex interactions between Abscisic Acid signaling mechanisms and biological processes in plants.

Table 4: Effect of Calcium Chloride, Cytokinin and Abscisic Acid on the stem diameter and flower bud size produced in flush 1 and 2.

Treatment	Rate of Concentration in mg/L	Stem diameter (mm)		Flower bud size(cm)	
		Flush 1	Flush 2	Flush 1	Flush 2
$CaCl_2$	250	8.44a	7.11a	5.30a	5.23a
$CaCl_2$	500	8.83a	7.56a	5.34a	5.23a
$CaCl_2$	750	8.89a	7.89a	5.4a	5.44a
Cytokinin	150	8.06a	6.83a	5.3a	5.16a
Cytokinin	250	8.56a	7.17a	5.13a	5.15a
Cytokinin	350	8.61a	7.33a	5.38a	5.36a
Abscisic Acid	5	8.78a	7.39a	5.45a	5.51a
Abscisic Acid	10	8.06a	6.81a	5.39a	5.42a
Abscisic Acid	15	8.44a	7.06a	5.26a	5.16a
Control	0	8.72a	7.39a	5.35a	5.30a
	LSD	1.08	1.36	0.33	0.36
	CV	13.53	19.92	6.41	7.22

Means followed by the same letter(s) along the column are not significantly different at 5% probability level for each flush.

4.1.4 Effect on Leaf Area

In the current study it was observed that different concentrations of Calcium Chloride (250, 500 or 750 mg/L) had a no effect on the leaf area of cut roses “Rhodos variety” (Appendix 14; Table 5). These outcomes imply that, in this specific variety, there is no direct relationship between the administration of calcium chloride and leaf area. According to El Habbasha and Ibrahim (2015), the impact of Calcium on the growth of a plants can be affected by multiple physiological, and environmental variables. Moreover, Aldon *et al*. (2018) explained that the amount of calcium that plants require varies depending on the stage of development and the species of the plant. This explains why the application of different concentrations of calcium chloride applied at different timings had no effect on the leaf area of cut roses “Rhodos.” Contrary to the findings, Youssef *et al.* (2017) reported that foliar treatments of calcium chloride resulted in a considerable increase in average leaf area when applied more than one time and at high concentrations in lettuce. Similarly, Ahmed and Rab (2019) reported a significant influence of calcium concentrations, application timing, and their combination on leaf area of gladiolus.

In this study leaf area was significantly influenced by Cytokinin application. The biggest leaf size of 76.67 and 72.44 cm^2 was observed with application of 350 mg/L Cytokinin in flush 1 and 2 respectively (Table 5). In this study, it is very likely that Cytokinin increased the leaf area of the Rhodos variety because of its ability to promote cell division and enhance cell expansion during the developmental stage of the leaf. According to Wu *et al.* (2021), cytokinin enables shoot apical meristems to continue growing and produce stem cells that eventually generate leaves during the early stages of leaf production.

The findings of this study are in agreement with those of Mondal & Sarkar (2018) studied on the Hybrid tea rose cv. Bugatti during spring-summer months. They observed that maximum leaf area was obtained from plants with cytokinin treatment compared to the control experiment that had the least leaf area. Similar outcomes were also noted by Sardoei (2014) who reported that leaf area increases with increasing concentration of growth regulators (Gibberellic Acid and Benzyl Adenine). Therefore, based on the findings of this study, the higher the concentration of the cytokinin application the larger the leaf and the higher the surface area for photosynthesis. Tamayo-Tenorio *et al*. (2017) reported that plants with larger leaves are able to effectively make their own food by sufficiently converting light energy from sun into chemical energy which results to growth of strong and healthy plants with better quality and enough food reservoir for postharvest life of the plant.

During the study, Abscisic acid treatment decreased the leaf area of the Rhodos variety compared to the control. It is possible that exogenous application of Abscisic acid induced stomata closure. This affected the gaseous exchange in the crop causing a reduction in photosynthesis which affected the growth and enlargement of leaves. Chen *et al.* (2020) indicated that when Abscisic acid simulated the impacts of water stress it causes reactions that are comparable to those brought on by internally produced Abscisic acid. Additionally, applying Abscisic acid cause stomatal closure, which lowers the amount of carbon dioxide that enters the leaf during photosynthesis (Negin *et al*., 2019). This therefore causes a limitation in the synthesis of carbohydrate which is crucial for cell division and proliferation therefore reducing the leaf area.

Similarly, Khaleghnezhad *et al.* (2021) reported that under all moisture levels in the experiment, the application of Abscisic acid exhibited a declining trend in both photosynthetic rate and leaf area in Dracocephalum moldavica L. under drought stress. They also observed that leaf area was drastically decreased by the decline in leaf growth in increasing concentrations of Abscisic acid. Roeder *et al.* (2023) also reported that after 19 days of Abscisic acid treatment, the expected leaf area was 20% smaller. Additionally, Abscisic acid treatment has compromised cell division, reduced leave growth and less leaf area Chen *et al.* (2022). It could be that this reduces the synthesis of carbohydrates in the plant, negatively impacting the growth and the quality of the cut flowers.

Table 5: Effect of Calcium Chloride, Cytokinin and Abscisic Acid on the leaf area produced in flush 1 and 2.

Treatment	Rate of Concentration in mg/L	Leaf Area in Flush 1 (cm^2)	Leaf Area in Flush 2 (cm^2)
$CaCl_2$	250	65.89b	63.17b
$CaCl_2$	500	66.94b	63.78b
$CaCl_2$	750	68.94b	63.89b
Cytokinin	150	75.00a	71.06a
Cytokinin	250	76.11a	71.44a
Cytokinin	350	76.67a	72.44a
Abscisic Acid	5	61.00c	54.11c
Abscisic Acid	10	61.11c	54.94c
Abscisic Acid	15	61.89c	57.22c
Control	0	66.06b	63.28b
	LSD	5.96	9.37
CV		9.33	15.82

Means followed by the same letter(s) along the column are not significantly different at 5% probability level within each flush.

4.1.5 Effect on Chlorophyll Content

Although Calcium Chloride at 250, 500 and 750 mg/L, had varying means, the treatments had no statistically significant effect ($p < 0.05$) on chlorophyll content 3 weeks after first application of treatments (after initial bending and pruning) both in flush 1 and 2. After the second application of treatments (at maroon stage) 500 and 750 mg/L of Calcium chloride significantly ($p < 0.05$) influenced chlorophyll content compared to the control (Appendix 15). Despite the application of treatments 2 times during pre-harvest of the Rhodos variety, 250 mg/L of Calcium chloride had no significant effect on chlorophyll content. The effects of 500 and 750 mg/L of Calcium Chloride on chlorophyll content was observed after a second application of the treatments. Therefore, based on these findings it is evident that Calcium translocation is slow and can influence the levels of chlorophyll content in roses upon build up within the plant.

The observations were in agreement with Zomorrodi *et al* (2022) who observed that applications of Calcium Chloride significantly increased the amount of chlorophyll in Periwinkle, particularly at the highest concentrations. Similar outcomes were observed by Xie *et al.* (2014) who reported that preharvest application of calcium chloride had a significant role in maintaining the chlorophyll content in of cowpea (*Vigna unguiculata* (L.). Additionally, Kazemi (2014) observed similar outcomes in tomato and Ilias *et al.* (2007) in Okra. It conceivable that the uptake and translocation of the calcium ions by the plant was slow (Pathak *et al*., 2021) and also, the effectiveness of calcium chloride on chlorophyll content could have been based on the amount of calcium accumulated by the plant (Guo *et al*., 2023). Therefore, this explains why the effect of calcium chloride was only visible in Rhodos variety after more than one time of application at high concentrations.

Treatment analysis indicated that cytokinin application at 250 and 350 mg/L had a significant effect on chlorophyll content ($p < 0.05$) 3 weeks after first application of treatments (after initial bending and pruning) both in flush 1 and 2. Application of 350mg/L had the highest means of 53.81 SPADS and 46.8 SPADS in flush 1 and 2. Cytokinin at 150 mg/ had no statistically significant effect ($p < 0.05$) on chlorophyll content compared to the control (0 mg/L) [appendix 15]. After the second application of treatments (at maroon stage), 150, 250, and 350mg/L of cytokinin significantly ($p < 0.05$) influenced chlorophyll content compared to the control (Appendix 10). While application of 350 mg/L of Cytokinin was not significantly different from application of 250 mg/L, where 350mg/L was applied higher chlorophyll content of 72.95 and 70.77 SPADS was observed in flush 1 and 2, respectively (Table 6). The effect of

150mg/L of Cytokinin on chlorophyll content was observed after a second application of the treatments. These findings indicate that cytokinin substantially influenced the concentration of chlorophyll content in the leaves of Rhodos variety 3 weeks after first application and at week Seven after second application in flush 1 and 2, respectively. Therefore, based on these findings it could be that cytokinin have potential of influencing chlorophyll content when their concentration within the crop is high.

Hormones gradually build up in target tissues and become more effective following 3 or more weeks of treatment application (Dobránszki & Mendler-Drienyovszki, 2014). This probably explains why the impact of cytokinin on chlorophyll content was evident after 3 weeks. Sosnowski *et al*. (2023) reported that floral induction caused fluctuations in endogenous cytokinin levels which affected the amount of chlorophyll in the leaves of plants. This therefore justified the need for a second application of cytokinin treatment in Rhodos variety when the plants were at maroon vegetative stage of growth. It very likely that the effects of cytokinin on chlorophyll content are more evident on higher concentration (250 and 350 mg/L) than on lower concentration (150 mg/L). This explains why 150 mg/L had no significant effect on chlorophyll content even after 3 weeks of first application of treatments and was less significant than 250 and 350 mg/L at seven weeks after second application of treatments.

In other studies, when cytokinin was sprayed to alfalfa at different quantities, ranging from 0.025% to 0.20%, a notable rise in the amount of chlorophyll was observed (Skalska, 1992). Similarly, Cavusoglu *et al.* (2021) observed that the highest concentrations of cytokinin application substantially increased the amounts of Chlorophyll a and b on pepper *(Capsicum annuum* L.). They also reported that when cytokinin was applied externally to the pepper plant, it promoted the growth of chloroplasts and increased chlorophyll production, decreased chlorophyll degradation, and maintained photosynthetic activity. Research by Talla *et al*. (2016) demonstrated that cytokinins positively affected the amount of photosynthetic proteins in plants such as the proteins that attach to chlorophyll and form chlorophyll-protein complexes, which enhance chlorophyll content in plants.

Although Abscisic Acid at 5, 10, and 15 mg/L, had varying means, the treatments had no statistically significant effect ($p < 0.05$) on chlorophyll content 3 weeks after first application of treatments (after initial bending and pruning) both in flush 1 and 2. Despite the application of treatments 2 times during pre-harvest of the Rhodos variety, 5 mg/L Abscisic Acid had no significant effect on chlorophyll content. Compared to the control, application of 10 and 15 mg/L of Abscisic Acid significantly reduced the chlorophyll content with application of 15 mg/L having the lowest chlorophyll content of 64.74 and 65.13 SPADS in flush 1 and 2, respectively (Table 5; Appendix 15). The effects of 10 and 15 mg/L of Abscisic Acid on chlorophyll content was observed after a second application of the treatments application while after first application (at week 3) there was no significant effect. It is likely that endogenous level of abscisic acid increased with continued exogenous application of higher concentration of treatments 10 and 15 mg/L which reduced the chlorophyll content in the Rhodos variety. It is also possible that the increase in endogenous Abscisic Acid stimulated the aging process in plants therefore causing degeneration of chlorophyll within the leaves.

According to Yang *et al.* (2014), genes responsible for degradation of chlorophyll are strongly expressed when the concentration of Abscisic Acid is high. This possibly explains why the chlorophyll content was significantly reduced after the second application of 10 and 15 mg/L and why 5 mg/L had no effect on the chlorophyll content. Wang *et al.* (2018) in a study in Arabidopsis observed similar outcomes as the current study. They reported a significant decrease in chlorophyll content and yellowing in plants treated with Abscisic Acid. According to Gao *et al.* (2016), Abscisic Acid is known to cause leaf senescence by causing the expression of several genes linked to senescence or chlorophyll breakdown in Arapidopsis.

Table 6: Effect of Calcium Chloride, Cytokinin and Abscisic Acid on the chlorophyll content in flush 1 and 2.

Treatment	Rate of Concentration in mg/L	Flush 1		Flush 2	
		At week 3	At week 7	At week 3	At week 7
$CaCl_2$	250	47.27b	66.57d	41.24b	66.37d
$CaCl_2$	500	47.57b	68.17c	41.88b	67.44c
$CaCl_2$	750	48.74b	68.37c	42.42b	67.54c
Cytokinin	150	48.18b	70.15b	42.29b	68.33b
Cytokinin	250	52.48a	71.84a	46.36a	70.66a
Cytokinin	350	53.81a	72.93a	46.80a	70.77a
Abscisic Acid	5	47.17b	65.09d	40.51b	66.15d
Abscisic Acid	10	46.73b	64.75e	40.27b	65.58e
Abscisic Acid	15	47.48b	64.74f	41.34b	65.13f
Control	0	48.34b	66.34d	42.39b	66.15d
	LSD	2.90	2.66	3.68	2.34
	CV	6.33	7.21	9.35	3.77

Means followed by the same letter(s) along the column are not significantly different at 5% probability level within each flush.

4.1.6 Effect on the Number of Suckers

Compared to the control (0 mg/L), Calcium Chloride application at 250, 500, and 750 mg/L and Abscisic Acid application at 5, 10, and 15 mg/L on Cut roses "Rhodos variety" were observed to have no effect on the number of suckers produced per plant (Table 7; Appendix 16). Calcium Chloride is primarily responsible of strengthening cell wall while Abscisic Acid responsible of plant response to stress. It could be that Abscisic Acid influenced the response of Rhodos variety to stresses resulting from environmental fluctuations while Calcium Chloride strengthened the cell walls while playing no active role in regulating the emergence and development of suckers. According to Müller and Leyser (2011), hormones including cytokinins, and auxin control the shoot branching and development of auxiliary shoots. Moreover, Demidchik *et al.* (2018) reported that calcium is definitively attributed to the strengthening and stabilization of the cell wall. Vishwakarma *et al*. (2017) also reported that Abscisic Acid is majorly characterised with stress response in plants. The development of suckers, which includes branching from the primary stem, is primarily associated with growth processes instead of stress reactions (Salama & Elsherbeny, 2016) and rigidity of cell walls (Zhang *et al*., 2021).

Analysis of Variance revealed that Cytokinin application at 150, 250, and 350 mg/L significantly ($p < 0.05$) increased the number of suckers on Rhodos variety during production. Application of 350 mg/L had the highest number of suckers at 17.83 and 15.67 both in flush 1 and 2 (Table 7; Appendix 16). While treatment with 150 and 250 mg/L cytokinin had varying means, they expressed no statistical difference. Cytokinin has the ability to enhance lateral growth and suppress apical dominance. This explains why Cytokinin application increased the number of suckers in the Rhodos variety in the current study. Wang *et al.* (2017) reported that cell division in the meristematic tissues is activated when cytokinin levels are high in a specific region of the plant, such as close to the nodes or along the stem. It is certain that because of the accelerated cell division branching is enhanced and therefore more suckers are formed. This explains the reason for more suckers in the highest concentration of cytokinin (350 mg/L) in this study.

Ragini *et al.* (2019) conducted a study to find out the effect of cytokinins on bulbous flower crops and observed that corms treated with cytokinin produced the maximum number of suckers per corm. Similarly, Manasa *et al*. (2017) conducted research on the influence of growth regulators on vegetative parameters of gladiolus cv. Summer Sunshine. The outcomes revealed that cytokinin recorded the highest number of suckers per plant. Reshmi and Sheela (2016) in a study on the effect growth regulators on heliconia varieties reported that cytokinin produced the highest number of total suckers in all the varieties of heliconia in experiment 1 and 2. While sucker's development in bulbous crop because of cytokinin application is favourable, in tea hybrid cut roses they are not favourable (Ragini *et al.,* 2019). This is because they grow rapidly and run the risk of starving the primary part of the plant off its nutrients and water. Consequently, the quality of the cut rose is compromised and the cost of labor increased.

Table 7: Effect of Calcium Chloride, Cytokinin and Abscisic Acid on the number of suckers produced in flush 1 and 2.

Treatment	Rate of Concentration in mg/L	Number of suckers in Flush 1	Number of suckers in Flush 2
$CaCl_2$	250	8.00c	7.67c
$CaCl_2$	500	7.83c	7.67c
$CaCl_2$	750	8.17c	8.00c
Cytokinin	150	13.17b	11.00b
Cytokinin	250	15.17b	13.00b
Cytokinin	350	17.83a	15.67a
Abscisic Acid	5	7.67c	7.00c
Abscisic Acid	10	8.33c	8.00c
Abscisic Acid	15	8.17c	8.00c
Control	0	8.33c	8.00c
	LSD	2.64	3.020
	CV	22.16	18.73

Means followed by the same letter(s) along the column are not significantly different at 5% probability level within each flush.

4.1.7 Effect on Bent Peduncle

Analysis of treatments indicated that application of Calcium Chloride at 750 mg/L significantly ($p < 0.05$) reduced the number of bent peduncle peduncle formation in the Rhodos variety, which promoted erect posture of the stems. It had the lowest number of bent peduncles of 2.0667 and 1.1333 in flush 1 and 2, respectively (Table 8). Application of Calcium Chloride at 250 and 500 mg/L had no significant influence on the number of bent peduncles compared to the control (0 mg/L). It is conceivable that the high application of Calcium chloride (750 mg/L) in Rhodos variety increased the endogenous calcium within the crop, creating strong cellular structure which countered the effect of auxins on altering plant wall plasticity. This therefore reduced the incidences of bent peduncles.

Abdolmaleki *et al.* (2015) reported that plants suffering from a calcium deficit may exhibit physiological problems such as weakening stems and heightened vulnerability to bending of their necks. Therefore, application of Calcium Chloride application of Calcium can reduce stem weakening and bending. Further, El-Beltagi *et al*. (2022) studied that at high concentrations of calcium cut roses can create more robust and resilient cellular structures of the cell wall in their stems. Based on the findings of this study, it is doubtless that growers can ensure optimum stem rigidity and structural integrity with reduced bent peduncles by preventing the signs of calcium deficiency in cut roses. The findings of this study are in agreement with those of Elshawa *et al.* (2023) who indicated that using calcium chloride pre-harvest treatments reduced neck bending in gerbera cut flowers.

During the entire growing, application of 150, 250, and 350 mg/L of Cytokinin; 5, 10, and 15 mg/L of Abscisic Acid had no significant influence on the number of bent peduncles compared to the control (0 mg/L) [Table 8; Appendix 17]. Auxin alters the plant wall plasticity and therefore causing cell differential growth. It is very likely that the involvement of auxins in differential growth in Rhodos variety overshadowed the activity of Cytokinin and Abscisic acid making them to have no effect on the bent peduncles. Prior research by Jing *et al.* (2020) has demonstrated that the bending is caused by relative variations in cell expansion and division on either side of the bud. Zaccai *et al.* (2009) also reported that the prevalence of bent peduncles is higher when plants are young and develop more quickly, primarily in the warmer months. Additionally, Philosoph- Hadas *et al.* (2005) reported that auxin concentrations in plants have the ability to drive differential growth, which causes bending. This is because auxin regulate plant growth, development, and reaction to external stimuli (Muday, 2001; Caumon & Vernoux, 2023).

Table 8: Effect of Calcium Chloride, Cytokinin and Abscisic Acid on the number of bent peduncles produced in flush 1 and 2.

Treatment	Rate of Concentration in mg/L	Bent Peduncles in Flush 1	Bent Peduncles in Flush 2
$CaCl_2$	250	2.83a	2.67a
$CaCl_2$	500	2.83a	2.67a
$CaCl_2$	750	2.07b	1.13b
Cytokinin	150	3.83a	3.33a
Cytokinin	250	3.83a	3.33a
Cytokinin	350	4.00a	4.00a
Abscisic Acid	5	4.17a	4.33a
Abscisic Acid	10	4.00a	4.00a
Abscisic Acid	15	4.17a	4.67a
Control	0	4.17a	4.33a
	LSD	2.10	3.30
	CV	50.42	59.96

Means followed by the same letter(s) along the column are not significantly different at 5% probability level within each flush.

4.1.8 Effect on Flush days

Compared to the control (0 mg/L), Calcium Chloride at 250, 500, and 750 mg/L, had no statistically significant effect on numbers of days taken by the flowers to mature (Table 9; Appendix 18). It is very likely that Calcium Chloride in the current study increased the rigidity of the cell wall and was not directly involved in influencing the growth and development of cut roses "Rhodos variety". Demidchik *et al*. (2018) also observed that calcium is definitively attributed to the strengthening and stabilization of the cell wall.

Analysis of variance indicated that Cytokinin application at 350mg/L significantly ($p < 0.05$) influenced the number of days taken by the Rhodos variety to mature (Appendix 18). Flowers treated with 350 mg/L of Cytokinin took 51.72 and 53.67 days. Compared to the control (0 mg/L), Cytokinin at 150 and 250 mg/L, had no statistically significant effect on numbers of days taken by the flowers to mature (Table 9; Appendix 18). Cytokinin promotes cell division and increase cell expansion. It is possible that at high concentration Cytokinin enhanced more cell division and expansion, lengthening the vegetative phase which delayed the plant from moving to the reproductive phase. This therefore increased the number of days taken to harvest flowers treated with cytokinin. It has been reported that Cytokinin essentially encourages cell division and vegetative development (Werner *et al*., 2021), delay senescence (Mantilla *et al*., 2021), and change nutrient distribution (Mok. 2019) therefore lengthening the time needed to harvest. Cho *et al*. (2022), reported that cytokinin prolongs the development period of the plant by encouraging the synthesis of additional vegetative tissue and preserving leaf functioning. Furthermore, cytokinins promote the branching on the stem by diminishing apical dominance, which makes the plant bushier and slow to blossom (Cao *et al.*, 2022). It is likely that high application of cytokinin in the present study encouraged vegetative growth and therefore delayed the time of the plant to move to the floral phase.

Abscisic Acid significantly ($p < 0.05$) reduced the number of days taken by the Rhodos variety to mature (Table 9; Appendix 18). Flowers treated with 15 mg/L of Abscisic Acid took 49.06 and 50.67 days to mature in flush 1 and flush 2, respectively (Table 9). Compared to the control (0 mg/L), Abscisic Acid at 5 and 10 mg/L had no statistically significant effect on numbers of days taken by the flowers to mature. It is conceivable that Abscisic acid accelerated the transition to the floral phase and caused a dormant-like state in the vegetative tissues to begin earlier. Because of its critical involvement in stress reactions, plants respond to environmental challenges like drought and cold by increasing their levels of Abscisic acid, which signals them to preserve resources and speed up reproductive growth (Vishwakarma *et al*., 2017; Shu *et al*., 2018). Furthermore, Abscisic acid controls stomatal closure, which aids in water management and further communicates to the plant that it must swiftly finish its life cycle (Abhilasha & Roy Choudhury. 2021). It is possible that the exogenous application of Abscisic Acid in the current study caused a dormant-like state in the vegetative tissues of the Rhodos variety and accelerated the transition to floral phase. Therefore, this reduced the number of days needed to harvest cut roses.

Table 9: Effect of Calcium Chloride, Cytokinin and Abscisic Acid on the number of maturation days taken in flush 1 and 2.

Treatment	Rate of Concentration in mg/L	Maturation Days Flush 1	Maturation Days Flush 2
$CaCl_2$	250	49.78b	52.44b
$CaCl_2$	500	49.89b	52.67b
$CaCl_2$	750	49.67b	52.33b
Cytokinin	150	50.44b	53.00b
Cytokinin	250	50.22b	53.00b
Cytokinin	350	51.72a	53.67a
Abscisic Acid	5	49.56b	52.22b
Abscisic Acid	10	49.56b	52.22b
Abscisic Acid	15	49.06c	50.67c
Control	0	49.78b	52.33b
	LSD	1.47	1.55
	CV	3.13	3.14

Means followed by the same letter(s) along the column are not significantly different at 5% probability level within each flush.

4.1.9 Effect on the Number of Petals

Analysis of treatment effect showed that the application of Calcium chloride, Cytokinin, and Abscisic Acid at all the tested rates had no significant ($p < 0.05$) effect on the number of petals compared to the control (0 mg/L) (Table 10; Appendix 19). These outcomes imply that, in the Rhodos variety, there is no relationship between the administration of calcium chloride, Cytokinin, Abscisic Acid treatments and the number of petals per flower head. Additionally, genetic variables are primarily responsible of the development of petals. Han *et al*. (2018) reported that in contrast to the impacts of the growth regulators, genetic variables regulate the expression of genes involved in petal formation, growth and pattern development. According to Hong *et al*., (2021) Petal specification is primarily determined by the A and B genes based on the ABC paradigm for blooming. The interaction of these genes results in the formation of the floral organs (Vasisth & Sharma, 2022). Additionally, Bowman & Moyroud (2024) reported that the development of sepals and carpels is controlled by one class C gene and a mixture of class A gene, respectively. Together, class B and class A genes determine the development of the petals, and class B and class C genes encourage the growth of the stamens (Bowman & Moyroud, 2024).

Table 10: Effect of Calcium Chloride, Cytokinin and Abscisic Acid on the number of petals produced in flush 1 and 2.

Treatment	Rate of Concentration in mg/L	Number of Petals in Flush 1	Number of Petals in Flush 2
$CaCl_2$	250	35.00a	34.22a
$CaCl_2$	500	35.78a	34.78a
$CaCl_2$	750	34.89a	34.00a
Cytokinin	150	35.00a	34.11a
Cytokinin	250	35.11a	34.30a
Cytokinin	350	35.89a	34.89a
Abscisic Acid	5	35.33a	34.33a
Abscisic Acid	10	35.67a	34.67a
Abscisic Acid	15	35.44a	34.44a
Control	0	35.33a	34.33a
	LSD	1.94	1.87
	CV	5.85	5.78

Means during Flush 1 and 2 were not significantly different at 5% probability level.

4.2 Effect of Calcium Chloride, Cytokinin and Abscisic Acid Application on Postharvest Quality of Rose Cut-Flower.

The effect of Calcium Chloride, Cytokinin and Abscisic acid on postharvest quality of cut roses (*Rosa hybrida* L.) cv. Red Rhodos was assessed at different rates and at different times of application. The analysis of variance showed that chlorophyll content, petal colour, weight loss and vase life were affected by the different rates and different times of application of Calcium Chloride, Cytokinin and Abscisic acid.

4.2.1 Effect on Chlorophyll Content

Calcium Chloride applied at postharvest at the rates of 250, 500, and 750mg/L) significantly increased the chlorophyll content while treatments applied at preharvest at 250, 500, and 750mg/L had no effect on the chlorophyll content at postharvest (Table 11; Appendix 20). However, combined application of the treatments both at preharvest and postharvest at 250, 500, and 750mg/L significantly decreased the chlorophyll content probably due to high concentration in the crop. It could be that the calcium chloride application at postharvest helped in maintating the cell integrity, reduced cellular degradations and therefore preserving the chlorophyll content. Additionally, according to Saftner *et al*. (1998) postharvest application of calcium it has ability to reduce the respiration rate.

Similar observations were reported by El-Beltagi *et al*. (2022) in broccoli. They observed that postharvest application of calcium chloride preserved the overall number of phenols and flavonoids while delaying yellowing and the breakdown of chlorophyll content in broccoli. According to Pareek *et al*. (2017), fresh produce can retain their green color for longer when treated with calcium chloride because it stabilizes cell membranes and inhibits enzymatic breakdown reactions that cause chlorophyll degradation. On a similar study in French beans Kasim & Kasim *et al*. (2015) reported that treatments with high concentrations of calcium chloride had a negative impact on the green color of freshly chopped green beans. Based on the findings of this study, it is very likely that the administration of calcium chloride at high concentration could have contributed to the chlorophyll degradation. Additionally, Tanaka and Tsuji, (1980) observed that high concentrations of Calcium Chloride Degrades Chlorophyll. Treatments with calcium chloride can also have a direct impact on the routes that harvested produce uses to synthesize chlorophyll (Jha *et al*., 2023). Although calcium is necessary for a number of enzymatic processes related to the manufacture of chlorophyll, these processes can be interfered with by high or unbalanced calcium concentrations (Kasim & Kasim, 2015).

The findings also showed that Cytokinin application done both at preharvest and at preharvest and postharvest at 150, 250, and 350 mg/L significantly increased the chlorophyll content of harvested flowers at postharvest. It was observed that 350 mg/L of cytokinin applied at preharvest recorded the highest chlorophyll content of 69.23 and 62.5 at postharvest in flush 1 and 2, respectively (Table 11). Cytokinin application done only at postharvest at 150, 250, and 350 mg/L had no significant effect on the chlorophyll content compared to the control (Appendix 20).

Cytokinin takes at least 3 weeks to accumulate in the plant after application (Dobránszki & Mendler-Drienyovszki, 2014). It is very likely that cytokinin showed no effect in chlorophyll content after the postharvest application because of the slow translocation rate in the crop. However, the positive impact of cytokinin in flowers treated at postharvest was evident because of the already build up levels of cytokinin in the plant at preharvest. Additionally, Cytokinins boost the synthesis of chlorophyll, and extend the time that leaves remain photosynthetic (Sosnowski *et al*., 2023). This explains why preharvest and pre and postharvest increased the chlorophyll content of the leaves at postharvest thereby increasing the freshness of the flowers at vase.

Chlorophyll content at postharvest is very important because it helps to keep the freshness of the cut flowers for more days. When degraded, chlorophyll leads to leaf yellowing (Luo *et al*., 2019) which affects the ornamental value of cut flowers. In research on impacts of cytokinin on crops Sosnowski *et al*. (2023) reported that externally administered cytokinin enhances the amount of chlorophyll in aging leaf tissues by inhibiting the loss of this pigment and postponing the senescence process. By preventing chlorophyll loss and preserving the green hue of the leaves, cytokinin inhibit or retard the aging process of plants (Burr *et al*., 2020). This condition arises because of phytohormones' suppression of the breakdown of green pigment. In a study on the invitro apples Dobránszki & Mendler-Drienyovszki (2014) reported that the effect of cytokinin on the leaves were visible after three weeks of application. This explains why the application of cytokinin at postharvest, during the current study had no significance on the chlorophyll content.

The chlorophyll content in flowers treated with Abscisic Acid only at preharvest, both preharvest and postharvest, and only at postharvest significantly reduced the chlorophyll content. Application of 15 mg/L of Abscisic Acid only at preharvest recorded the lowest concentration of 44.2 and 43.53 in flush 1 and 2, respectively (Table

11). Because of its active role in aging and senescence, it is justifiable that Abscisic acid stimulated the breakdown of chlorophyll therefore reducing its conentartion in the plant. The findings of this study are in agreement with Ferrante *et al*. (2004) who reported that Chlorophyll concentration dropped sharply within a 24-hour period of exogenous application of Abscisic acid treatments. Consistent to this study, Wang *et al*. (2018) observed that long-term Abscisic acid treatment suppressed growth and decreased chlorophyll levels physiologically, but short-term Abscisic acid treatment had no effect on chlorophyll content. It also produced leaf yellowing and hindered chloroplast division. It has been reported by Xie *et al*. (2022) that Abscisic acid accelerates senescence. Chlorophyll degradation happens during senescence because of cellular structures breaking down (Thakur *et al*., 2016). Therefore, it is very likely that in this study that the exogenous application of Abscisic acid hastened leaf senescence, which resulted in the breakdown and reduction of chlorophyll content.

Zhao *et al*. (2016) reported that the first 48 hours following Abscisic acid administration considerably increased the expression of genes related to chloroplast production, dehydration impact responses, and chlorophyll breakdown. According to Gao *et al*. (2016) Abscisic acid controls the expression of genes and the activity of enzymes that regulate the chain reactions leading to the synthesis of chlorophyll, which has an indirect effect on the process. Elevated Abscisic acid levels have the potential to suppress the expression of genes that code for the enzymes needed for the manufacturing of chlorophyll (Yamburenko *et al*., 2015), so reducing the amount of chlorophyll that is produced and accumulates in plant tissues.

Table 11: Effect of Calcium Chloride, Cytokinin and Abscisic Acid on the postharvest chlorophyll content in flush 1 and 2.

Treatment	Application Time	Rate in mg/L	Chlorophyll Content in Flush 1	Chlorophyll Content in Flush 2
$CaCl_2$	Preharvest	250	55.53c	53.10c
$CaCl_2$	Preharvest	500	55.70c	52.83c
$CaCl_2$	Preharvest	750	56.93c	53.43c
$CaCl_2$	Pre and Postharvest	250	52.40d	49.33d
$CaCl_2$	Pre and Postharvest	500	52.70d	49.83d
$CaCl_2$	Pre and Postharvest	750	52.60d	49.40d
$CaCl_2$	Postharvest	250	64.17b	58.23b
$CaCl_2$	Postharvest	500	64.53b	58.73b
$CaCl_2$	Postharvest	750	64.43b	58.60b
Cytokinin	Preharvest	150	63.63b	56.53b
Cytokinin	Preharvest	250	65.77b	58.80b
Cytokinin	Preharvest	350	69.23a	62.50a
Cytokinin	Pre and Postharvest	150	63.93b	57.73b
Cytokinin	Pre and Postharvest	250	66.47b	59.43b
Cytokinin	Pre and Postharvest	350	66.43b	59.43b
Cytokinin	Postharvest	150	55.73c	53.83c
Cytokinin	Postharvest	250	55.83c	53.33c
Cytokinin	Postharvest	350	55.47c	52.83c
Abscisic Acid	Preharvest	5	44.07i	41.27i
Abscisic Acid	Preharvest	10	46.60f	44.77f
Abscisic Acid	Preharvest	15	44.20h	43.53h
Abscisic Acid	Pre and Postharvest	5	46.53f	44.77f
Abscisic Acid	Pre and Postharvest	10	47.53f	44.80f
Abscisic Acid	Pre and Postharvest	15	44.40g	44.37g
Abscisic Acid	Postharvest	5	50.30e	48.07e
Abscisic Acid	Postharvest	10	51.37e	48.13e
Abscisic Acid	Postharvest	15	50.20e	47.63e
Control	No Application	0	55.33c	51.77c
		LSD	2.92	3.54
		CV	3.19	4.14

Means followed by the same letter(s) along the column for flush 1 and 2 are not significantly different at 5% probability level.

4.2.2 Effect on the Petal Color

Calcium chloride treatment application only at preharvest, both at preharvest and postharvest, and only at postharvest (250, 500, and 750 mg/L) increased the Lightness with 750mg/L applied at preharvest and postharvest being the highest at 28.93 and 30.01 in flush 1 and 2, respectively (Table 12; Appendix 21). While 250 mg/L of Calcium chloride applied at postharvest increased the lightness in the petal colors, the significance was lesser than other concentrations of calcium chloride. This signified that at low concentrations calcium chloride application at postharvest have less effective in influencing the lightness in petal colour. Irrespective of the different concentration and different times of application, Calcium chloride significantly increased the redness of the petal color (Table 12; Appendix 22). In both flushess, the yellowness of the petal color responded significantly to Calcium chloride (250, 500, and 750 mg/L) applied only at preharvest, both at preharvest and postharvest, and only at postharvest (Table 12; Appendix 23).

Exogenous application of Calcium increases the level of calmodulin in plants. Calmodulin influences the transportation of photosynthetic products and the resistance mechanisms to various biotic and abiotic stresses. It is likely that application of various concentrations of Calcium Chloride at different times, increased the level of calmodulin in Rhodos variety. This helped in transportation of photosynthetic products necessary for petal colour development and retaining of the integrity of the cells in the reproductive structures of the plant even after separation from the mother plant. Therefore, the flowers produced their pigments more effectively and had brighter, richer hues.

According to Thor (2019) the rigidity and stability of the cell wall are dependent on calcium. Flowers can wilt and lose some of their brightness due to cell wall breakdown caused by insufficient calcium (Elshawa *et al*., 2023). It has been reported that application of calcium chloride supports the maintenance of ideal cell turgor pressure, which keeps fresh produce turgid and erect and enhances their entire brightness and aesthetic appeal (Martins *et al*., 2020). According to Guo *et al*. (2021) application of calcium chloride enhanced calcium contents in the plant, improved the physiological functions of calmodulin (Wang *et al*., 2023), and heightened the expression of genes responsible for anthocyanin biosynthesis (Michailidis *et al*., 2017) in the petals. Consequently, sufficient amounts of calcium encourage the synthesis and stability of pigments like carotenoids, anthocyanins, and flavonoids (Zhu *et al*., 2023) that give flowers their color. Zhu *et al*. (2019) reported that Calcium facilitates the plant's transportation of photosynthetic products, supplying the building blocks needed for the synthesis of anthocyanins.

Cytokinin application at 150, 250, and 350mg/L during preharvest, and preharvest and postharvest significantly increased the lightness of the petals. However, the significance on the lightness of Petal color varied with the concentration of cytokinin and the time of application. Cytokinin treatment applied only in the postharvest had no significant effect on lightness of petal color compared to the control (0 mg/L) [Table 12; Appendix 21]. Irrespective of the different concentration and different times of application, cytokinin significantly increased the redness of the petal color. Nevertheless, 350mg/L cytokinin applied both in the field and in postharvest had the highest redness of 57.83 and 62.44 both in flush 1 and flush 2 (Table 12; Appendix 22). In both flushes, the yellowness of the petal color responded significantly to Cytokinin (150, 250, and 350 mg/L) applied only at preharvest, both at preharvest and postharvest, and only at postharvest. The outcomes showed that 350mg/L of cytokinin applied at preharvest recorded the highest yellowness of 17.62 and 18.89 in flush 1 and flush 2 respectively (Table 12; Appendix 23). Cytokinin has an ability to delay senescence in plants (Kaur & Jhanji, 2023). It is possible that the Cytokinin delayed the breakdown of Carotenoids and anthocyanins pigments which are responsible for the red colour in plants. This therefore, helped the plants look more radiant and retained their color for long. Additionally, Shibuya and Ichimura (2016) observed that application of cytokinin to cut flowers prolonged the time that they looked fresh and brilliant, which increased their overall radiance by retarding the natural aging process.

Abscisic acid treatment applied at preharvest, and at preharvest and postharvest decreased the lightness of the petal color with 15 mg/L of Abscisic Acid applied at preharvest and postharvest having the lowest lightness of 6.49 and 6.98 in flush 1 and 2, respectively (Table 12; Appendix 21). However, application of Abscisic acid only at postharvest had no effect on the lightness of the petals. On the other hand, Abscisic acid significantly decreased the redness of the petal with 15 mg/L of Abscisic Acid at preharvest having the lowest redness of 31.15 and 34.47 in flush 1 and flush 2, respectively (Table 12; Appendix 22). Abscisic Acid application had no significant effect on the yellowness of the petal color in all the different concentrations and the different times of application (Table 12; Appendix 23). Abscisic Acid is a stress hormone that increases senescence in plants. The increase in senescence reduces the concentration of anthocyanins and carotenoids therefore making the flowers reduce the brightness and radiance of the petals of the Rhodos variety.

Xia *et al*. (2020) reported that the yellow-to-red hue of flowers is largely attributed to the presence of carotenoids. While investigating into the effects of Abscisic acid in Camellia sinensis, Baldermann *et al*. (2013) observed that the carotenoid levels decreased with increasing Abscisic acid concentration in the plant. Moreover, Liu *et al.* (2020) reported that Application of Abscisic acid altered the carotenoid concentration of flower petals, hence affecting their coloring. Ahmad and Tahir. (2016) also reported that application of Abscisic acid at certain developmental phases or in high doses hastened the senescence process in flowers, causing wilting, fading, and decreased brightness.

Table 12: Effect of Calcium Chloride, Cytokinin and Abscisic Acid on petal color in flush 1 and 2.

Treatment	Application Time	Rate in mg/L	Lightness (L*)		Redness (a*)		Yellowness (b*)	
			Flush 1	Flush 2	Flush 1	Flush 2	Flush 1	Flush 2
$CaCl_2$	Preharvest	250	27.44a	29.44a	45.84e	49.24e	14.17b	15.22b
$CaCl_2$	Preharvest	500	27.29a	29.32a	47.39d	50.91c	15.42b	16.54b
$CaCl_2$	Preharvest	750	27.41a	29.42a	49.70d	53.41d	16.24b	17.45b
$CaCl_2$	Pre/Postharvest	250	27.29a	29.30a	48.33d	51.90c	13.93c	14.96c
$CaCl_2$	Pre/Postharvest	500	27.70a	29.87a	49.68d	53.38d	15.58b	16.75b
$CaCl_2$	Pre/Postharvest	750	28.93a	31.01a	55.13b	59.52b	15.97b	17.18b
$CaCl_2$	Postharvest	250	26.25d	28.14d	47.67d	51.22c	16.46b	17.68b
$CaCl_2$	Postharvest	500	28.49a	30.76a	49.89c	53.48d	15.43b	16.57b
$CaCl_2$	Postharvest	750	28.12a	30.14a	52.09c	55.84d	14.47b	15.52b
Cytokinin	Preharvest	150	26.53c	28.49c	46.13e	49.55d	16.50b	17.70b
Cytokinin	Preharvest	250	26.74b	28.73b	46.66e	50.13c	16.48b	17.69b
Cytokinin	Preharvest	350	27.13a	29.15a	49.63d	53.30d	17.62a	18.90a
Cytokinin	Pre/Postharvest	150	27.23a	29.25a	52.89b	56.81c	15.14b	16.26b
Cytokinin	Pre/Postharvest	250	27.13a	29.14a	53.76b	57.79b	14.16b	15.23b
Cytokinin	Pre/Postharvest	350	27.67a	29.47a	57.83a	62.44a	15.30b	16.52b
Cytokinin	Postharvest	150	22.80e	22.61e	48.53d	52.14c	15.48b	16.62b
Cytokinin	Postharvest	250	22.65e	22.53e	48.24d	51.73c	14.36b	15.43b
Cytokinin	Postharvest	350	21.56e	22.39e	47.92d	51.37c	15.99b	17.25b
ABA	Preharvest	5	8.54f	9.18f	33.73h	36.16h	7.812d	7.24d
ABA	Preharvest	10	8.95f	9.62f	37.27g	40.05g	7.15d	7.69d
ABA	Preharvest	15	7.12g	7.65g	32.26i	34.66i	8.84d	9.49d
ABA	Pre/Postharvest	5	6.64h	7.14g	36.76g	39.49g	8.31d	9.00d
ABA	Pre/Postharvest	10	6.88h	7.43g	32.15i	34.47j	8.46d	9.09d
ABA	Pre/Postharvest	15	6.49i	6.98g	37.41g	40.18g	8.46d	9.13d
ABA	Postharvest	5	20.52e	22.05e	36.90g	39.67g	6.78d	7.28d
ABA	Postharvest	10	19.57e	20.65e	36.24g	39.13g	7.50d	7.75d
ABA	Postharvest	15	20.53e	22.15e	34.74h	37.24h	6.87d	8.83d
Control	No application	0	19.26e	19.82e	40.65f	45.43f	6.97d	7.47d
		LSD	1.80	1.95	2.36	2.56	2.35	2.52
		CV	5.05	5.08	3.23	3.25	11.48	11.46

Means followed by the same letter(s) along the column for flush 1 and 2 are not significantly different at 5% probability level.

4.2.3 Effect on the Weight Loss

Calcium chloride reduced the % weight loss in Rhodos variety in highest concentrations of treatment (750 mg/L applied both at preharvest and postharvest, and 750 mg/L applied at postharvest) [Table 13; Appendix 24]. However, the other treatment applications applied at preharvest (250, 500, and 750 mg/L), at the pre and postharvest (250, and 500 mg/L) and at postharvest (250, and 500 mg/L) had no significant effect the weight loss [Table 13; Appendix 22]. Calcium helps to maintain the integrity of the cell. Therefore, application of Calcium Chloride enhanced the integrity of the cells in Rhodos variety, reducing the rate of degeneration which in turn reduced the weight loss. However, based on these findings it is evident that Calcium translocation is slow and can only reduce weight loss in roses upon build up within the plant.

These findings are in agreement with Thakur *et al*. (2019) who did a study on effects of calcium chloride on fresh fruits. They reported that over the course of 20 days of storage, fresh cut guava, muskmelon, and papaya all saw a significant decrease in weight loss or absence of weight loss when exposed to calcium Chloride. The current study was also consistent with Chepngeno *et al*. (2016) who reported the least amount of weight lost by tomatoes and African eggplant hydro cooled in water containing calcium chloride. These findings concur with those of Lou *et al.* (2020), who discovered that cut flower of Perpetual Carnation treated with calcium chloride at postharvest had a reduced weight loss.

According to Thakur *et al*. (2019) calcium has an ability to maintain the functional and structural integrity of membrane systems by strengthening the cell wall and protecting them from deterioration. Ramezanian *et al.* (2018) reported that calcium reduced the transpiration rate by preventing loss of water from the produce's surfaces, which maintained its moisture content. As a result, the reduced transpiration reduced the percentage weight loss. In fruits and vegetables, Gao *et al*. (2020) observed that Calcium chloride prolonged shelf life by inhibiting the activity of the enzymes that break down cells, which further prevented weight loss from deterioration. Therefore, this serves as the major underlying reason for the reduction in weight or the absence of weight loss in cut flowers treated with calcium chloride.

Analysis of treatment showed that Cytokinin was only able to significantly reduce weight loss when applied in the highest concentration (350 mg/L) at preharvest (Table 13). On the other hand, the rest of the cytokinin treatment had no significant effect on the weight loss of cut Rhodos variety (Table 13; Appendix 24). Cytokinin helps to delay senescence in plants. It could be that by delaying senescence in Flowers cytokinin helped the plant retain their structural strength therefore reducing the rate of weight loss. Consistent to the current outcomes, Ainalidou *et al.* (2016) found that preharvest treatment of cytokinin effectively lowered weight reduction in kiwifruit by 33% and prolonged storage by two months when compared to the control; these observations demonstrated the effectiveness of Cytokinins in preventing structural deterioration in fruits. Moreover, Marzouk & Kassem (2011) reported that cytokinin treatment prior to harvest greatly decreased the reduction in weight in Thompson seedless grapes. In research on the effects of growth regulators on the quality of cucumber Qian *et al*., (2018) observed that application of cytokinin significantly reduced the weight loss of cucumber fruit at postharvest.

According to Li *et al*. (2023), the capacity of Cytokinin to deter senescence in fresh produce becomes especially useful during processing following harvest. By delaying the natural senescence process, plants treated with cytokinin have the ability to retain their water content and their structural strength for extended periods of time (Danilova *et al*., 2020). Gao *et al*. (2016) observed that early senescence causes weight loss, but delay in senescence slows it down, keeping the peach fruit fresh and commercially viable throughout storage and transit. Moreover, cytokinin ability to reduce physiological responses to stress in plants (Cortleven *et al*., 2019) strengthens its role in preventing weight loss. This keeps the cut flowers hydrated and healthy, reducing weight loss and increasing market value all the way through the postharvest process.

Abscisic acid (5, 10, and 15 mg/L) significantly reduced % weight loss when applied at preharvest, preharvest and at postharvest, and at the highest concentration of postharvest application (15 mg/L) [Table 13; Appendix 24]. It is possible that Abscisic acid significantly reduced the weight loss in Rhodos variety because of its ability to induce stomata closure to reduce moisture loss at postharvest. Liu *et al*. (2015) observed that exogenous Abscisic acid administration, considerably reduced postharvest lettuce's weight loss. According to Dar *et al*., (2017), Abscisic acid is considered as a stress hormone and that it helps plants cope with abiotic stress. Through stomata closure, it reduces the moisture loss in fresh produce (Negin *et al*., 2019). Huang *et al*. (2021), reported that significant water loss during storage resulted in both the loss of marketable weight and clear quality problems like shriveling in kiwi fruit. According to Hung *et al*. (2011), transpiration is thought to be the primary factor causing leafy vegetable quality degradation and weight loss. This is because vegetable water loss after harvesting is a serious issue since it causes withering of the leaves and fresh weight loss, which affects the produce' appearance and shelf life (Mahajan *et al*., 2008).

Table 13: Effect of Calcium Chloride, Cytokinin and Abscisic Acid on weight loss in flush 1 and 2.

Treatment	Application Time	Rate in mg/L	Weight Loss in Flush 1	Weight Loss in Flush 2
$CaCl_2$	Preharvest	250	18.03a	16.31a
$CaCl_2$	Preharvest	500	17.48a	15.93a
$CaCl_2$	Preharvest	750	16.84a	15.24a
$CaCl_2$	Pre and Postharvest	250	17.15a	15.33a
$CaCl_2$	Pre and Postharvest	500	17.21a	15.44a
$CaCl_2$	Pre and Postharvest	750	16.65b	15.14b
$CaCl_2$	Pre and Postharvest	250	17.46a	15.76a
$CaCl_2$	Pre and Postharvest	500	17.39a	15.75a
$CaCl_2$	Pre and Postharvest	750	15.94b	14.31b
Cytokinin	Preharvest	150	17.00a	15.49a
Cytokinin	Preharvest	250	17.13a	15.25a
Cytokinin	Preharvest	350	15.91b	14.24b
Cytokinin	Pre and Postharvest	150	17.89a	16.20a
Cytokinin	Pre and Postharvest	250	17.64a	15.20a
Cytokinin	Pre and Postharvest	350	17.33a	15.69a
Cytokinin	Postharvest	150	16.83a	16.16a
Cytokinin	Postharvest	250	18.33a	16.86a
Cytokinin	Postharvest	350	18.42a	16.91a
Abscisic Acid	Preharvest	5	10.04c	9.82c
Abscisic Acid	Preharvest	10	8.41c	8.42c
Abscisic Acid	Preharvest	15	9.53c	8.98c
Abscisic Acid	Pre and Postharvest	5	9.22c	8.76c
Abscisic Acid	Pre and Postharvest	10	8.94c	8.56c
Abscisic Acid	Pre and Postharvest	15	9.54c	9.35c
Abscisic Acid	Postharvest	5	18.13a	16.70a
Abscisic Acid	Postharvest	10	18.08a	16.31a
Abscisic Acid	Postharvest	15	16.64b	14.96b
Control	No Application	0	17.62a	15.95a
		LSD	1.676	1.74
		CV	6.61	7.46

Means followed by the same letter(s) along the column for flush 1 and 2 are not significantly different at 5% probability level.

4.2.4 Effect on Vase life of Rose Cut-Flower

Analysis of treatment revealed that calcium chloride significantly ($p < 0.05$) increased the number of days the flowers spent in vase [Table 14; Appendix 25]. High concentration of calcium chloride was noted to be more effective than lower concentrations. This explains why 250 mg/L of calcium chloride at postharvest (Lowest concentration applied at postharvest) had no significant effect. At preharvest application 750 mg/L of calcium chloride demonstrated the highest influence in vase life where it had an average of 14.67 and 12.67 in flush 1 and 2 (Table 14). At preharvest and postharvest application, A6 had the most significant effect on vase life with 14.67 and 13 days in flush 1 and 2 (Table 14). It is conceivable that calcium accumulated in plants promoting the bonding of pectin polymers in the cell wall structure that boosts mechanical strength and delays cell disintegration.

The observations of this study on the effect of calcium chloride on vase life on cut roses were consistent to the study by Abdolmaleki *et al*. (2015). They reported that the Vase life of "Dolce Vita" roses was increased by varying calcium chloride concentrations with the highest concentration of calcium chloride and salicylic acid having the longest vase life. Similarly, Akintoye *et al.* (2016) reported that calcium chloride significantly improved the Vase life of Heliconia. They reported no wilted flowers in calcium chloride treatment after 9 days in vase which was contrary to other treatment. Park & Kim. (2022) reported that calcium spray effectively decreased bending and strengthened the scape by more than 10%, extending the gerbera flowers vase life. Additionally, Ehsanimehr *et al.* (2024), reported that application of calcium chloride prior to harvesting prolonged the vase life and antioxidant capability of cut roses while also delaying senescence.

Prior research has demonstrated the advantageous impact of exogenous calcium application in mitigating environmental variations in plants. The physiological processes responsible for these beneficial effects have been identified as osmotic adjustment and enhanced antioxidant responses which in turn enhance the life of a plant (Henry *et al*., 2021). According to Divya Sharma *et al.* (2017), Calcium controls the architecture, signalling pathways, and functionality of membranes by its capacity of bonding to the phospholipid bilayer. As a result, this engagement makes it easier for plants to mitigate harmful variations in the environmental conditions by strengthening and improving the structural stability and integrity of their membranes. Additionally, earlier observations by Timalsina *et al*. (2023) indicated that calcium builds up in plants promoting the bonding of pectin polymers in the centre of lamella to produce a cell wall structure that boosts mechanical strength and delays stem bending. This explains why calcium chloride application in the current study increased the vase life of Rose cut-flower Variety Rhodos.

Analysis of treatment revealed that Cytokinin significantly ($p < 0.05$) increased the number of days the flowers spent in vase [Table 14; Appendix 25]. Application of cytokinin (150, 250, and 350 mg/L) at preharvest, and at preharvest and postharvest significantly increased the vase life of Rhodos variety. The highest significance was noted in the highest concentrations (250 and 350 mg/L) because of the gradual accumulation of cytokinin within the plant (Appendix 25). However, application of cytokinin at postharvest had no significance on the vase life days. It could be that cytokinin was able to increase vase life because of its ability to counter the effect of ethylene. Additionally, it is likely that cytokinin slowed down the aging process of plant components by delaying the breakdown of their protein and chlorophyll because of its ability to delay senescence. In a study on the response of cut flowers to cytokinin and 1-metylcylcopropene Mirzakhani *et al.* (2020) reported similar outcomes. They observed that higher concentration of cytokinin reduced the amount of ethylene present in cut flowers, which in turn extended the vase life. Similarly, Kaur *et al*. (2017) reported that cytokinin inhibited petal senescence by blocking the production of ethylene or by lessening the cells' sensitivity to it and therefore increasing the vase life of cut flowers. Additionally, Shaya *et al*. (2019) reported that degeneration was slowed down when the concentration of cytokinins was elevated at the abscission layer in the fruitlets of persimmon.

Analysis of treatment revealed that abscisic acid significantly ($p < 0.05$) reduced the vase life of Rhodos variety. As a result, 15 mg/L of Abscisic Acid applied at preharvest and postharvest had the least number of days in vase with 10.33 and 9.67 days in flush 1 and 2 (Table 14). At lower concentration, Abscisic acid (5 and 10 mg/L at preharvest, preharvest and postharvest, and postharvest; and 15 mg/L at postharvest) had no significant effect on the vase life of Rhodos variety [Table 14; Appendix 25]. Abscisic Acid is a stress hormorne that induces aging. It is likely that after the separation from the mother plant, Abscisic Acid stimulated the aging process in the flowers at postharvest therefore reducing the vaselife. It is also possible that Abscisic acid enhanced closure of stomata as a response to abiotic stress, affecting water uptake and gaseous exchange, thereby causing early senescence. Similar to the outcomes of this study on the effects of Abscisic acid on vase life, Costa *et al.* (2016) reported that abscisic acid decreased the stem longevity of cut gladiolus flowers by increasing the senescence rate. The findings were also consistent with Zhong and Ciafre, (2011), who reported that Abscisic Acid treatment hastens the senescence of lily petals, indicating that Abscisic Acid plays a direct function in the initial stages of deterioration in petals independent of endogenous ethylene levels. Geng *et al*. (2015) also agreed with the current findings and reported accelerated opening of cut buds and reduction in the time that flowers and leaves remained in the vase when higher concentrations of abscisic acid were applied in in cut lilies.

According to Shibuya & Ichimura (2016) Abscisic acid speeds up senescence and the decomposition of cut flowers. Upon harvesting, flowers are deprived of their nutrition source and exposed to a number of stressors, including mechanical harm, hormone imbalances, and water shortage (Rabiza-Świder *et al*., 2020). As a result, plants produce more Abscisic acid in reaction to these stresses which is harmful to cut flowers. Aalifar *et al*. (2020) reported that Abscisic acid shortens vase life by enhancing stomata closure. Even though stomata closure helps save water in plants, it reduces the amount of water absorbed by cut flowers (Kitamura *et al*., 2017). Limitations in water intake hasten the process of dehydration, resulting in withering and early senescence (Kitamura *et al*., 2017). According to Imadi et al. (2016), dehydration and early senescence causes tissue disintegration and a loss of turgidity. This hastens the decomposition of cellular components such as structural proteins and chlorophyll, which gives cut flowers their vivid color. Therefore, based on the findings of this study, in case of high Abscisic acid levels, cut roses age more quickly and show symptoms including petal abscission, wilting, and yellowing.

Table 14: Effect of Calcium Chloride, Cytokinin and Abscisic Acid on vase life in flush 1 and 2.

Treatment	Application Time	Rate in mg/L	Vase life in Flush 1	Vase life in Flush 2
$CaCl_2$	Preharvest	250	12.33b	11. 67b
$CaCl_2$	Preharvest	500	12.67b	11.67b
$CaCl_2$	Preharvest	750	14.67a	12.67a
$CaCl_2$	Pre and Postharvest	250	13.00b	11.67b
$CaCl_2$	Pre and Postharvest	500	13.00b	12.00b
$CaCl_2$	Pre and Postharvest	750	14.67a	13.00a
$CaCl_2$	Postharvest	250	11.67c	10.67c
$CaCl_2$	Postharvest	500	12.67b	11.67b
$CaCl_2$	Postharvest	750	12.67b	11.67b
Cytokinin	Preharvest	150	12.33b	11.67b
Cytokinin	Preharvest	250	13.67a	12.00b
Cytokinin	Preharvest	350	13.67a	12.67a
Cytokinin	Pre and Postharvest	150	13.00b	11.67b
Cytokinin	Pre and Postharvest	250	13.33b	12.00b
Cytokinin	Pre and Postharvest	350	14.33a	12.67a
Cytokinin	Postharvest	150	11.33c	10.33c
Cytokinin	Postharvest	250	12.00c	10.67c
Cytokinin	Postharvest	350	12.00c	11.00c
Abscisic Acid	Preharvest	5	11.33c	10.33c
Abscisic Acid	Preharvest	10	11.33c	10.33c
Abscisic Acid	Preharvest	15	10.67d	9.67d
Abscisic Acid	Pre and Postharvest	5	11.00c	10.00c
Abscisic Acid	Pre and Postharvest	10	11.00c	10.33c
Abscisic Acid	Pre and Postharvest	15	10.33e	9.67e
Abscisic Acid	Postharvest	5	11.67c	10.33c
Abscisic Acid	Postharvest	10	12.00c	10.67c
Abscisic Acid	Postharvest	15	12.00c	11.00c
Control	No Application	0	11.607c	10.330c
		LSD	1.26	1.13
		CV	6.27	6.16

Means followed by the same letter(s) along the column for flush 1 and 2 are not significantly different at 5% probability level.

CHAPTER FIVE

SUMMARY, CONCLUSION AND RECOMMENDATIONS

5.1 Summary of Findings

The study aimed at determining the effects of Calcium chloride, Cytokinin, and Abscisic Acid on cut rose's cv. Rhodos. The specific objective that guided the study included; to determine the effect of calcium chloride, cytokinin and abscisic acid application on the growth of rose flower and to determine the effect of calcium chloride, cytokinin and abscisic acid application on the postharvest quality of rose flower. The parameters of growth considered included; number of shoots, stem length, stem diameter, flower bud size, leaf area, chlorophyll content, number of suckers, physiological disorders, flush days, and number of petals. The postharvest parameters taken into account included; chlorophyll content, petal color, weight loss and vase life. In the research, Randomized Complete Block Design (RCBD) was used to demonstrate the level of variation of growth parameters that were influenced by Calcium chloride, Cytokinin, and Abscisic Acid. Complete Randomised Design (CRD) was used demonstrate the level of variation of postharvest parameters.

The outcomes showed that application of cytokinin increased the number of shoots produced per plant, stem length, leaf area, and number of suckers compared to the control, Calcium Chloride and Abscisic Acid. On the other hand, Abscisic acid reduced the stem length, and leaf area. Cytokinin applied at 250 and 350 mg/L increased chlorophyll content 3 weeks after first application of treatments. After the second application of treatments (at maroon stage) all the concentrations of cytokinin and Calcium Chloride (500 and 750 mg/L) increased chlorophyll content. After the second application of the treatments (at maroon stage) 10 and 15 mg/L of Abscisic Acid significantly reduced the chlorophyll content.

It was observed that the highest concentration of Calcium chloride (750 mg/L) reduced the incidences of bent peduncles in Rhodos variety. When applied at high concentration of 15mg/L, Abscisic Acid significantly reduced the flush days of Rhodos variety with 0.7 days in flush 1 and 1.66 in flush 2. Highest rates of cytokinin (350 mg/L) extended the flush days of Rhodos variety with 1.9 days in Flush 1 and 1.3 days in flush 2.

Application of Cytokinin, Abscisic acid, and Calcium chloride on Rhodos variety at preharvest had no effect on stem diameter, flower bud size, and number of petals.

Analysis of variance on postharvest parameters revealed that, Calcium Chloride applied at postharvest significantly increased the chlorophyll content while treatments applied at preharvest had no effect on the chlorophyll content at postharvest. However, treatments applied both at preharvest and postharvest significantly decreased the chlorophyll content due to high concentration in the crop. Cytokinin application done both at preharvest, and at preharvest and postharvest significantly increased the chlorophyll content of harvested flowers at postharvest. 350 mg/L of cytokinin applied at preharvest recorded the highest chlorophyll content of 69.23 and 62.5 at postharvest in flush 1 and 2 respectively. Cytokinin application done only at postharvest had no significant effect on the chlorophyll content compared to the control. The chlorophyll content in flowers treated with Abscisic Acid at preharvest, at preharvest and postharvest, and at postharvest significantly reduced. 15 mg/L Abscisic Acid applied at preharvest recorded the lowest concentration of 44.2 and 43.53 in flush 1 and 2, respectively.

Calcium chloride treatment application at preharvest, at preharvest and postharvest, and at postharvest increased the Lightness with 750mg/L applied at preharvest and postharvest being the highest at 28.93 and 30.01 in flush 1 and 2, respectively. Cytokinin application significantly increased the lightness of the petals on treatment in the preharvest, and preharvest and postharvest. However, the significance on the lightness of Petal color varied with the concentration of cytokinin and the time of application. Cytokinin treatment applied only in the postharvest had no significant effect on lightness of petal color compared to the control. Abscisic acid treatment applied at preharvest, and at preharvest and postharvest decreased the lightness of the petal color with 15 mg/L of Abscisic Acid applied at preharvest and postharvest having the lowest lightness of 6.49 and 6.98 in flush 1 and 2, respectively. Nevertheless, application of Abscisic acid only at postharvest had no effect on the lightness of the petals.

Irrespective of the different concentration and different times of application, Calcium chloride and cytokinin significantly increased the redness of the petal color. Where 350mg/L cytokinin was applied both in the postharvest and in postharvest had the highest redness of 57.83 and 62.44 both in flush 1 and flush 2. Abscisic acid significantly decreased the redness of the petal with 15 mg/L of Abscisic Acid at preharvest having the lowest redness of 31.15 and 34.47 in flush 1 and flush 2. In both flushs, the yellowness of the petal color responded significantly to Calcium chloride and Cytokinin applied at preharvest, at preharvest and postharvest, and at postharvest. The observations showed that 350 mg/L applied at preharvest recorded the highest yellowness of 17.62 and 18.89 in flush 1 and flush 2 respectively. Abscisic Acid application had no significant effect on the yellowness of the petal color in all the different concentrations and the different times of application.

Analysis of treatment showed that calcium chloride reduced the % weight loss in Rhodos variety in highest concentrations of treatment (750 mg/L applied both at preharvest and postharvest, and at postharvest). Cytokinin was only able to significantly reduce weight loss when applied in the highest concentration (350 mg/L) at preharvest. Abscisic acid (5, 10, and 15 mg/L) significantly reduced % weight loss when applied at preharvest, preharvest and at postharvest, and at the highest concentration of postharvest application (15 mg/L).

Analysis of treatment revealed that calcium chloride and Cytokinin significantly ($p < 0.05$) increased the number of days the flowers spent in vase while abscisic acid significantly ($p < 0.05$) reduced the vase life. High concentration of calcium chloride was noted to be more effective than lower concentrations. At preharvest application 750mg/L of calcium chloride demonstrated the highest influence in vase life where it had an average of 14.67 and 12.67 in flush 1 and 2. At preharvest and postharvest application, A6 had the most significant effect on vase life with 14.67 and 13 days in flush 1 and 2. In cytokinin application, the highest significance was noted in the highest concentrations (250 and 350mg/L) because of the gradual accumulation of cytokinin within the plant. However, application of cytokinin at postharvest had no significance on the vase life days. Treatment analysis revealed that high concentration of Abscisic Acid reduced the vase life days of Rhodos variety. As a result, 15 mg/L of Abscisic

Acid preharvest and postharvest applied at had the least number of days in vase with 10.33 and 9.67 days in flush 1 and 2. At lower concentration, Abscisic acid (5 and 10 mg/L at preharvest, preharvest and postharvest, and postharvest; and 15 mg/L at postharvest) had no significant effect on the vase life of Rhodos variety.

5.2 Conclusion

From this study, it can be concluded that application of cytokinin increased the number of shoots produced per plant, stem length, leaf area, and number of suckers in cut flowers. On the other hand, Abscisic acid reduced the stem length, and leaf area. At high rates Cytokinin increased the levels of chlorophyll content after first application while lower rates increased chlorophyll content after the second application. High rates of Calcium chloride also increased the chlorophyll content after second application of treatment. At high rates of application, Abscisic Acid, significantly reduced the chlorophyll content and especially after second treatment application. It was also observed that at high rates of application, Calcium chloride reduced incidences of bent peduncles. Additionally, high rate of Cytokinin increased flush days while high rate of Abscisic acid reduced the flush days of cut roses. However, application of Cytokinin, Abscisic acid, and Calcium chloride on Rhodos variety at preharvest had no effect on stem diameter, flower bud size, and number of petals.

Based on the current study, it can be concluded that Calcium Chloride applied at postharvest significantly increased the chlorophyll content while treatments applied at preharvest had no effect on the chlorophyll content at postharvest. Treatments applied both at preharvest and postharvest significantly decreased the chlorophyll content. Cytokinin application at preharvest, and at preharvest and postharvest significantly increased the chlorophyll content of harvested flowers at postharvest. Cytokinin application at postharvest had no significant effect on the chlorophyll content. The chlorophyll content in flowers treated with Abscisic Acid significantly reduced at postharvest. Moreover, Calcium chloride application increased the Lightness at postharvest. Cytokinin when applied at preharvest, and preharvest and postharvest significantly increased the lightness of the petals while Abscisic acid decreased. Also, different rates and different times of application, Calcium chloride and cytokinin significantly increased the redness and yellowness of the petal color. On the other hand, Abscisic acid significantly decreased the redness of the petal. Calcium chloride (applied at both preharvest and postharvest) and cytokinin (applied at preharvest) reduced the % weight loss in Rhodos variety as seen in the outcomes. Further, Calcium chloride and Cytokinin increased the number of days the flowers spent in vase while abscisic acid reduced the vase life.

5.3 Recommendations

Based on the findings of the current study, the following recommendation was made.

i. To achieve optimal growth and quality in cut flowers, growers should adopt the application of Cytokinin (250 and 350 mg/L) and Calcium Chloride (500 and 750 mg/L) during growth while avoiding use of Abscisic Acid. However, high concentration of cytokinin may increase the number of suckers present on individual stems.

ii. To achieve optimal postharvest quality of cut roses, growers can apply calcium chloride (500 and 750 mg/L) and cytokinin (250 and 350 mg/L) at preharvest and/or at postharvest.

iii. To increase vase life in cut roses, growers can apply calcium chloride (750 mg/L) and Cytokinin (250 and 350 mg/L) and avoid application of abscisic acid because it reduces the vase life.

5.4 Suggestions for Further Research

i. The propagation was done during the cold months of the year, June and July. However, a 5% success rate was observed when propagation was done in Murang'a which had cold weather and 90% success rate was observed when the propagation was repeated in Naivasha which had warm weather. It is therefore recommended that further research be conducted to investigate the effect of cold and warm temperatures on bud dormancy during propagation of rose flowers. This can help improve the propagation success rate, reduce the time and cost spent during propagation. Therefore, making the propagation process efficient.

ii. The results showed that Cytokinin had better growth performance than Calcium chloride and Abscisic Acid but this cannot be fully concluded since soilless media, one variety, one type of rose, and a control was involved during the study. As a result, further study is recommended on soil media and other types of roses for a more extended duration of two years, as roses are perennial plants and longer data collection periods yield superior results.

iii. The research was conducted in single geographical location, Ruiru, Kenya. Further study can be done in various geographical regions to collect data on roses cultivated throughout Kenya, particularly in the areas that produce flowers.

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APPENDICES

Appendix 1: Research Site Map - Kiambu County.

Source: Maptive Software

Appendix 2: Clearance Form from Chuka University Ethics Review Committee

CHUKA UNIVERSITY

Knowledge is Wealth *(Sapientia divitia est)* Akili ni Mali

CHUKA UNIVERSITY INSTITUTIONAL ETHICS REVIEW COMMITTEE

Telephones: 020-2310512/18 **P. O. Box 109-60400, Chuka**
Direct Line: 0772894438 **Email: info@chuka.ac.ke,** **Website: www.chuka.ac.ke**

16th May, 2023

REF: CUIERC/ NACOSTI/386
TO: **Regina Wakomi Mburu**

RE: Effect of Calcium Chloride, Cytokinin and Abcisic Acid on Growth and Postharvest Quality of Rose Flower

This is to inform you that *Chuka University IERC* has reviewed and approved your above research proposal. Your application approval number is *NACOSTI/NBC/AC-0812.* The approval period is 16th May, 2023 – 16th May, 2024.

This approval is subject to compliance with the following requirements;

i. Only approved documents including (informed consents, study instruments, MTA) will be used
ii. All changes including (amendments, deviations, and violations) are submitted for review and approval by *Chuka University IERC.*
iii. Death and life threatening problems and serious adverse events or unexpected adverse events whether related or unrelated to the study must be reported to *Chuka University IERC* within 72 hours of notification
iv. Any changes, anticipated or otherwise that may increase the risks or affected safety or welfare of study participants and others or affect the integrity of the research must be reported to *Chuka University IERC* within 72 hours
v. Clearance for export of biological specimens must be obtained from relevant institutions.
vi. Submission of a request for renewal of approval at least 60 days prior to expiry of the approval period. Attach a comprehensive progress report to support the renewal.
vii. Submission of an executive summary report within 90 days upon completion of the study to *Chuka University IERC.*

Prior to commencing your study, you will be expected to obtain a research license from National Commission for Science, Technology and Innovation (NACOSTI) https://oris.nacosti.go.ke and also obtain other clearances needed.

Yours sincerely

Dr. Benjamin Kanga
SECRETARY

Chuka University is....... ...Inspiring Environmental Sustainability for Better Life

Appendix 3: National Commission of Science, Technology and Innovation (NACOSTI) Permit

REPUBLIC OF KENYA

NATIONAL COMMISSION FOR SCIENCE,TECHNOLOGY & INNOVATION

Ref No: 135369

Date of Issue: 17/July/2023

RESEARCH LICENSE

This is to Certify that Miss.. Reginah Wakomi Mburu of Chuka University, has been licensed to conduct research as per the provision of the Science, Technology and Innovation Act, 2013 (Rev.2014) in Kiambu on the topic: EFFECT OF CALCIUM CHLORIDE, CYTOKININ, AND ABSCISIC ACID ON GROWTH AND POSTHARVEST QUALITY OF ROSE FLOWER for the period ending : 17/July/2024.

License No: **NACOSTI/P/23/27601**

135369

Applicant Identification Number

Director General
NATIONAL COMMISSION FOR SCIENCE, TECHNOLOGY & INNOVATION

Verification QR Code

NOTE: This is a computer generated License. To verify the authenticity of this document, Scan the QR Code using QR scanner application.

See overleaf for conditions

Appendix 4: Propagation Spraying Records

Olij Kenya Propagation

Olij Kenya Propagation
P.O. Box 479 - 20117, Naivasha
Tel: +254 715982132
E-mail: CS.KE@DummenOrange.com
Bank of Africa (K) Ltd
Swift: AFRIKENX
CODE: 019-01002
Euro Acc: 03038850022
USD Acc: 03038850010
KShs Acc: 03038850007
BRANCH: WESTLANDS
NAIROBI KENYA

Variety:- Red Rhodos

Client Name:- Reginah Mburu

Order/Batch No:- 231/7/23

Grafting Period: 3rd July 2023-5th August 2023

SPRAY RECORDS:-

Week no.	Date	Chemical	Remark
Week 27	03-Jul	Equation pro	Downy
	05-Jul	Teldor+Agral 90	Botrytis
	07-Jul	topsin	Botrytis
Week 28	10-Jul	Acrobat+kanemite	Downy+mites
	12-Jul	Switch	Botrytis
	14-Jul	Daconil	Botrytis
Week 29	17-Jul	Melody duo +floramite	Downy+mites
	19-Jul	Tecto	Botrytis
	21-Jul	Scala	Botrytis
Week 30	24-Jul	Acrobat+Silwet	Downy+mites
	26-Jul	previcure energy	Downy
	28-Jul	Ridomil+rufast	Downy+mites
Week 31	31-Jul	Melody duo +silwet	Downy+mites
	02-Aug	Previcure energy	Downy
	04-Aug	ortivatop+Arima	Downy+mites

WE OLIJ KENYA PROPAGATION LTD GIVES QUALITY[GUARANTEE] CERTIFICATION:- as below

1. Plants were visibly healthy and free of live pest and diseases.
2. During propagation IPM programme is followed against Spider Mites.
3. A set protocol for cleaning and hygiene were strictly followed during the propagation period.

DECLARATION:-

This is to declare that Olij Kenya Propagation Ltd does not use any form of genetic modification in

OLIJ KENYA PROPAGATION LTD

Appendix 5: Propagation Feeding Records

Olij Kenya Propagation

Olij Kenya Propagation
P.O. Box 479 - 20117, Naivasha
Tel: +254 715982132
E-mail: CS.KE@DummenOrange.com
Bank of Africa (K) Ltd
Swift: AFRIKENX
CODE: 019-01002
Euro Acc: 03038850022
USD Acc: 03038850010
KShs Acc: 03038850007
BRANCH: WESTLANDS
NAIROBI KENYA

Variety:- Red Rhodos

Client Name:- Reginah Mburu

Order/Batch No:- 231/7/23

Grafting Period: 3rd July 2023-5th August 2023

FERTIGATION RECORDS:-

TANK A

Product	Composition	Nutrients Elementals	Molecular Mass (g/mol)
calcium nitrate	$5[ca(NO3)_2.2H_2O].NH_4NO_3$	15.5N, 19ca	1080
Potassium nitrate	KNO_3	13.7N, 38.6 K	101.1
Fe-Chelate 11%	Fe-EDTA	11 Fe	508
Fe-Chelate 6%	Fe-EDTA	6 Fe	1118

TANK B

Product	Composition	Nutrients Elementals	Molecular Mass (g/mol)
mono potassium Phosph	KH_2PO_4	22.7N, 28.7 K	136.1
magnesium nitrate	$Mg(NO_3)_2.6H_2O$	9.5Mg, 10.9N	256
magnesium sulfate	$MgSO_4.7H_2O$	9.7Mg, 13S	246.4
potassium sulfate	K_2SO_4	44.8K, 18.3S	174.3
potassium nitrate	KNo_3	13.7N, 38.6K	101.1
manganese sulphate	$MnSO_4H_2O$	32Mn	169
zinc sulphate	$ZnSO_4.H_2O$	23Zn	287.5
borax	$Na_2B_4O_7.10H_2O$	11B	381.2
Copper sulphate	$CuSO_4.5H_2O$	25Cu	241.7
sodiummolybdate	$Na_2MoO_4.2H_2O$	40Mo	241.9

Appendix 6: Flush 1 Spray Records

Date	Type	Session	Targe	Areas	olume	Equipment	Cocktail
23-Aug-23	Drenching	Morning	TP	Full Greenhouse	100000	Central Unit Station	NEMATECH S (TUBES)@10+@0+@0+@0
09-Sep-23	Beneficials Application	Morning	SM	Full Greenhouse	100000	By hand	PHYTOTECH (TUBES x 2000)@15+@0+@0+@0
14-Sep-23	Beneficials Application	Morning	SM	Full Greenhouse	100000	By hand	REAL PHYTOSEIULUS (1000 x TUBE)@15+@0+@0+@0
19-Sep-23	Chemical Spray	Early Afternoon	CAT	Full Greenhouse	1000	Central Unit Station	PROVE 1.92@60+B.T. (Bacillus Thuringiensis)@50+@0+@0
22-Sep-23	Beneficials Application	Morning	SM	Full Greenhouse	100000	By hand	REAL PHYTOSEIULUS (1000 x TUBE)@1665+@0+@0+@0
28-Sep-23	Chemical Spray	Afternoon	CAT	Full Greenhouse	1000	Central Unit Station	UPHOLD@40+ATTRACKER@40+B.T. (Bacillus Thuringiensis)@50+@0
30-Sep-23	Beneficials Application	Morning	SM	Full Greenhouse	0	By hand	REAL PHYTOSEIULUS (1000 x TUBE)@0+@0+@0+@0
01-Oct-23	Crop Wash	Morning	MB	Full Greenhouse	500	Central Unit Station	DETERGENT@300+@0+@0+@0
03-Oct-23	Beneficials Application	Morning	SM	Full Greenhouse	100000	By hand	REAL CALIFORNICUS (12500 x TUBE)@25+@0+@0+@0
08-Oct-23	Beneficials Application	Early Afternoon	SM	SPOTS	100000	By hand	PHYTOTECH (TUBES x 2000)@0+REAL PHYTOSEIULUS (1000 x TUBE)@750+@0+@0
12-Oct-23	Chemical Spray	Afternoon	TP	Full Greenhouse	1000	Central Unit Station	DELEGATE 250 WG@20+ATTRACKER@100+B.T. (Bacillus Thuringiensis)@50+@0
16-Oct-23	Chemical Spray	Early Morning	TP	Full Greenhouse	1000	Central Unit Station	DELEGATE 250 WG@20+ATTRACKER@100+B.T. (Bacillus Thuringiensis)@50+@0
17-Oct-23	Crop Wash	Morning	MB	Full Greenhouse	1000	Central Unit Station	OMO Detergent@200+@0+@0+@0
19-Oct-23	Spot Chemical Spray	Morning	SM	SPOTS	1500	Central Unit Station	SILWET GOLD@60+@0+@0+@0
20-Oct-23	Chemical Spray	Afternoon	TP	Full Greenhouse	1000	Central Unit Station	DELEGATE 250 WG@20+ATTRACKER@100+B.T. (Bacillus Thuringiensis)@50+@0
29-Oct-23	Chemical Spray	Early Morning	PM	Full Greenhouse	1800	Central Unit Station	NIMROD 25 EC@200+THIOVIT JET@150+GRANFOL K@100+@0
30-Oct-23	Chemical Spray	Afternoon	TP	Full Greenhouse	1000	Central Unit Station	DELEGATE 250 WG@20+ATTRACKER@100+B.T. (Bacillus Thuringiensis)@50+TELDOR 50@100
31-Oct-23	Spot Chemical Spray	Early Afternoon	SM	SPOTS	1200	Central Unit Station	ARIMA 30 SC@60+@0+@0+@0
04-Nov-23	Chemical Spray	Early Afternoon	PM	Full Greenhouse	2000	Central Unit Station	LUNA TRANQUILITY SC 500 @70+NIMBECIDINE@100+@+@
06-Nov-23	Spot Chemical Spray	Morning	SM	Full Greenhouse	1200	Central Unit Station	SILWET GOLD@60+@0+@0+@0
10-Nov-23	Chemical Spray	Early Morning	PM	Full Greenhouse	3000	Central Unit Station	NIMROD 25 EC@200+THIOVIT JET@150+GRANFOL K@100+SILWET GOLD@10
12-Nov-23	Spot Chemical Spray	Early Afternoon	SM	Full Greenhouse	1500	Central Unit Station	ARIMA 30 SC@60+@0+@0+@0
14-Nov-23	Chemical Spray	Morning	PM	Full Greenhouse	3000	Central Unit Station	NIMROD 25 EC@200+THIOVIT JET@150+LITHOVIT@100+@0
15-Nov-23	Chemical Spray	Afternoon	TP	Full Greenhouse	1800	Central Unit Station	AVIRMEC 1.8 EC@50+AFIFEN 10.8 %@100+@0+@0

Appendix 7: Flush 2 Spray Records

Date	Type	Session	Targe	Areas	olume	Equipment	Cocktail
17-Nov-23	Beneficials Application	Morning	SM	Full Greenhouse	100000	By hand	PHYTOTECH (TUBES x 2000)@139+@0+@0+@0
20-Nov-23	Beneficials Application	Morning	SM	Full Greenhouse	100000	By hand	AMBLYTECH (TUBES x 25000)@32+REAL CALIFORNICUS (12500 x TUBE)@10
21-Nov-23	Chemical Spray	Early Afternoon	AP	Full Greenhouse	1800	Central Unit Station	AVIRMEC 1.8 EC@50+AFIFEN 10.8 %@100+@0+@0
22-Nov-23	Chemical Spray	Early Afternoon	PM	Full Greenhouse	3000	Central Unit Station	SOLVIT 175 EW@100+THIOVIT JET@150+LITHOVIT@100+SILWET GOLD@10
25-Nov-23	Chemical Spray	Early Morning	SM	Full Greenhouse	4000	Central Unit Station	BIOMITE@200+SILWET GOLD@30+@0+@0
26-Nov-23	Chemical Spray	Early Afternoon	PM	Full Greenhouse	2800	Central Unit Station	NIMROD 25 EC@250+THIOVIT JET@150+LITHOVIT@100+AGRAL 90@30
29-Nov-23	Chemical Spray	Early Morning	SM	Full Greenhouse	4500	Central Unit Station	FLORAMITE 240 SC@50+SILWET GOLD@30+@0+@0
01-Dec-23	Chemical Spray	Afternoon	PM	Full Greenhouse	2200	Central Unit Station	NIMROD 25 EC@250+THIOVIT JET@150+AGRAL 90@30+@0
11-Dec-23	Chemical Spray	Early Morning	SM	Full Greenhouse	4500	Central Unit Station	BIOMITE@200+AGRAL 90@30+@0+@0
12-Dec-23	Crop Wash	Morning	MB	Full Greenhouse	2000	Central Unit Station	HYDROGEN PEROXIDE@300+@0+@0+@0
13-Dec-23	Chemical Spray	Afternoon	MB	Full Greenhouse	1000	Central Unit Station	CLOSER 240 SC@20+@0+@0+@0
15-Dec-23	Chemical Spray	Early Afternoon	PM	Full Greenhouse	2500	Central Unit Station	NIMROD 25 EC@250+THIOVIT JET@150+AGRAL 90@30+@0
17-Dec-23	Chemical Spray	Early Morning	SM	Full Greenhouse	4500	Central Unit Station	FLORAMITE 240 SC@50+SILWET GOLD@30+@0+@0
17-Dec-23	Chemical Spray	Afternoon	SBO	Full Greenhouse	400	Central Unit Station	CUPRO CAFFARO@200+@0+@0+@0
19-Dec-23	Chemical Spray	Morning	PM	Full Greenhouse	2600	Central Unit Station	NIMROD 25 EC@250+ALERT 50 SC@100+AGRAL 90@30+@0
20-Dec-23	Crop Wash	Morning	MB	Full Greenhouse	2000	Central Unit Station	OMO Detergent@200+@0+@0+@0
21-Dec-23	Chemical Spray	Early Afternoon	MB	Full Greenhouse	2500	Central Unit Station	CALYPSO 480 SC@80+B.T. (Bacillus Thuringiensis)@50+SILWET GOLD@30+@0
23-Dec-23	Chemical Spray	Early Morning	PM	Full Greenhouse	1800	Central Unit Station	NIMROD 25 EC@250+ALERT 50 SC@100+AGRAL 90@30+@0
26-Dec-23	Chemical Spray	Early Morning	SM	Full Greenhouse	4500	Central Unit Station	SILWET GOLD@70+@0+@0+@0
27-Dec-23	Crop Wash	Morning	MB	Full Greenhouse	2000	Central Unit Station	OMO Detergent@200+@0+@0+@0
28-Dec-23	Chemical Spray	Early Afternoon	MB	Full Greenhouse	2600	Central Unit Station	CALYPSO 480 SC@80+B.T. (Bacillus Thuringiensis)@50+SILWET GOLD@30+@0
30-Dec-23	Chemical Spray	Morning	PM	Full Greenhouse	1600	Central Unit Station	NIMROD 25 EC@250+ALERT 50 SC@100+AGRAL 90@30+MKP@100
04-Jan-24	Chemical Spray	Afternoon	PM	Full Greenhouse	1600	Central Unit Station	ENCLOSURE@250+TECTO 500 SC@80+AGRAL 90@30+MKP@200
05-Jan-24	Crop Wash	Morning	MB	Full Greenhouse	2000	Central Unit Station	HYDROGEN PEROXIDE@300+@0+@0+@0
08-Jan-24	Chemical Spray	Early Morning	SM	Full Greenhouse	4500	Central Unit Station	AMINO GOLD@80+@0+@0+@0

Appendix 8: Greenhouse Climate Monitoring Records

Summary Report

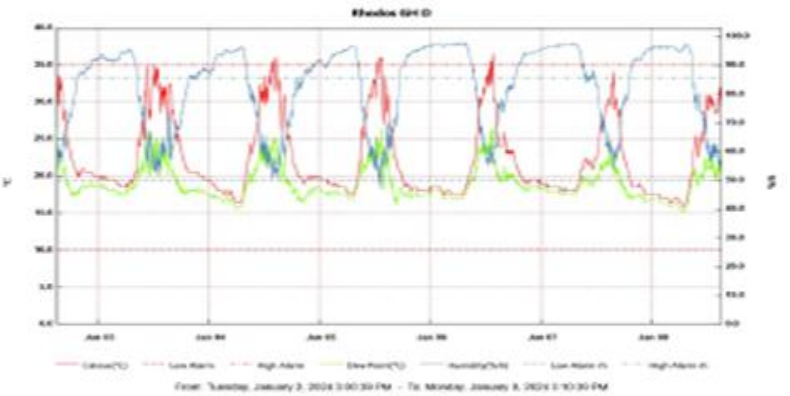
Serial Number	010180845
Logger Name	Rhodos GH D

First Reading	3:00:39 PM 02-01-2024	Alarm	*Triggered*
Last Reading	3:10:39 PM 08-01-2024	Logging Rate	5 Minutes
Elapsed Time	6d, 0h, 10m		
Total Readings	1731		

Celsius	Low Alarm 10.0°C	High Alarm 35.0°C
Minimum 16.0°C 6:15:39 AM 08-01-2024	Alarm Occurrences 0	Alarm Occurrences 9
Maximum 36.5°C 1:25:39 PM 06-01-2024	Total time in alarm N/A	Total time in alarm 2h, 10m
Average Reading 22.43°C Mean Kinetic Temperature 22.34°C (ΔH83.14472)	First Alarm Triggered N/A	First Alarm Triggered 10:30:39 AM 03-01-2024
Standard Deviation 5.19°C	Longest Alarm N/A	Longest Alarm 40m

Humidity	Low Alarm 50.0%rh	High Alarm 85.5%rh
Minimum 47.0%rh 1:10:39 PM 05-01-2024	Alarm Occurrences 2	Alarm Occurrences 16
Maximum 97.5%rh 5:05:39 AM 06-01-2024	Total time in alarm 10m	Total time in alarm 3d, 12h, 40m
Average Reading 82.84%rh	First Alarm Triggered 12:45:39 PM 05-01-2024	First Alarm Triggered 7:35:39 PM 02-01-2024
Standard Deviation 14.20%rh	Longest Alarm 5m	Longest Alarm 16h, 15m

Dew Point	
Minimum 15.2°C 6:15:39 AM 08-01-2024	Maximum 27.3°C 3:35:39 PM 03-01-2024
Average Reading 19.02°C	Standard Deviation 2.18°C



Appendix 9: Vaselife Climate Monitoring Records

Summary Report

Serial Number 010180845
Logger Name Rhodos - Vase

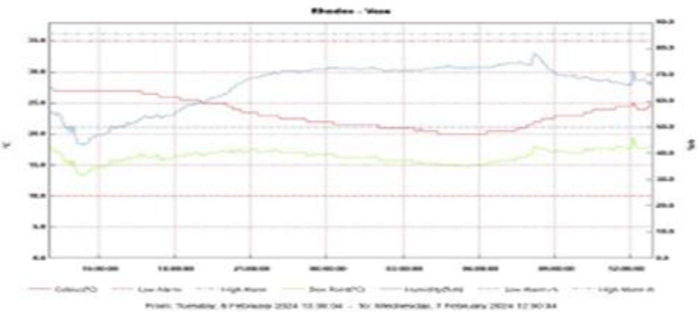
First Reading 13:05:04 06/02/2024
Last Reading 12:50:04 07/02/2024
Elapsed Time 23h, 45m
Total Readings 286

Alarm *Triggered*
Logging Rate 5 Minutes

Celsius	Low Alarm 10.0°C	High Alarm 35.0°C
Minimum: 20.0°C 04:25:04 07/02/2024	Alarm Occurrences: 0	Alarm Occurrences: 0
Maximum: 27.5°C 13:05:04 06/02/2024	Total time in alarm: N/A	Total time in alarm: N/A
Average Reading: 23.34°C; Mean Kinetic Temperature: 23.32°C (ΔH83.14472)	First Alarm Triggered: N/A	First Alarm Triggered: N/A
Standard Deviation: 2.34°C	Longest Alarm: N/A	Longest Alarm: N/A

Humidity	Low Alarm 50.0%rh	High Alarm 85.5%rh
Minimum: 43.0%rh 14:25:04 06/02/2024	Alarm Occurrences: 6	Alarm Occurrences: 0
Maximum: 78.0%rh 08:10:04 07/02/2024	Total time in alarm: 1h, 55m	Total time in alarm: N/A
Average Reading: 65.88%rh	First Alarm Triggered: 13:45:04 06/02/2024	First Alarm Triggered: N/A
Standard Deviation: 8.85%rh	Longest Alarm: 1h, 25m	Longest Alarm: N/A

Dew Point	
Minimum: 13.4°C 14:25:04 06/02/2024	Maximum: 19.5°C 12:05:04 07/02/2024
Average Reading: 16.42°C	Standard Deviation: 1.01°C



Appendix 10: Analysis of variance showing the effect of treatments on Lateral Shoots in cut roses in flush 1 and 2

Flush 1					
Source of Variation	df	SS	MS	F Value	P Value
Block	2	0.4222	0.2111	0.26	0.7718
Hormone	2	11.7284	5.8643	7.22	0.0013
Rate	2	1.2099	0.6049	0.74	0.4782
Hormone*Rate	4	3.3086	0.8272	1.02	0.4032
Flush 2					
Block	2	0.5556	0.2778	0.66	0.5192
Hormone	2	17.3580	8.6790	20.65	<.0001
Rate	2	0.3210	0.1605	0.38	0.6838
Hormone*Rate	4	0.9383	0.2346	0.56	0.6937

Appendix 11: Analysis of variance showing the effect of treatments on stem length in cut roses in flush 1 and 2

Flush 1					
Source of Variation	df	SS	MS	F Value	P Value
Block	2	36.1556	18.0778	1.03	0.3607
Hormone	2	2776.3209	1388.1605	79.34	<.0001
Rate	2	4.0247	2.0123	0.12	0.8915
Hormone*Rate	4	16.2716	4.0679	0.23	0.9193
Flush 2					
Block	2	23.0889	11.5444	0.57	0.5655
Hormone	2	1238.8889	619.4444	30.18	<.0001
Rate	2	0.6667	0.3333	0.02	0.9836
Hormone*Rate	4	0.8889	0.2222	0.01	0.9998

Appendix 12: Analysis of variance showing the effect of treatments on stem diameter in cut roses in flush 1 and 2

Flush 1					
Source of Variation	df	SS	MS	F Value	P Value
Block	2	3.4056	1.7028	1.28	0.2849
Hormone	2	1.6852	0.8426	0.63	0.5346
Rate	2	0.7222	0.3611	0.27	0.7636
Hormone*Rate	4	4.3704	1.0926	0.82	0.5171
Flush 2					
Block	2	6.6687	3.3343	1.60	0.2091
Hormone	2	2.1640	1.0820	0.52	0.5977
Rate	2	2.5454	1.2727	0.16	0.5462
Hormone*Rate	4	3.1723	0.7931	0.38	0.8225

Appendix 13: Analysis of variance showing the effect of treatments on flower bud size in cut roses in flush 1 and 2

Flush 1					
Source of Variation	df	SS	MS	F Value	P Value
Block	2	0.3042	0.1521	1.30	0.2774
Hormone	2	0.1373	0.0686	0.59	0.5577
Rate	2	0.0652	0.0326	0.28	0.7568
Hormone*Rate	4	0.4418	0.1104	0.95	0.4417
Flush 2					
Block	2	0.3307	0.1653	1.14	0.3263
Hormone	2	1.0017	0.5009	3.44	0.0369
Rate	2	0.0640	0.0320	0.22	0.8032
Hormone*Rate	4	0.4346	0.1086	0.75	0.5632

Appendix 14: Analysis of variance showing the effect of treatments on leaf area in cut roses in flush 1 and 2

Flush 1					
Source of Variation	df	SS	MS	F Value	P Value
Block	2	196.6722	98.3361	2.44	0.0935
Hormone	2	2908.5432	1454.2716	36.14	<.0001
Rate	2	8.3395	4.1698	0.10	0.9017
Hormone*Rate	4	52.1975	13.0494	0.32	0.8609
Flush 2					
Block	2	12.3167	6.1583	0.06	0.9401
Hormone	2	3566.8889	1783.4444	17.89	<.0001
Rate	2	37.4630	18.7315	0.19	0.8291
Hormone*Rate	4	20.3703	5.0926	0.06	0.9950

Appendix 15: Analysis of variance showing the effect of treatments on chlorophyll content in cut roses in flush 1 and 2 at preharvest.

Flush 1									
Source of variation	df	At week 3				At week 7			
		SS	MS	F Value	P Value	SS	MS	F Value	P Value
Block	2	0.72	0.36	0.04	0.96	49.92	23.96	7.35	0.0012
Hormone	2	287.63	143.82	15.10	<.0001	644.67	322.33	98.89	<.0001
Rate	2	71.87	35.93	3.77	0.027	21.21	10.61	3.25	0.04
Hormone*Rate	4	104.73	26.18	2.75	0.03	37.39	9.35	2.87	0.03
Flush 2									
Block	2	12.70	6.35	0.41	0.66	15.77	7.88	1.27	0.29
Hormone	2	7.20	3.60	0.23	0.79	1.70	0.85	0.14	0.87
Rate	2	10.50	5.25	0.34	0.71	0.76	0.38	0.06	0.94
Hormone*Rate	4	65.86	14.47	1.08	0.37	18.69	4.67	0.75	0.56

Appendix 16: Analysis of variance showing the effect of treatments on the number of suckers in cut roses in flush 1 and 2 at preharvest.

Flush 1					
Source of Variation	df	SS	MS	F Value	P Value
Block	2	0.5333	0.2667	0.05	0.9498
Hormone	2	650.2593	325.1296	62.81	<.0001
Rate	2	28.4815	14.2407	2.75	0.0739
Hormone*Rate	4	39.0741	9.7685	1.89	0.1280
Flush 2					
Block	2	0.2000	0.1000	0.03	0.9683
Hormone	2	181.5556	90.7778	29.28	<.0001
Rate	2	18.0000	9.0000	2.90	0.0808
Hormone*Rate	4	17.1111	4.2778	1.38	0.2803

Appendix 17: Analysis of variance showing the effect of treatments on bent peduncles in cut roses in flush 1 and 2.

Flush 1					
Source of Variation	df	SS	MS	F Value	P Value
Block	2	21.2333	10.6167	3.25	0.0473
Hormone	2	25.4815	12.7407	3.91	0.0268
Rate	2	0.4815	0.2407	0.07	0.9290
Hormone*Rate	4	2.5185	0.6296	0.19	0.9409
Flush 2					
Block	2	29.4000	14.7000	3.34	0.0585
Hormone	2	14.5185	7.2593	1.65	0.2201
Rate	2	0.0741	0.0370	0.01	0.9916
Hormone*Rate	4	1.9259	0.4815	0.11	0.9777

Appendix 18: Analysis of variance showing the effect of treatments on flush days in cut roses in flush 1 and 2.

Flush 1					
Source of Variation	df	SS	MS	F Value	P Value
Block	2	0.4667	0.2333	0.10	0.9092
Hormone	2	12.2469	6.1234	2.50	0.0884
Rate	2	0.7654	0.3827	0.16	0.8555
Hormone*Rate	4	1.8271	0.4568	0.19	0.9447
Flush 2					
Block	2	23.0887	11.5444	4.24	0.0179
Hormone	2	11.2840	5.6420	2.07	0.1329
Rate	2	0.7654	0.3827	0.14	0.8691
Hormone*Rate	4	0.7901	0.1975	0.07	0.9902

Appendix 19: Analysis of variance showing the effect of treatments on the number of petals in cut roses in flush 1 and 2.

Flush 1					
Source of Variation	df	SS	MS	F Value	P Value
Block	2	12.4222	6.2111	1.45	0.2407
Hormone	2	0.9136	0.4568	0.11	0.8990
Rate	2	2.3951	1.1975	0.28	0.7568
Hormone*Rate	4	6.5679	1.6420	0.38	0.8199
Flush 2					
Block	2	8.9556	4.4778	1.13	0.3272
Hormone	2	0.4691	0.2346	0.06	0.9424
Rate	2	0.0988	0.0494	0.01	0.9876
Hormone*Rate	4	3.8272	0.9568	0.24	0.9136

Appendix 20: Analysis of variance showing the effect of treatments on chlorophyll content in cut roses at postharvest in flush 1 and 2.

Flush 1					
Source of Variation	df	SS	MS	F Value	P Value
Hormone	2	3318.7514	1659.3757	522.18	<.0001
Rate	8	86.7202	10.8400	3.41	0.0031
Hormone*Rate	16	1329.4931	83.0933	26.15	<.0001
Flush 2					

Hormone	2	1893.4867	946.7433	201.98	<.0001
Rate	8	58.5889	7.3236	1.56	0.1581
Hormone*Rate	16	564.8444	35.3040	7.53	<.0001

Appendix 21: Analysis of variance showing the effect of treatments on the Lightness (L*) of Petal colour in cut roses at postharvest in flush 1 and 2.

Flush 1					
Source of Variation	df	SS	MS	F Value	P Value
Hormone	2	3821.5377	1910.7689	1921.33	<.0001
Rate	8	327.3637	40.9205	41.15	<.0001
Hormone*Rate	16	670.1194	41.8825	42.11	<.0001
Flush 2					
Hormone	2	4763.7459	2381.8729	1685.03	<.0001
Rate	8	322.7699	40.3462	28.54	<.0001
Hormone*Rate	16	1006.1383	62.8836	44.49	<.0001
Rate	8	327.3637	40.9205	41.15	<.0001

Appendix 22: Analysis of variance showing the effect of treatments on the Redness (a*) of Petal colour in cut roses at postharvest in flush 1 and 2.

Flush 1					
Source of Variation	df	SS	MS	F Value	P Value
Hormone	2	3289.9509	1644.9755	383.40	<.0001
Rate	8	263.0642	32.8830	7.66	<.0001
Hormone*Rate	16	173.9699	10.8731	2.53	0.0056
Flush 2					
Hormone	2	4162.7807	2081.3904	853.98	<.0001
Rate	8	433.9867	54.2483	22.26	<.0001
Hormone*Rate	16	352.0638	22.0040	9.03	<.0001

Appendix 23: Analysis of variance showing the effect of treatments on the Yellowness (b*) of Petal colour in cut roses at postharvest in flush 1 and 2.

Flush 1					
Source of Variation	df	SS	MS	F Value	P Value
Hormone	2	1054.0477	527.0239	146.94	<.0001
Rate	8	7.4901	0.9363	0.26	0.9757
Hormone*Rate	16	104.2484	6.5155	1.82	0.0528
Flush 2					
Hormone	2	1432.1267	716.0634	301.75	<.0001
Rate	8	16.5944	2.0743	0.87	0.5440
Hormone*Rate	16	133.3529	8.3346	3.51	0.0003

Appendix 24: Analysis of variance showing the effect of treatments on weight loss in cut roses at postharvest in flush 1 and 2.

Flush 1					
Source of Variation	df	SS	MS	F Value	P Value
Hormone	2	507.5678	253.7839	242.21	<.0001
Rate	8	133.7911	16.7239	15.96	<.0001
Hormone*Rate	16	288.1997	18.0125	17.19	<.0001

Flush 2					
Hormone	2	301.4219	150.7109	134.02	<.0001
Rate	8	141.9302	17.7413	15.78	<.0001
Hormone*Rate	16	203.0192	12.6887	11.28	<.0001

Appendix 25: Analysis of variance showing the effect of treatments on Vaselife in cut roses at postharvest in flush 1 and 2.

Flush 1					
Source of Variation	df	SS	MS	F Value	P Value
Hormone	2	46.8887	23.4444	39.36	<.0001
Rate	8	11.7778	1.4722	2.47	0.0232
Hormone*Rate	16	32.6667	2.0417	3.43	0.0004
Flush 2					
Hormone	2	44.1728	22.0864	46.73	<.0001
Rate	8	11.8025	1.4753	3.12	0.0057
Hormone*Rate	16	11.6049	0.7253	1.53	0.1216