INTEGRATED MANAGEMENT OF *Agrobacterium tumefasciens* Cavara*;* THE CAUSAL AGENT OF CROWN GALL DISEASE IN ROSES (*Rosa hybrida*)

MARY OPISA ONIANG'O

A Thesis Submitted to the Graduate School in Partial Fulfilment of the Requirements for the Award of Doctor of Philosophy Degree in Plant Pathology of Egerton University

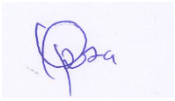
EGERTON UNIVERSITY

OCTOBER, 2025

DECLARATION AND RECOMMENDATION

Declaration

This thesis is my original work and has not been submitted for examination in any other university.

Signature  Date: 1st October 2025

Mary Opisa Oniang'o

SD15/23552/14

Recommendation

This thesis has been submitted with our approval as supervisors according to Egerton University regulations.

Signature  Date: 1st October 2025

Prof. Daniel O. Otaye (Ph.D.)

Department of Biological Sciences

Egerton University

Signature  Date 1 st October 2025

Dr. Japhet M. Muthamia (Ph.D.)

Department of Biological Sciences

Egerton University

Signature For  posthumous Date: 1 st October 2025

Prof. Oliver S. Achwanya (Ph.D.)

Department of Biological Sciences

Egerton University

DEDICATION

To my dear husband Rono and daughters Susan, Sharlene, Vanessa, Mitchell, Arelle and my grand daughter Tessylaine for their moral support and encouragement. To my dear parents Mr. and Mrs. Oniang'o for the strong foundation of education they gave me that has led to this great achievement.

ACKNOWLEDGEMENTS

I would like to thank the almighty God for giving me an opportunity to pursue my studies at Egerton University. I would also like to thank the Egerton University for allowing me to study my Ph.D. degree and for the thorough training I have received from the institution while undertaking my undergraduate and master's degrees. Much gratitude also goes to my supervisors, the late Prof. Oliver Achwanya, Prof. Daniel Otaye and Dr. Japhet Muthamia, Dean of students for their suggestions, advice and critical evaluation of this project, guidance in publications and writing up this thesis. Special thanks also go to Prof. Nzula Kitaka, former Director of Graduate School Egerton University, Prof. Julius Kipkemboi, former Dean Faculty of Science, for their encouragement and moral support. My appreciation also goes to the Managing Director Browns Plantations Kenya LTD Mr. Simeon Hutchison, for allowing me to carry out my project and for providing the physical facilities which were used to carry out my studies at James Finlay Kenya Limited Kericho County, Tarakwet farm, which has now been acquired by Browns Plantations LTD since 2023. I would also like to thank Mr. Robert Korir of KALRO- Kericho, Research and Development staff of Finlay flowers and Botanicals division for their moral support while carrying out the experiments. Special thanks also go to my parents Mr. and Mrs. Oniang'o, my daughters Susan, Vanessa, Shirleen, Mitchell and Arelle for their encouragement and moral support. My husband Mr. Kipkoech Rono Chepkwony for the financial support and encouragement as well as his patience throughout my studies. I also thank my colleague Mr. Aggrey Simiyu and Rev. Chumo whose prayers and encouragement greatly contributed towards the success of this project.

ABSTRACT

Crown gall caused by *Agrobacterium tumefasciens* is one of the major diseases currently threatening the flower industry in Kenya. The disease significantly reduces production of roses by 50 – 100 % on susceptible rose varieties. Currently, there is limited understanding on the effectiveness of various cultural, biological and chemical control measures used by the flower growers in Kenya. The main objective of this study was to contribute to the increased productivity and quality of marketable roses through integrated management of *A. tumefasciens*. The specific objectives were to test the effect of various biostimulants such as foltron, biozyme, alexin, hicure and codamine radicular through spray or drench in suppressing *A. tumefasciens* in roses. The effect of sterilizing agents such as Dettol 0.5 ml / L, 1.0 ml / L and hydrogen peroxide1.0 ml / L, agrowipe (botanic neem extract) undiluted and vegetable oil (fresh fri) undiluted were tested in suppressing crown gall growths. Biological agents: *Trichoderma asperellum* 0.25 Kg / Ha and 0.5 Kg / Ha and *Bacillus subtilis* 0.2 L / Ha and 0.4 L / Ha were also tested in suppressing *A. tumefasciens* in roses. The effect of selected pesticides at various rates as control agents of *A. tumefasciens* in roses was also tested. Experiments were conducted at James Finlay Kenya LTD Tarakwet farm Kericho County in greenhouse F36 planted with rose variety tropical amazon. Ten galls located 15 cm above the growing media – pumice, were tagged on each treatment replicate and the initial gall diameter measured using a Vanier caliper. The galls were cut using a sterilized roll cut, treatments applied, and the new crown galls growths were measured once a week for twelve months, then cut and weighed using servo weighing balance. The number of fresh crown gall growths on each plot were counted, removed and the diameter measured once a month for twelve months. The number of marketable stems was counted daily from each treatment replicate for a period of twelve months to determine the yield. The quality of stems was determined by sampling 30 stems once a week from each treatment replicate and stem length, weight, and head size measured. Similar experiments were also conducted in pots under controlled environment. Results showed that the various biostimulants increased yield and quality of roses. Botanic neem extract and vegetable oil suppressed crown gall growths and had a higher yield and quality of roses than copper oxychloride. Previcur energy and enrich BM did not suppress *A. tumefasciens*. *Bacillus subtilis* and *T. asperellum* suppressed crown gall growth under controlled environment. It was concluded that integrated management of *A. tumefasciens* using biologicals, biostimulants and plant extracts increased yield and quality of roses and therefore recommended for use in the flower industry.

TABLE OF CONTENTS

DECLARATION AND RECOMMENDATION ii

COPY RIGHT iii

DEDICATION iv

ACKNOWLEDGEMENTS v

ABSTRACT vi

LIST OF TABLES xii

LIST OF FIGURES xiviv

LIST OF PLATES xvi

LIST OF ACRONYMS AND ABBREVIATIONS xvii

CHAPTER ONE 1

INTRODUCTION 1

1.1. Background information 1

1.2. Statement of the problem 3

1.3. Objectives 4

1.3.1. General objective 4

1.3.2. Specific objectives 4

1.4. Hypotheses 4

1.5. Justification 4

1.6. Scope and limitations 5

CHAPTER TWO 6

LITERATURE REVIEW 6

2.1. Floriculture industry in the world 7

2.1.1. Floriculture industry in Africa 7

2.1.2. Flower industry in Kenya 11

2.1.3. Roses production constraints 14

2.1.4. Integrated disease and pest management for roses 17

2.1.5. Post-harvest handling and cold chain management 19

2.1.6. Global competitiveness and future prospects of Kenya's flower industry 23

2.1.7. Competition from other producing countries 25

2.1.8. Environmental and Climate Risks 26

2.1.9. Agronomy of *Rosa hybrida* 28

2.1.10. Crown gall disease in roses 30

2.1.11. Genome structure of *Agrobacterium tumefasciens* 36
2.1.12. Pathogenesis of *Agrobacterium tumefasciens* 36
2.1.13. Symptoms of crown gall in roses 39
2.1.14. Host range and distribution 41
2.1.15. Biotypes of *Agrobacterium tumefasciens* 44
2.2. Management of *Agrobacterium tumefasciens* 46
2.2.1. Cultural control 48
2.2.2. Biological control using *Agrobacterium radiobacter* 50
2.2.3. Biological control using *Trichoderma* spp. 52
2.2.4 Biological control using *Bacillus subtilis* 53
2.2.5. Chemical control 54
2.2.6. Future prospects in crown gall management 56
2.3. Induced systemic resistance 57
2.4. Use of Biostimulants 59
CHAPTER THREE 63
MATERIALS AND METHODS 63
3.1. Study site 63
3.2. Testing the efficacy of various biostimulants in controlling *Agrobacterium tumefasciens* in roses 65
3.2.1. Green house experiment 65
3.2.2. Pot trials 70
3.3. Testing effect of sterilizing agents and pesticides in suppressing crown gall growth 73
3.3.1. Green house trials 73
3.3.2. Pot experiment 74
3.3.3. Data collection 75
3.4. Testing effect of *Bacillus subtilis* and *Trichoderma asperellum* in suppressing *Agrobacterium tumefasciens* 75
3.4.1. Green house trial 75
3.4.2. Pot trials 76
3.5. Testing effect of selected pesticides in controlling *Agrobacterium tumefasciens* 76
3.5.1. Pot trials 76
3.5.2. Green house experiment 76
3.6. Data analysis 77
CHAPTER FOUR 78

RESULTS **78**
4.1. Effect of various biostimulants in suppressing *Agrobacterium tumefasciens* in roses .78
4.2. Green house experiment 78
4.2.1. Yield of roses 78
4.2.2. Fresh galls, diameter and weight of crown gall tumours 80
4.2.3. Flower quality- stem length, weight head length and width 83
4.3. Pot experiment 84
4.4. The effect of Agrowipe (botanic neem extract), Fresh fri (vegetable oil) sterilizing agents and pesticides in controlling *Agrobacterium tumefasciens* 85
4.4.1. Yield of marketable stems 86
4.4.2. Diameter, weight and fresh crown gall tumours 88
4.4.3. Quality of rose stems 89
4.4.4. Pot experiment - mean tumour diameter and tumour weight 90
4.5. Effect of selected fungicides in controlling *Agrobacterium tumefasciens* 93
4.5.1. Field experiment – flower quality (stem length and head size) 93
4.5.2. Mean yield and stem weight 94
4.5.3. Mean fresh galls and gall diameter 94
4.6. Pot experiment 95
4.6.1. Mean fresh galls and gall diameter 95
4.7. Effect of selected biological agents in controlling *Agrobacterium tumefasciens* 99
4.7.1. Field trial - mean production and stem weight 99
4.7.2. Fresh galls and gall diameter 100
4.7.3. Stem length and head size 101
4.8. Pot trials 101
CHAPTER FIVE **105**
DISCUSSIONS **105**
5.1. Effect of selected biostimulants in suppressing *Agrobacterium tumefasciens* in roses 105
5.2. Effect of sterilizing agents, agrowipe, Fresh fri and fungicide in suppressing crown gall growth 107
5.3. Effect of *Bacillus subtilis a*nd *Trichoderma asperellum* in controlling *Agrobacterium tumefasciens* 109
5.4. Effect of Previcur energy and enrich BM in controlling *Agrobacterium tumefasciens* 112

CHAPTER SIX 115
CONCLUSIONS AND RECOMMENDATIONS.....115
6.1. Summary 115
6.2. Conclusions 115
6.3. Recommendations 116
6.4. Suggestions for further research……..117
REFERENCES.....119
APPENDICES……..…...147
Appendix 1: Raw data for yield of roses on plots treated with various biostimulants ……..147
Appendix 2: Raw data for galls diameter on plots treated with various biostimulants …….148
Appendix 3: Raw data for fresh galls on plots treated with various biostimulants……150
Appendix 4: Raw data for stem length on plots treated with various biostimulants……..151
Appendix 5: Raw data for stem weight on plots treated with various biostimulants…….152
Appendix 6: Raw data for rose head length on plots treated with various biostimulants…..153
Appendix7: Raw data for fresh galls on plots treated with sterilizing agents……155
Appendix 8: Raw data for yield of roses on plots treated with various sterilizing agents….156
Appendix 9: Raw data for gall diameter on plots treated with various sterilizing agents…..157
Appendix 10: Raw data for gall weight on plots treated with various sterilizing agents…...158
Appendix 11: Raw data on yield of roses on plots treated with various fungicides……..159
Appendix 12: Raw data on gall diameter on plots treated with various fungicides……...159
Appendix 13: Raw data on fresh galls on plots treated with various fungicides……...160
Appendix 14: Raw data on stem length of roses on plots treated with various fungicides…161
Appendix 15: Raw data on roses head length on plots treated with various fungicides……161
Appendix 16: Raw data of roses head width on plots treated with various fungicides……..162
Appendix 17: Raw data on stem weight of roses on plots treated with various fungicides…163
Appendix 18: Raw data on yield of roses on plots treated with various biologicals…….163
Appendix 19: Raw data for fresh galls on plots treated with various biologicals……..…164
Appendix 20: Raw data for fresh galls on plots treated with various biologicals (pot trial)..165
Appendix 21: Raw data on rose stem length on plots treated with various biologicals…….166
Appendix 22: Raw data on roses stem weight on plots treated with various biologicals…..166
Appendix 23: Raw data on roses head width on plots treated with various biologicals……167
Appendix 24: Raw data on roses head length on plots treated with various biologicals…...168
Appendix 25: Analysis of variance for various biostimulants ……..……169
Appendix 26: Analysis of variance for various sterilizing agents ...…….....173

Appendix 27: Analysis of variance for various fungicides……180
Appendix 28: Analysis of variance for various biologicals……...178
Appendix 29: Publication 1……...….185
Appendix 30: Publication 2……....186
Appendix 31: Research permit……...187

LIST OF TABLES

Table 1: Various biostimulant treatments, their composition and mode of application 68

Table 2: Roses feeding program for macro and micro elements……..69

Table 3: Mean yield of roses and percentage increase on plots treated with various biostimulants (pooled data for one year)……79

Table 4: Mean fresh galls, gall diameter and gall weight on plots treated with various biostimulants (pooled data for one year)……..…….81

Table 5: Mean stem length, head length, head width and stem weight on plots treated various biostimulants (pooled data for one year) ……..…….83

Table 6: Mean new galls gall diameter and gall weight on plots treated with various biostimulants (pooled data for one year)…….....85

Table 7: Mean yield of marketable stems on plots treated with copper oxychloride, vegetable oil, botanic neem extract, chloroxylenol 4.8 %, hydrogen peroxide and untreated control (pooled data for one year)…….,,,…..……..87

Table 8: Effect copper oxychloride, vegetable oil, botanic neem extract, chloroxylenol 4.8 %, Hydrogen peroxide and untreated control on crown gall tumours rose variety tropical amazon (pooled data for one year).....88

Table 9: Effect of copper oxychloride, vegetable oil, botanic neem extract, chloroxylenol 4.8 %, hydrogen peroxide and untreated control on quality of roses variety tropical amazon (pooled data for one year) 89

Table 10: Effect copper oxychloride, vegetable oil, botanic neem extract, chloroxylenol 4.8 %, hydrogen peroxide 50 % and untreated control on crown gall tumours rose variety tropical amazone.....90

Table 11: Mean production and stem weight on plots treated with previcur energy, enrich BM and control (water only) 93

Table 12: Mean production and mean stem weight on plots treated with previcur energy, enrich BM and control (water only) ……..……...…….94

Table 13: Mean fresh gall and gall diameter on plots treated with previcur energy, enrich BM and control (water only) ……..……..95

Table 14: Mean fresh gall and gall diameter on plots treated with previcur energy, enrich and control (water only) – pot experiment……...……..96

Table 15: Mean production and stem weight on plots treated with *Bacillus subtilis*, *Trichoderma asperellum* and control (water only) ……..…99

Table 16: Mean fresh galls, gall diameter and gall weight on plots treated with *Bacillus subtilis*, *Trichoderma asperellum* and control (water only) …….100

Table 17: Mean stem length and head size on plots treated with *Bacillus subtilis*, *Trichoderma. asperellum* and control (water only) ……..…..……....101

Table 18: Mean stem length and head size on plots treated with *Bacillus subtilis*, *Trichoderma asperellum* and control (water only) ……..…….....102

LIST OF FIGURES

Figure 1: Map of Kenya showing Kericho County ……..…..…….……64

Figure 2: Biostimulants experimental lay out; R1 – R4 replicates, G guard rows, 1-11randomized plots……...……67

LIST OF PLATES

Plate 1: (a) Healthy rose plants, (b) rose plants infected by crown gall…….…….40

Plate 2: Pot experimental layout for testing various biostimulants……..……72

Plate 3: (a) Crown gall treated with copper oxychloride before cutting gall (b) after cutting gall (c) gall at 12 months……..……91

Plate 4: (a) Crown gall treated with fresh fri before cutting gall (b) after cutting gall (c) gall at 12 months…….91

Plate 5: (a) Crown gall treated with agrowipe before cutting gall (b) after cutting gall (c) gall at 12 months……..……...……91

Plate 6: (a) Crown gall treated with dettol 0.5 ml / L before cutting gall (b) after cutting gall (c) gall at 12 months.....91

Plate 7: (a) Crown gall treated with dettol 1.0 ml / L before cutting gall (b) after cutting gall (c) gall at 12 months……..…….92

Plate 8: (a) Crown gall treated with hydrogen peroxide before cutting gall (b) after cutting gall (c) gall at 12 months…….……....92

Plate 9: (a) Crown gall treated with water only (control) before cutting gall (b) after cutting gall (c) gall at 12 months…….….92

Plate 10: (a) Crown gall treated with previcur energy 3.0 ml / L before cutting gall (b) after cutting gall (c) gall at 12 months……..….97

Plate 11: (a) Crown gall treated with previcur energy 4.0 ml / L before cutting (b) after cutting gall (c) gall at 12 months…….....97

Plate 12: (a) Crown gall treated with enrich BM 2 g / L before cutting gall (b) after cutting gall (c) gall at 12 months…….97

Plate 13: (a**)** Crown gall treated with enrich BM 4 g / L before cutting gall (b) after cutting gall (c) gall at 12 months……...…….….98

Plate 14: (a) Crown gall treated with water only (control) before cutting gall (b) after cutting (c) gall at 12 months …….……....98

Plate 15: (a) Crown gall treated with *B. subtilis* 2 ml / L before cutting gall (b) after cutting gall (c) gall at 12 months …….……..103

Plate 16: (a) Crown gall treated with *B. subtilis* 4 ml / L before cutting gall (b) after cutting gall (c) gall at 12 months …….103

Plate 17: (a) Crown gall treated with *T. asperellum* 0.25 g / L before cutting gall (b) after cutting gall (c) gall at 12 months.……....103

Plate 18: (a) Crown gall treated with *T. asperellum* 0.5 g / L before cutting gall (b) after cutting gall (c) gall at 12 months ……..104

Plate 19: (a) Crown gall treated with water only (control) before cutting gall (b) after cutting gall (c) gall at 12 months …..……...….104

LIST OF ACRONYMS AND ABBREVIATIONS

AFDB	African Development Bank
ANOVA	Analysis of variance
CM	Centimetre
Co.	Company
CRD	Completely Random Design
Dudutech K	Dudu technology Kenya
EDTA	Ethylenediaminetetraacetic acid
GLOBAL G.A.P	Global Good Agricultural Practices
HCD	Horticultural Crops Directorate
IFTEX	International Floriculture Trade Fair
ISR	Induced Systemic Resistance
IPM	Integrated pest management
KFC	Kenya Flower Council
KEPHIS	Kenya Plant Health Inspectorate Service
MPS	Milieu Programma Sierteelt
LSD	Least Significant Difference
LTD	Limited
MAP	Mono ammonium phosphate
MKP	Mono potassium phosphate
NO	Numbers
PGPR	Plant Growth Promoting Rhizobacteria
RCBD	Randomised Completely Block Design.
WP	Wettable powder

CHAPTER ONE
INTRODUCTION

1.1. Background information

Kenya's horticultural industry has grown to become a cornerstone of both national and international agricultural trade. Ranked as the third-largest foreign exchange earner after tourism and tea, the industry is not only critical to Kenya's economy but also serves as a major livelihood source for millions of people (Chepoghisho, 2019). The horticultural sector encompasses a wide variety of crops, including fruits, vegetables, potatoes, and flowers, but among these, the floriculture industry has emerged as the most globally competitive subsector. Kenya is now recognized as one of the leading producers and exporters of cut flowers, with a particular dominance in rose production, which constitutes over 70 % of its flower exports (Resource Trade Earth, 2023). The geographical distribution of flower production across regions such as Lake Naivasha, Kinangop, Nakuru, Limuru, Athi River, Thika, Kiambu, and Eldoret reflects the country's agro-ecological diversity. These high-altitude regions, with their volcanic soils, moderate temperatures, and reliable sunlight, provide ideal growing conditions for high quality flowers throughout the year (AFDB, 2013). Strategic proximity to Jomo Kenyatta International Airport in Nairobi further enhances export logistics, enabling rapid access to key European markets. The Netherlands, the United Kingdom, Germany, and other EU nations remain the largest destinations for Kenyan flowers, with over 70 % of exports heading to Europe (Horticultural Crops Directorate, 2022).

From an economic perspective, the floriculture industry plays an outsized role relative to its scale. In 2023, the sector contributed approximately USD 1.09 billion in export revenue, equivalent to about 1.25 % of national GDP (Mordor Intelligence, 2024). Beyond direct revenue generation, the industry sustains more than 500,000 jobs and indirectly supports the livelihoods of nearly 2 million people. The high participation of women is estimated at 70 % of the workforce underscores the industry's role in gender empowerment and poverty alleviation, particularly in rural communities (Central Bank of Kenya, 2021; Floral Daily, 2023). Employment extends beyond farm-level activities to include a range of ancillary services such as packaging, transport, catering, and housing around flower production hubs (Riungu & Kariuki, 2019). However, the industry is not without its challenges. Disease pressure is one of the most persistent threats to profitable flower production. Roses, which dominate Kenya's floriculture exports, are susceptible to a wide range of fungal and bacterial diseases such as powdery mildew (*Sphaerotheca pannosa*), downy mildew (*Peronospora*

sparsa), black spot (*Diplocarpon rosae*), verticillium wilt (*Verticillium dahliae*), and crown gall (*Agrobacterium tumefaciens*) (Kariuki, 2015). Crown gall, in particular, has emerged as a major constraint due to its persistence and lack of effective chemical or biological control measures.

Crown gall disease, caused by *A. tumefasciens*, poses a unique challenge because of its genetic mechanism of pathogenicity. The bacterium transfers a portion of its Tumor-Inducing (Ti) plasmid DNA into the host plant's genome, leading to uncontrolled cell division and tumor formation at the crown, roots, and stems of infected plants (Furuya *et al*., 2004). Roses infected by crown gall disease develop rounded tumours which are brown to brownish black in colour, they change from smooth, spongy to a rough texture as they age. As the tumours expand, they destroy the adjacent healthy tissue preventing the normal flow of water and nutrients (Agrios, 1997). This process not only weakens the structural integrity of the plant but also interferes with nutrient and water transport, thereby reducing flower quality and yield (Collins, 2001). The disease has a long survival period in soil, with reports indicating persistence for over ten years, which makes eradication extremely difficult (Agrios, 2005).The first major outbreak of crown gall in Kenya was documented in 1998, devastating several commercial rose farms (Arim, 2011). Investigations revealed that the pathogen was introduced through infected rose rootstocks imported from Israel, highlighting the risks associated with international plant material movement (Maina *et al.,* 2013). Since then, crown gall has remained a recurrent problem in rose production systems. Despite the use of copper-based compounds as a management strategy, their effectiveness has been limited due to the pathogen's resistance and the phytotoxic effects on crops (Agrios, 2005). Breeding programs to develop resistant rose varieties have also made little progress, as most commercially popular varieties remain highly susceptible (Arim, 2011).

In addition to disease challenges, the floriculture industry in Kenya faces mounting pressure from environmental and social sustainability concerns. Large-scale flower cultivation has raised questions about water usage, particularly in ecologically sensitive areas like Lake Naivasha, where intensive irrigation competes with domestic and environmental water needs (Odongo, 2023). The extensive use of agrochemicals, while critical for disease and pest management, has also generated debates about occupational health risks, pesticide residues, and biodiversity impacts (Dinham, 2003). Socially, while the industry has been celebrated for providing employment, it has also faced criticism regarding labor conditions, gender equity, and workers' rights (Dolan *et al.,* 2012). Nonetheless, the sector continues to adapt to these challenges. Certification schemes such as Fairtrade, MPS, and the Floriculture Sustainability

Initiative (FSI) have been embraced to ensure compliance with environmental and social standards, particularly for export to European markets where consumer awareness is high (Hawkins *et al.,* 2019). Additionally, research into integrated pest and disease management (IPDM), water-use efficiency, and climate-smart agricultural practices is being promoted to enhance the long-term sustainability of the industry (Kenya Flower Council, 2021).

In conclusion, Kenya's horticultural and floriculture industries represent a critical component of the global flower trade and a vital source of foreign exchange and livelihoods domestically. While the sector is internationally competitive, its sustainability is threatened by persistent plant diseases such as crown gall, alongside environmental and social challenges. Addressing these constraints through research, innovation, and sustainability driven practices will be crucial for ensuring Kenya maintains its position as one of the world's leading exporters of cut flowers

1.2. Statement of the problem

Crown gall disease caused by *A. tumefasciens* is one of the major diseases currently threatening the flower industry in Kenya. Presently, *A. tumefasciens* is widely spread in Kenyan nurseries, commercial production areas, and uncultivated fields. Pathogenic strains of *A. tumefasciens* can live saprophytically in soil for up to ten years and the disease significantly reduces production of roses by 50 – 100 %. Seriously infected fields do not attain their production targets and are usually uprooted early before the average seven years' life span of productive roses. Currently, there is limited understanding on the effectiveness of various cultural, biological and chemical control measures used by the flower growers in Kenya. As a result, flower growers are left vulnerable to the disease and spend considerable amount of money trying various methods to control or minimize the effect of *A. tumefasciens* in roses. A study done on management of crown gall disease of roses in Kenya showed that rose growers planted seedlings from certified propagators and practiced integrated disease management to avoid infections and manage crown gall. The wounds on the plants, mainly caused by harvesting were painted with copper-based fungicides but their effect has never been satisfactory. The use of synthetic pesticides has affected negatively the export of horticultural produce by detection of traces above the regulatory residue levels leading to market loss. An increase in the quality and quantity of roses has been observed by removing galls and pasting with vegetable oils, drenching *Trichoderma asperellum, Bacillus subtilis* and use of sterilizing agents such as hydrogen peroxide and dettol. Crown gall tumours have been observed to completely dry up by applying botanic neem extract such as agrowipe. In addition, some rose

growers in Kenya have observed an increase in the quality and quantity of rose stems treated with various biostimulants. However, limited quantitative scientific experiments have been conducted hence the reason for this study.

1.3. Objectives

1.3.1. General objective

The general objective of this research was to contribute to the increased productivity and quality of marketable roses through integrated management of *Agrobacterium tumefasciens*, the causal agent of crown gall disease in roses.

1.3.2. Specific objectives

i. To test the effect of various biostimulants in suppressing *A. tumefasciens* in roses.
ii. To test the effect of sterilizing agents such as dettol and hydrogen peroxide, agrowipe (botanic neem extract and vegetable oil (fresh fri) in suppressing crown gall growths after cutting galls.
iii. To test the effect of biological agents; *Trichoderma asperellum* and *Bacillus subtilis* in suppressing *A. tumefasciens* in roses.
iv. To test the effect of selected pesticides at various rates as control agents of *A. tumefasciens* in roses.

1.4. Hypotheses

i. Biostimulants do not suppress *A. tumefasciens* in roses.
ii. Sterilizing agents such as dettol and hydrogen peroxide, agrowipe (botanic neem extract) and vegetable oil (fresh fry) do not supress growth of crown galls growths after cutting galls.
iii. Biological agents: *Trichoderma asperellum* and *Bacillus subtilis* do not suppress *A. tumefasciens* in roses.
iv. Selected pesticides do not control *A. tumefasciens* in roses.

1.5. Justification

Crown gall disease caused by *A. tumefasciens* is of economic importance in Kenya since it causes reduction in the yield of roses by reducing the crop life cycle, quality and quantity of rose stems. A survey done in East Africa in 2012 on the losses caused by *A. tumefasciens* showed that crown gall incidence on susceptible varieties was between 75 – 95

% when the crop was only three years old. The high incidence of infection led to a decision to uproot the crop before the normal 7 years of the crop cycle hence leading to high crop losses. Current rose management regimes in Kenya lack protocol for this disease hence farmers resort to untested solutions. In addition, there are no registered pesticides in Kenya and East Africa for controlling crown gall disease which continues to threaten the flower industry. A survey done in 2012 showed that a big head variety treated twice with dygall *(Agrobacterium radiobacter* strain K84) at the seedling stage had 100 % infection. In one and a half years, the production had reduced by 38 %. Galls formed where the stems were cut during harvesting and on leaf node eyes. In addition, trials conducted with copper-based fungicides showed that galls did not develop on the treated area but developed somewhere else on the treated stem indicating the treatment was localized and not able to contain the disease. Biostimulants have been known to stimulate plant growth, mitigate stress-induced limitations, increase yield and may contribute to reducing the use of chemical fertilizers and pesticides. The use of biologicals such as *Bacillus subtilis*, *Trichoderma asperellum* and plant extracts such as the botanic neem extract to control *A. tumefasciens* are alternatives to sustaining high production in roses and are key components to integrated management. They are also safer to the user and environment. There is an urgent need to look for effective and safe control measures to ensure competitiveness of the rose crop. This research therefore gives information on the integrated management strategies to control, suppress or minimize the effect of crown gall disease in roses. This research will help address Sustainable Development Goal 8 (SDG 8) ensuring economic growth and providing employment especially for women and youth.

1.6. Scope and limitations

The research was conducted at James Finlay Kenya limited Tarakwet farm Kericho County in a commercial rose green house. A rose variety, tropical amazone that was susceptible to *A. tumefasciens* was used. Pot trials were also conducted to measure the effect of various treatments closely. For the greenhouse trial, a randomised completely block design (RCBD) was used while for the pot trial completely randomised design was used (CRD). The research was conducted for a period of four years. The limitation of the study was unavailability of clean planting material from propagators of rose seedlings in Kenya.

CHAPTER TWO
LITERATURE REVIEW

2.1. Floriculture industry in the world

The floriculture industry is a significant sub-sector of global agriculture that encompasses the production, marketing, and distribution of cut flowers, potted plants, foliage, bedding plants, and other ornamentals. Beyond its aesthetic value, floriculture has evolved into a commercial enterprise that contributes substantially to global trade, employment generation, and rural livelihoods. Historically, the modern floriculture industry traces its roots to the late nineteenth century in the United Kingdom, where flower cultivation was predominantly carried out on large estates for elite consumption and decorative purposes (Nahe *et al*., 2024). Over time, what began as a luxury-oriented activity transformed into a highly industrialized and globalized business, driven by consumer demand, advancements in technology, and integration into international trade networks. Globally, the valuation of the floriculture industry stood at approximately USD 50.04 billion in 2022, with projections indicating steady growth to nearly USD 58.03 billion by 2028 (Trend Economy, 2022). This growth is fueled by several factors, including the increasing consumer preference for aesthetically appealing indoor and outdoor spaces, the growing role of flowers in social and cultural events such as weddings, funerals, and festivals, and the rising demand for environmentally friendly decorative plants in urban areas. Furthermore, technological innovations in greenhouse systems, hydroponics, climate control, and cold-chain logistics have enabled producers to meet year-round demand while minimizing post-harvest losses (Kenya Flower Council, 2019).

The global trade structure of floriculture reveals cut flowers as the dominant product category, accounting for nearly half of worldwide trade. Bedding plants, potted ornamentals, and cut foliage follow an important segment that serve both domestic and international markets (Agricultural Marketing Resource Center, n.d.). The industry is characterized by highly concentrated trade flows. For instance, the United States, Germany, the Netherlands, the United Kingdom, Russia, and France constitute the top importers of cut flowers, collectively accounting for over 65 % of global demand (Trend Economy, 2022). Notably, the Netherlands remains a global hub for flower auctions and distribution, with the Aalsmeer flower auction handling nearly 40 % of worldwide cut flower trade. Through re-exports, Dutch traders supply more than 70 % of the flowers sold across Europe, illustrating the country's central role in global value chains (Berg, 2013). Roses (*Rosa* spp.), belonging to the Rosaceae family, are the most cultivated ornamental based on their beautiful flowers (Fougère-Daneza *et al*.,2015;

Raymond *et al*., 2018). They rank among the top five ornamental plants worldwide (National Agriculture Statistics Service, 2020). In the United States, the sale of roses in 2019 generated USD 200 million from the production of 39 million plants by 1729 operations (Li *et a*l., 2019). A key trend shaping the industry in the twenty-first century is the increasing emphasis on sustainability. Consumers, particularly in Europe and North America, are becoming more conscious of environmental and social issues linked to flower production, such as pesticide use, water consumption, and workers' welfare (Van der Velden & Smit, 2019). As a result, certification schemes such as Fairtrade, Rainforest Alliance, and the Floriculture Sustainability Initiative (FSI) are gaining momentum, helping producers meet the demands of ethically and environmentally sensitive markets (Dolan, 2012). Cold-chain advancements and improved logistics have further facilitated the integration of producers in Africa, Latin America, and Asia into international markets, ensuring that flowers can be transported across continents while maintaining freshness and quality (Knuth *et al*., 2025).

In summary, the floriculture industry has transformed from a niche luxury pursuit into a dynamic, globalized agricultural enterprise. Its resilience and growth potential are anchored in technological innovation, consumer preferences for beauty, symbolism, and the ability of producers to adapt to sustainability standards and logistical demands. The sector's continued expansion is expected not only to enhance international trade but also provide socio-economic benefits in producing regions across the Global South.

2.1.1. Floriculture industry in Africa

Africa has increasingly positioned itself as one of the most dynamic hubs for global floriculture due to its favorable agro-climatic conditions, relatively low labor costs, and growing investments in infrastructure and logistics. The continent's diverse geography provides unique production advantages. High-altitude areas with cool temperatures, such as those found in Kenya's Rift Valley or Ethiopia's central highlands, are especially well-suited for cultivating high-quality roses and other premium cut flowers (Yeshiwas & Workie, 2018). Moreover, Africa's geographical proximity to Europe and the Middle East offers a logistical advantage, allowing flowers to be transported quickly and efficiently to major consumer markets. Kenya and Ethiopia dominate the African floriculture landscape, jointly contributing to more than 85 % of the continent's flower exports.

Kenya is widely regarded as the leader in African floriculture, earning the title of the "flower garden of Europe." The country accounts for the largest share of Africa's flower exports, with the industry valued at approximately USD 1.09 billion in 2023 (Mordor

Intelligence, 2024). The production of flowers is majorly concentrated around Lake Naivasha, Athi River, Thika, Nakuru, and Eldoret region that provide favorable climatic conditions including high altitude, moderate temperatures, and volcanic soils (AFDB, 2013). The sector is not only a major foreign exchange earner but also a key source of employment directly employing over 150,000 workers on farms and indirectly supporting nearly 2 million livelihoods (Central Bank of Kenya, 2021). Kenya's floriculture is dominated by roses, which make up over 70 % of exports, followed by carnations, alstroemeria, lilies, and summer flowers (Horticultural Crops Directorate, 2022). The main export destinations includes Netherlands, UK, Germany, and other EU markets. Despite its success, the sector faces challenges such as rising airfreight costs, water stress around Lake Naivasha, and compliance with increasingly strict phytosanitary standards. To maintain competitiveness, many farms have adopted certification schemes like Fairtrade, Rainforest alliance and MPS, which addresses sustainability and ethical trade (Hawkins *et al.*, 2019).

Ethiopia has emerged as the second-largest exporter of flowers in Africa, benefiting from its highland regions near Addis Ababa, which offer ideal growing conditions. The sector produces more than 130,000 tons of flowers annually, with roses as the dominant crop (Mebrat *et al.*, 2022). Government incentives, including tax holidays, land allocation, and infrastructural investment, have played a critical role in attracting foreign direct investment (FDI) into the industry. Dutch, Indian, and Israeli investors have established large-scale farms, creating thousands of jobs (Yeshiwas & Workie, 2018). The Ethiopian government's policy framework has made floriculture one of the fastest growing sectors, contributing significantly to foreign exchange earnings. However, concerns have been raised about labor rights, pesticide use, and environmental degradation around Lake Ziway and other production zones (Mebrat, 2022). Ethiopia's competitiveness is also hampered by logistical challenges, as the country relies heavily on airfreight through Bole International Airport, which often faces congestion. Nevertheless, Ethiopia's floriculture continues to grow steadily, with sustainability and compliance becoming key areas of focus for exporters (Yeshiwas & Workie, 2018).

Tanzania is relatively smaller but growing player in African floriculture. The sector is concentrated in the northern regions of Arusha, Kilimanjaro, and Tanga, where high altitudes and fertile soils provide favorable conditions for growing roses, carnations, and other ornamentals (Yeshiwas & Workie, 2018). Government support through favorable policies and incentives has encouraged both domestic and foreign investors to participate in the industry. Although Tanzania's export volumes remain much lower than Kenya and Ethiopia, the industry is expanding rapidly, targeting both European and Middle Eastern markets. Some of the major

challenges faced by the Tanzanian floriculture industry includes; limited airfreight capacity, high production costs and infrastructural limitations. However, Tanzania has a significant potential for growth if investments in logistics and certification schemes are scaled up to meet international standards.

Uganda's floriculture industry is centered near Lake Victoria, particularly in areas close to Entebbe and Kampala. The sector benefits from fertile soils, reliable rainfall, and favorable temperatures that allow year-round production (Langan, 2011). Roses and chrysanthemum cuttings are the dominant exports, with the majority destined for Netherlands and other European countries. Uganda's industry is smaller in scale as compared to Kenya and Ethiopia, but it plays an important role in rural employment and export diversification. Some of the constraints includes; high freight costs, limited access to financing, and compliance with strict EU market standards. However, the government of Uganda has identified floriculture as a strategic export subsector and is working to improve their infrastructure and access to markets (Yeshiwas & Workie, 2018).

South Africa has a long tradition of ornamental horticulture that primarily serves domestic and regional markets, though exports to Europe and Asia are also significant. Unlike Kenya and Ethiopia, South Africa's sector is more diversified, producing proteas, roses, chrysanthemums, and other indigenous flowers that enjoy niche demand abroad (Coetzee & Hoffman, 2018). The country's well developed infrastructure, including ports and airports, provides an advantage for export logistics. However, production costs are higher than in East Africa, largely due to higher wages and input costs. South Africa's floriculture industry is also increasingly shaped by sustainability standards and certification requirements, reflecting strong consumer awareness in its target markets.

Despite these country specific differences, several common themes emerge across African floriculture. Foremost among these, is the challenge of high logistics and freight costs. Since flowers are highly perishable commodities, they rely heavily on airfreight to reach distant consumer markets in Europe and beyond. In Kenya and Ethiopia, for instance, freight costs account for a substantial proportion of total production expenses, reducing competitiveness against Latin American producers supplying the United States. Water scarcity is another shared challenge, particularly acute in regions such as Kenya's Lake Naivasha and Ethiopia's Lake Ziway, where large-scale irrigation competes with domestic and ecological water needs (Njuguna, 2010). Environmental concerns have grown around these hotspots, drawing attention from local communities and international advocacy groups. Furthermore, the African floriculture sector is increasingly shaped by sustainability and social responsibility imperatives.

European consumers are more aware of the social and environmental costs of production, and certification schemes such as Fairtrade, Rainforest Alliance, and MPS have become essential for market access. These certifications promote safe working conditions, gender equity, reduced pesticide use, and environmental conservation (Mebrat, 2022). While larger farms have been able to comply with certification requirements, small scale producers often struggle due to the financial burden of audits and infrastructure improvements. This creates a structural imbalance that favors larger, foreign owned farms, raising questions about inclusivity and equitable development (Hawkins *et al*., 2019).

Nevertheless, floriculture continues to play a vital role in social development across Africa. The industry provides employment opportunities for women, who constitute the majority of the workforce. Employment in flower farms not only generates income but also contributes to women's empowerment, education of children, and poverty reduction in rural areas (Riungu & Kariuki, 2019). In Ethiopia, for instance, the expansion of floriculture has created tens of thousands of jobs for women in rural communities, contributing to both economic participation and social transformation. Similarly, in Kenya, flower farming has stimulated the growth of secondary industries, including packaging, transport and housing, creating broader multiplier effects for local economies. Looking ahead, Africa's comparative advantages in floriculture remain strong. Year round production, favorable climatic conditions, and geographical proximity to Europe are enduring strengths that will continue to anchor the continent's competitiveness (Cental Bank of Kenya, 2021). However, addressing systemic challenges such as logistics inefficiencies, environmental sustainability, and compliance with international standards will be critical for long term growth. Expanding research into integrated pest management (IPM), water efficient irrigation and disease resistant varieties will also be key to ensuring the resilience of African floriculture against climate change and emerging plant health threats.(Kenya Flower Council, 2021)

To conclude, Africa's floriculture industry reflects both significant opportunities and notable challenges. Nations like Kenya and Ethiopia showcase how floriculture can drive rural economic transformation while elevating the continent's role as a key player in global flower production. Emerging players such as Tanzania and Uganda highlight the continent's vast untapped potential, while South Africa provides valuable insights into diversification and niche marketing. With sustained investment, enhanced sustainability practices, and deeper integration into global markets, African floriculture is well positioned to play an even more influential role in the future of the global flower trade.

Despite the remarkable growth of the sector, several obstacles continue to hinder its full potential. Rising freight costs significantly reduce profit margins, particularly for countries that depend heavily on air transport to reach distant markets in Europe, Asia, and North America. Water scarcity, worsened by climate change and competition with other agricultural and domestic needs, adds further pressure on production sustainability. In addition, fluctuating energy costs increase the expenses of running greenhouses, irrigation systems, and cold storage facilities, thereby affecting overall competitiveness. Furthermore, stringent phytosanitary regulations imposed by the European Union Africa's largest flower market demand high levels of compliance, requiring producers to invest in costly monitoring, certification, and integrated pest management systems. Collectively, these challenges underscore the vulnerability of the African flower industry and highlight the urgent need for innovation, diversification of markets, and stronger policy support to ensure long-term resilience (Kenya Flower Council, 2021). There is worldwide concern about important social issues, such as how labour is treated, gender equity, and pesticide safety (Van der *et al.*, 2012). Nevertheless, Africa retains a strong competitive edge in floriculture. Its ability to support year round production, coupled with its geographic proximity to European markets and the availability of skilled labour, positions the continent as an attractive and sustainable hub for long term industry growth (Padmini & Kodagoda, 2017).

2.1.2. Flower industry in Kenya

Kenya, like Ecuador and Colombia, is blessed with favourable geographical and climatic conditions that make it favourable to produce flowers in large quantities (Adeola *et al.*, 2018). The equatorial positioning of the country ensures yearly day and night lengths are almost equal. Therefore, the growing conditions are stable and predictable. Also, the elevated altitudes of places like Naivasha and the Mount Kenya region result in cooler temperatures, which favour cut flower quality because the flowers will develop more vibrant colours, strong stems, and have a longer vase life (Adeola *et al.*, 2018). Good volcanic soils, steady supply of fresh water resources, and good hours of sunshine also make Kenya flower industry competitive. Most large commercial flower farms are located within 100 kilometres of Nairobi, the country's central hub for logistics and international exports. These favourable natural conditions, combined with strong infrastructural support, enable consistent year-round production and supply of cut flowers (The Flower Hub, 2025).

The national economy of Kenya relies heavily on agriculture and horticulture has developed into a key sector for foreign exchange earnings.

The flower industry in Kenya has evolved beyond being a purely agricultural activity, it now stands as a critical pillar of the national economy. It generates significant foreign exchange earnings, contributes substantially to GDP and supports hundreds of thousands of livelihoods both directly and indirectly. Beyond employment, the sector stimulates growth in related industries such as transport, packaging, cold chain logistics, and agrochemical supply. Its role in empowering rural communities, especially women who form a large proportion of the workforce, further underscores its importance. As such, floriculture in Kenya is not only a driver of agricultural development but also a catalyst for broader socio economic transformation (Mwase, 2015).

It has an impact on foreign exchange earnings, the creation of jobs, rural development, and its contribution to the infrastructure and service sectors. Thus, it is a dominant component of Kenya's horticultural subsector and plays a direct role in shaping the livelihoods of hundreds of thousands of people along the value chain (Bermudez & Mumbi, 2024). The horticultural subsector generates nearly US $1 billion annually, which places it within the top three subsector of foreign exchange earnings alongside tea and tourism. Additionally, in 2023, the horticulture subsector was roughly 33 % of the national Gross Domestic Product (GDP), while flower exports were roughly 1.25 % of the GDP and the total export revenues were equivalent to billions of Kenyan shillings in annual inflows (Bermudez & Mumbi, 2024).

This highlights the scale and strategic significance of the flower industry to Kenya's economy, rural development, and livelihoods. Although it may seem modest when expressed as a percentage of the overall economy, the sector generates inflows worth billions of Kenyan shillings each year. Export revenues from flowers also play a vital role in providing the country with much needed foreign currency reserves, which help stabilize the exchange rate and finance essential imports such as fuel, machinery, and pharmaceuticals (Bermudez & Mumbi, 2024). The flower industry has steadily grown into one of the most competitive players in the global floral market, with Kenyan flowers highly regarded for their superior quality, affordable pricing, and dependable year round supply (Mordor Intelligence, 2024).

Flower farms in Kenya are spread across different regions, but they are strategically concentrated in areas with the most favorable production conditions. The majority of flowers grown for export are cultivated around Lake Naivasha, where approximately 1,200 hectares are under cultivation. Of this area, about 70 % (roughly 840 hectares) is devoted to rose production, which remains the most dominant and commercially valuable crop. Other key regions with significant flower farming activities include the areas around Mount Kenya, Athi

River, Kitale, Thika, Nakuru, Kericho, Kiambu, Nyandarua, Uasin Gishu, Trans Nzoia, and parts of Eastern Kenya. (Kenya Flower Market Update, 2023).

The high concentration of farms in these regions has created employment opportunities for thousands of people, particularly women, and has also stimulated the growth of supporting industries such as packaging, cold chain logistics, agrochemical supply, and air freight services.

Roses, carnations, and alstroemeria are the primary cut flowers that are grown in Kenya and they control the volumes of exports. However, diversification has also contributed to increasing the resilience of the industry and its competitiveness. The varieties of other flowers, including gypsophila, lilies, eryngiums, arabicum, hypericum, statice, and a broad range of summer flowers are also grown with the niche and seasonal market demand (Kenya Flower Council, 2021). This diversity will help Kenyan producers provide diversified products to meet the diverse consumer preferences in the international markets, especially when the seasons are at their highest point, like the Valentine Day, Mother's Day and Christmas.

Most of the flowers from Kenya are exported to Europe where Netherlands is the entry and distribution point for these flowers. Tyce, (2020) indicates, in his research, that The Netherlands has a good auction system for flowers. Key markets include the UK, Germany and France that take up the majority of Kenyan flower imports. Although Kenya still enjoys a stable demand from traditional flower markets in Europe, new markets are coming up. Kenyan flowers are increasingly in demand in the United States, Japan, Dubai and Russia. The country is set to diversify its exports away from dependence on the EU ((Tyce, 2020).

The flower business in Kenya is successful it had their problems still remaining. Due to the high perishability of flowers, they are shipped via air which makes them expensive. The high freight charges are constantly reducing profit margins. Also, Kenya's taxes and rules sometimes make it hard for exporters to compete (Thursd, 2023). Due to the stringent plant health and environmental regulations of the European Union, there has been continuous investment in integrated pest management, chemical residue monitoring and sustainable agriculture (Kenya Flower Council, 2021). The continuing threats posed by climate change, water shortages and increasing energy cost need innovative adaptation strategies. Kenya's flower industry is global competitive. It has some natural advantages. Secondly, it enjoys access to expanding international markets. Thirdly, its production system is becoming increasingly professionalised. The sector is expected to retain its significance as one of Kenya's greatest pillars of the economy as well as a leading supplier of flowers in the world with continuous investment in sustainability, technological innovation, and market diversification (Mwase, 2015).

2.1.3. Roses production constraints

Although rose production in Kenya is highly profitable and globally competitive, the industry faces multiple constraints that hinder its sustainability, productivity, and long term profitability. These constraints can be broadly categorized into biotic factors (pests and diseases) and abiotic factors (climatic and economic stresses). Together, they significantly reduce yields, compromise flower quality, increases production costs, and threaten Kenya's reputation as a leading supplier of premium flowers in international markets (Muñoz *et al.,* 2021).

Powdery mildew, downy mildew, black spot, crown gall, botrytis blight and rose mosaic virus complex are the primary diseases that affect roses (Jeniffer, 2017). The disease affects multiple stages of the crop production cycle, ranging from field cultivation to post harvest handling and processing. Its impact is not limited to reducing plant vigor and yield in the field but also extends to compromising the quality, appearance, and shelf life of harvested flowers during storage and transportation. Consequently, it poses one of the most pressing challenges for producers, as it directly threatens both productivity and the competitiveness of Kenya's floriculture industry in international markets. (Muñoz *et al.,* 2021).

One of the most devastating foliar diseases associated with rose is Powdery Mildew of roses *(Sphaerotheca pannosa var. rosea),* which can thrive in cool nights and warm days in high altitude areas of Kenya making such environmental favourable to the pathogen. (Sharon, 2023). The disease is capable of thriving under moderate humidity conditions without requiring free water on the leaf surface, making it a persistent threat in both open field and greenhouse production systems. Its symptoms include short, deformed new shoots covered with a whitish-gray fungal growth. Initial signs typically appear on older leaves as irregular, slightly raised dark green to reddish blisters, which are later covered with a powdery layer of fungal spores. As the infection progresses, the affected leaves turn reddish, purple, then yellow and eventually drop prematurely (Alan, 2021).

Extensive infestations cause not only loss of the photosynthetic ability, but also diminished quality of the flowers and retarded growth of the plants, directly. Compared to powdery mildew, downy mildew *(Sphaeroheca pannosa)* cannot proliferate without high humidity and wet and cloudy weather. This illness is especially catastrophic in green houses that lack proper ventilation or when it is raining heavily in the open farms (Sharon, 2023). Infected leaves get purple to reddish angular lesions which can be seen with ease on the upper leaf surface, and sporulation is greyish on the underside.

It has the potential to defoliate causing annual losses in the production of 10 to 12 % (Salgado-Salazar *et al.,* 2018). Besides lowering the flower production, downy mildew compromises the plants making them vulnerable to secondary infections and other environmental stress factors. Black spot *(Diplocarpon rosae)* is one of the most prevalent fungal infections of rose in the world including Kenya. Warm and wet weather prefer it especially in the rainy season. The symptoms are spotted in circular forms with fringed edges on leaf leaves, and usually with yellow halo. Leaves change their color to yellow and fall earlier than normal as the infection envelops them, particularly in more vulnerable cultivars (Sharon, 2023). Extreme defoliation causes plant weakness and decreases flower bud development, worsens flower size and flowering, and increases the susceptibility of plants to drought and cold stress. Prolonged infections also reduce the productive life of rose bushes, thus amplifying the cost of production to the farmers (Salgado-Salazar *et al.,* 2018).

Agrobacterium tumefasciens (syn: *Rhizobium radiobacter*) is a soil borne pathogen that poses a threat to the yield and quality of roses (Li *et al*., 2019). *Agrobacterium tumefasciens* can survive in the soil, the rhizosphere of many plants, and other and other rnatural environments (Barton *et al.,* 2018; Pulawaska, 2010). Even outside nursery conditions, in green houses, crown gall has caused a 60 % decrease in rose cut flower production in southern France, where intensive soilless cultivation conditions were always employed regulary (Poncet *et al*., 1996). It can persist in symptomless rose plants and it is a major cause for the spread of crown gall diseases, causing in huge economic losses (Marti *et al*., 1990). The pathogen has been extensively studied by researchers, as it is a significant factor in endangering the health of roses, leading to poor rooting, a decrease in number of shoot formations and the yield loss of cut flowers, affecting their commercial production (Kawaguchi, 2008 *et al;* Poncet, 1996).

Crown gall enters the plants through wounds commonly as a result of pruning or mechanical damage. The illness causes the development of tumor like galls around the roots and stems, the galls that interfere with the vascular system thus reduces the absorption of water and nutrients. Plants that are infected frequently look stunted, low level of vigor and no flower production (Oksel *et al.,* 2024). In severe cases, the disease causes galling at the crown of the plant, a condition that disrupts the normal flow of nutrients and water within the vascular system. This damage often leads to stunted growth, wilting, and eventual plant death. Such outcomes translate into significant economic losses for producers, as entire blocks of crops may be rendered unproductive. Beyond direct yield reduction, growers also face increased costs in crop management, replanting, and disease control. At a broader scale, widespread crown gall infestations undermine the profitability and sustainability of the floriculture

industry, with ripple effects on employment, export earnings, and rural livelihoods. The bacteria have the ability to span years in the soil and treating the crown gall involves a high level of sanitation and keen propagation practice (Collins, 2001).

One of the most destructive after harvest diseases of the rose is known as Botrytis blight *(Botrytis cinerea)*, also commonly known as grey mold. The disease is very latent during harvest and the symptoms are revealed during transportation and storage in the humid conditions (Kariuki, 2015). The first symptoms of infections manifest themselves in tiny spots or blisters on petals, but soon develop into huge necrotic spots, causing petal collapse and decomposition (Bika *et al.,* 2020). Since roses are very perishable, and they are air shipped to far markets, any infection of botrytis during transportation to the markets may lead to huge losses of rejections at the receiving markets which translates to direct losses incurred by the exporters (Sharon, 2023). The prevention of botrytis is based on the tight control of the cold-chain, ventilation, and handling of fruits to avoid bruising leading to the creation of entry points by the pathogen (Bika *et al.,* 2020).

Rose mosaic is caused by a complex of several viruses including, Prunus Necrotic Ringspot Virus (PNRSV) and Apple Mosaic Virus (AMV). The most common way of these viruses is via the use of infected planting material, and therefore, the management of this aspect on nursery level is of paramount importance (Alan, 2021). Showing a variety of symptoms based on cultivar, season, and environmental conditions, the common symptoms of the condition are wavy or zigzag chlorotic lines, mottled patterns, clearing of veins, or ring spots on leaves (Alan, 2021). Infected plants will be characterized by slowed growth of plants, small flowers, few flowers and cold intolerance. In other situations, the infected plants can be asymptomatic yet act as points of inoculum. Since there is no cure to viral infections, the best method of management is by prevention using certified disease-free planting material (Vazquez-Iglesias *et al.,* 2020).

In addition to diseases, rose production in Kenya faces several other challenges, particularly the heavy pressure from insect pests such as mites, thrips, and aphids. These pests not only inflict direct damage on leaves, buds, and flowers resulting in reduced aesthetic quality and market value but also act as vectors for viral pathogens that further compromise plant health. Their rapid multiplication under favorable conditions makes them difficult to manage, often necessitating frequent pesticide applications, which in turn increase production costs and raise concerns about resistance development, environmental contamination, and compliance with strict international residue standards. If left uncontrolled, pest infestations can cause significant yield losses, disrupt continuous harvesting schedules, and ultimately reduce the

competitiveness of Kenyan roses in export markets. (Vazquez-Iglesias *et al.*, 2020). Production is further complicated by abiotic stresses such as variation temperatures, water shortages and salinity of soils. The increase in prices of agrochemicals, labor, and energy contributes to the increase and makes the farmers less profitable. In addition, the phytosanitary requirements in major export markets are very strict necessitating constant surveillance and hence disease and pest control processes are not only very expensive but also labor-intensive (Kenya Flower Council, 2021).

Indeed, Kenya is one of the world's leading producers and exporters of roses. However, a combination of diseases and production challenges significantly limits the industry. To overcome these constraints, integrated management such as resistant varieties, use of certified planting material, better cultural practices, biological control and wise use of fungicides are needed. There is also a need to strengthen research and extension services and to train farmers so as to facilitate sustainable production suitable for ensuring Kenya's competitiveness (Vazquez-Iglesias *et al.,* 2020).

2.1.4. Integrated disease and pest management for roses

Based on the business model, roses are compromised by several pathogens and pests that interact with production practices and the post harvest chain (Daughtrey & Buitenhuis, 2020). Because no one tactic will eliminate risk, using integrated pest and disease management (IPDM / IPM) is the most cost effective and sustainable way forward. IPM involves the use of a combination of prevention, cultural controls, biology, chemistry, monitoring and post-harvest hygiene to manage pests. The site selection process takes into account that the soil conditions for planting should be well drained. Ventilation and heat management in tunnels prevent the downy and powdery mildews from occurring. By removing and destroying infected plant debris, pruning out diseased shoots and avoid leaving cuttings to rot, sanitizing tools and benches, the spread of crown gall and viruses will be reduced (Yan *et al.,* 2021). Management is done with the use drip or subsurface irrigation where possible avoiding overhead irrigation late in the day to keep foliage dry. This reduces downy mildew, powdery mildew and botrytis risks. Crop hygiene can also be achieved by keeping appropriate plant spacing and pruning to allow light penetration and air movement, along with immediate removal of symptomatic plants or sections (Arya, 2019). Healthy fertilization by avoiding too much nitrogen and regular water supply can help improve the health and resistance of plants against diseases. Using tissue-culture plants as well as certified disease free nursery stock helps in reducing the introduction of viruses such as rose mosaic complex and bacterial pathogens (Daughtrey & Buitenhuis,

2020). Nurseries should also apply indexing and hot water or thermotherapy treatments when appropriate, and focus on cultivars that have some known tolerance or resistance to local disease pressures when planting. They should be adopted in conjuction with other intergrated pest management tactics since no one tactic gives total control (Arya, 2019).

Consistent trapping for early signs of thrips which will act as vector viruses by using yellow and blue sticky traps and simple disease forecasting on humidity and temperature will allow rose farmers to spray on time instead of spraying when roses have already been infected. Proper keeping of records of disease incidence and spray history to show up resistance development (Dudutech, 2022). Many farms employ bacteria and fungi that fight other harmful fungi and bacteria. The antagonistic fungi and bacteria applied in most of the farms include *Trichoderma* spp. and *Bacillus subtilis* formulations that have the ability to suppress soil borne pathogens and some foliar diseases (Evalon *et al.*, 2024).

Natural enemy conservation such as predatory mites and parasitoids is also associated with keeping insect vectors and populations of pests small, which reduces the risk of transmitting the virus (Julliete *et al.,* 2020). Alternatives that can be applied are biopesticides and botanical extracts, which are softer to use and can meet export maximum residue level (MRL), requirements (Ruparo, 2011). Fungicides should be applied only when monitoring justifies treatment and resistance management guidelines are followed. It is important to rotate modes of action, use of single site fungicides or nutrients should not occur alone and respect Post Harvest Interval (PHI). Most of the chemicals used worldwide for roses are multi site protectants and single site systemic fungicides. Choice of fungicides must also consider local resistance patterns (FAO, 2012). Training of spray operators to calibrate the equipment and adjust the droplet size and spray coverage, as well as use PPE (Pesonal Protective Equipment) to get the maximum benefit and safety from the application (Loise *et al.,* 2015).

It is advisable to reduce mechanical injury during harvesting and handling to reduce entry points for *Botrytis* and *Agrobacterium*. Ullah *et al*., (2024) recommend cooling flowers immediately (pre-cooling / hydrocooling) and keeping an unbroken cold chain to slow the development of pathogens. It is compulsory that sanitary procedures through innovation like short dips or aqueous treatment meet destination MRLs and phytosanitary regulations. They must be practiced on packing line, cold room and transport containers. Following worldwide guidelines like GLOBAL G.A.P Guidelines and systems like MPS and KFC offers traceability which reduces rejection risks at destination markets.

Many certifications require IPM practices, residue monitoring, and worker training to be documented. It is essential to have extension services, grower cooperatives and public /

private research partnerships to disseminate best practices, diagnostics and updated spray calendars (Kenya Flower Council, 2023). Fund creations of local disease resistant varieties bred and screened for appropriate Kenyan environment as well as the speedy development of molecular diagnostics for early detection, particularly for viruses. Research on affordable forecasting tools and locally effective biological control agents offer high returns to the sector (Thomas & David, 2014).

In the long run, it is cheaper to put preventive measures in place such as, proper sanitation, monitoring and scouting for pests and diseases to safeguard market access. On the other hand, controls that are reactive outbreak after outbreak of pests or diseases tend to be costly (Borbaruah, 2023). In order for small and medium growers to adopt prevention based approaches, certification and technical support is offered as an incentive. Often, an integrated, locally adapted IPM program; combining clean planting material, good cultural practices, monitoring, biologicals and conservative, well managed chemical use is the most effective path to reducing disease constraints in Kenyan rose production and maintaining access to high value export markets (Kenya Flower Council, 2021).

2.1.5. Post-harvest handling and cold chain management

Kenya's flower industry's success depends on the efficiency of post-harvest handling and the cold chain, not just field production. The quality of flowers, particularly roses, deteriorates after harvesting as they are highly perishable commodities (Musyoki & Kihara, 2025). The post harvest stage includes harvesting methods, packaging, storing, transporting, and distribution. The above processes ensure that Kenyan flowers reach overseas markets fresh, attractive and long lasting. If post harvest management is poor, it can lead to severe monetary losses through reduced vase life, petal browning, *Botrytis* infection, and even rejection in destination markets (The Flower Hub, 2025).

The harvest is usually carried out at dawn or late in the evening when the temperature of the field is low. This is done to reduce respiration rate and wilting. There is a process for harvesting roses that is specific to the flower types. Roses are harvested when they are at the precise stage of maturity. In different stages a rose may be harvested for different markets (Musyoki & Kihara, 2025). For example, the tight bud stage is favoured for the far away market as in the European Union. Moreover, for the regional market, the open bloom stage or the mother bloom stage is used. Employees are trained to use sterilized, sharp secateurs to avoid bruising and tearing of stems. Bruises and tears create entry points for pathogens like *Botrytis cinerea*, which can hamper quality. Flowers should be placed in sterilized buckets filled with

clean water and preservative immediately after harvest. This prevents growth of microbes and increases vase life. (Dhiman *et al.,* 2021).

After harvesting, flowers are immediately sent to grading and packing houses close to the fields. Flowers are classified according to their International Grading standards based on stem length, bud and flower size, color and quality. Grading should be consistent, as buyers of variable systems like an auction system and supermarkets need uniformity (Chandel *et al.,* 2023). Flowers that are damaged, undersized, or diseased are rejected to maintain consignment overall quality. Pre-cooling is an important postharvest handling step. Once harvested, flowers are cooled quickly to decrease their field heat using forced air cooling systems to bring their temperature down to approximately 0 to 2 ° C within a few hours. As a result, metabolic activity is slowed, respiration rates are reduced and fungal pathogens do not develop. From here on, a strict temperature control should be maintained since a break in the cold chain, even for a short time, can cause irreversible quality loss (Mwase, 2015).

Kenya's flower export sector has developed however, the country is facing cold chain challenges. The reliability of cold storage is affected by high air freight charges, insufficient refrigerated cargo capacity and unreliable power supply in part of the country (Musyoki & Kihara, 2025). Moreover, congestion at airports cause delays in the shipment. There is also an increase in demand around the world for sustainability, which means carbon footprints in transport must also be reduced (Musyoki & Kihara, 2025).

In Kenya, flower farms are mostly to be found in the rural areas. For instance, Naivasha, Thika, Nakuru, and the Mount Kenya region. The establishment of large commercial farms has spurred the growth of nearby towns and development of infrastructure like houses, schools, hospitals and local investments (Adeola *et al.,* 2018). Numerous farms are involved in CSR (Cooperate Social Responsibility) initiatives, including setting up a health clinic, installing clean water supply systems, and providing education for children. The programs raise quality of life, create jobs, and strengthen the socio economic fabric of rural communities. Thus, the flower industry in Kenya not only forms part of agricultural exports but also assists in rural development and community welfare (Opondo, 2006). Flower production is linked with many other engineering sectors. Suppliers of fertilizers, pesticides, seeds, greenhouse materials, and irrigation equipment have benefited by the growth of the flower industry (Sharma, 2024). The logistics and transport sectors are thriving due to their dependence on refrigerated trucks, air freight as well as packaging material (The Flower Hub, 2025).

Nairobi's Jomo Kenyatta International Airport (JKIA) has emerged as one of the busiest cargo airports in Africa due to horticultural exports, comprising of up to 50 % of flower

airfreight. The multiplier effects from these links are very expansive and go to other businesses dealing with all cargo movement and related services benefiting from exports thus benefiting the economy courtesy of the horticulture sector of Kenya. The Kenyan flower industry contributes significantly to the Government's fiscal revenues through taxes, export levies and license fees. Money generated through taxes helps in building and providing necessary public services. According to experts, exporters have seen some reduction in their profits due to high taxes and rising freight and operational costs (Tyce, 2020).

With dynamic growth over the years, the Kenyan flower industry has been a key component in boosting economic growth. However, it continues to remain exposed to serious shocks. Operations can face major difficulties due to global recessions, pandemics, changing freight charges, and stringent regulations in the European Union. For example, in 2020 due to COVID-19 pandemic, flowers international demand fell significantly thereby causing huge financial losses and temporary layoffs on farms and in related sectors (Chebet, 2021). Despite these interruptions, the business has shown considerable resistance by taking advantage of new markets in the Middle East and Asia, more efficient production, post harvest systems and technology across supply chains (Sigion *et al.,* 2024).

Kenya's flower business plays a significant role in the economy. A significant amount of employment is created by this industry. This also provides necessary foreign exchange and stimulates rural development. It also creates linkages with many other industries (Agriculture Institute, 2025). The sector's social and gender impacts only add to its credentials not just as an export earner, but also as an agent of change in the Kenyan communities. To ensure the industry's sustainability in the long run, it must find a balance between profit, worker welfare, fair trade practices and the environment (Mwase, 2015).

The Kenyan government considers the flower industry to be strategic because of its ability to earn foreign exchange and create rural jobs so it is well regulated. The aforementioned goals of a company along with the overall national policies of a country will affect the project. The national policies incudes land use, taxation policies, export licensing policy, labor standard policy, environmental management policy (Ngutu *et al*., 2018). Government regulators like Horticultural Crops Directorate (HCD) under Agriculture and Food Authority (AFA), ensure compliance to the standards of horticultural exports through enforcement of quality and safety requirements of the market. However, producers often worry about high taxes, permission licenses by the bureaucracy and their inconsistent enforcement which raises production costs and lowers competitiveness (Chatterjee, 2025). Kenya has remained part and parcel of regional trade agreements including the East African Community (EAC), as well as preferential trade

agreements with the European Union and other partners to maintain access to key export markets and to support sector growth.

Most of the flowers produced in Kenya are exported to the European Union (EU). Therefore, producers must comply with strict phytosanitary requirements and pesticide Maximum Residue Limits (MRLs). The European Union has pressed Kenyan manufacturers to practice integrated pest management, minimize use of harmful chemicals, and improve traceability systems to enhance environmental safety and customer health (Kenya Flower Council, 2021). Not following the rules can cause your shipments to be turned down. One can also lose out on important buyers in the process. In a similar vein, the United States and Japan are also demanding their own quality and safety standards in relation to imports, making export management more complex (Kenya Flower Council, 2023).

Many flower farms in Kenya have adopted sustainability certification schemes to improve credibility and access to markets. This features GLOBAL G.A.P., Fairtrade and MPS (Milieu Programma Sierteelt) as well as Rainforest Alliance and KFC (Kenya Flower Council), Silver and Gold. Certification schemes deal with safe pesticide management, labour right, environmental protection, and worker welfare (Hoorn, 2024). Take Fairtrade, as an example, which provides better labour and creates premium funds which can be reinvested back in the community through schools, water supply systems, and health facilities. Adherence to such schemes is more a necessity than a choice as several buyers in Europe are now insisting on sourcing from certified farms (Kenya Flower Council, 2023).

The flower industry has been critiqued over its water consumption around Lake Naivasha, which mostly houses most farms. The declining level of water in lakes affecting different species is due to irrigation system. Since the use of much agrochemical causes concern over soil degradation, water pollution, and affects non target organisms (Saina *et al.*, 2019). Many farms have started adopting drip irrigation, rainwater harvesting, and water recycling in response to the demand for fresh water. Implementation of integrated pest management (IPM), biological control agents and lesser chemicals is becoming popular to keep footprints smaller. More farms are investing in renewable energy. For example, solar-powered pumping and cooling systems cut carbon emissions (Hortifresh, 2025).

Work in the flower industry has changed rural areas though there are concerns about working conditions, salaries and gender equality. Sometimes, workers, especially women have faced challenges such as low pay, job loss, and being in contact with agrochemicals due to lack of relevant protective gears (Kaaria, 2022). To address these challenges, certified farms are required to uphold high standards of labor welfare and workplace safety. This includes

providing workers with appropriate protective clothing and equipment to safeguard them from agrochemical exposure and other occupational hazards. In addition, farms must ensure fair and timely wages, while also offering essential social benefits such as maternity and paternity leave. The right of employees to unionize is equally important, as it strengthens their collective voice and enables them to negotiate for improved working conditions. Growing awareness among consumers and buyers about ethical sourcing has made compliance with such standards not only a moral obligation but also a strategic advantage. Farms that demonstrate social responsibility and fair labor practices are increasingly seen as more credible partners in global value chains, which in turn enhances brand reputation, worker satisfaction, and overall productivity. (Opondo, 2006).

2.1.6. Global competitiveness and future prospects of Kenya's flower industry

Among the key competitive strengths is the natural and geographical advantages. Kenya has favourable climatic conditions (day to night temperature variation, sunshine, altitude), in Naivasha, Mt. Kenya and other highlands. They help produce cut flowers of great quality throughout the year (Adeola *et al.,* 2018). The existence of volcanic soils together with the availability of water in certain zones gives Kenya a head start against many competitors. There is an already established infrastructure and supply chain and well managed cold chain handling, packing houses, and export logistics. Plus, regulatory agencies and trade associations such as the Kenya Flower Council, KEPHIS (Kenya Plant Health Inspectorate Services), AFA (Agriculture and Food Authority, HCD (Horticultural Crops Directorate). Such compliance with international market standards and ensure traceability and quality (Kenya Flower Coucil 2021).

More growers in Kenya and other flower exporting countries are adopting more increasingly mandatory and voluntary certification such as GLOBAL G.A.P., Fairtrade and Rainforest Alliance, The Kenya Flower Council's Flowers and Ornamentals Sustainability Standard (FOSS). In fact, these certifications go beyond merely being labels. In fact, they practically involve sustainability practices. This includes integrated pest management, recycling systems, irrigation system, energy efficient greenhouse design, renewable energy sources, and improved worker safety and welfare (Bermudez & Ngige, 2024; Hortfresh Journal, 2024). With such credentials, competitors gain access to high value markets, such as in Europe, the UK and elsewhere. This is where environmental, social and governance (ESG) requirements are now a condition of trade agreements and buyer contracts. Certification has practically become essential for many exporters, with certified producers facing higher barriers

to entry, such as increased scrutiny and audit costs (Bermudez & Ngige, 2024). The increased attention towards sustainability suggests that market forces and regulations are steering the change in the floriculture sector. Despite inflation, rising energy prices and climate change affecting the industry, many new investors are still interested in flowers. For example, in Kenya, local and international players have begun the establishment and expansion of medium sized farms of the order of 10 to 20 hectares to meet growing global demand (Waitathu, 2025).

Innovation, partnership and market linkages are the main aims of events for the floriculture industry most notably IFTEX which has seen considerable growth in numbers, especially new growers. Many of them are investing in moderate scale production based on international sustainability and quality standards. These improvements are expanding to other areas as well. Other than the new developments, other areas like energy systems, cold-chain logistics, and other digital technologies are benefitting (Hortifresh, 2025). These improvements will make sure that exporting and other requirements are being fulfilled. Inspite of the global headwinds, the resilience of investor confidence illustrates long term potential of the sector as it shifts towards growth (FloralDaily (2023).

Kenya's cut flowers are predominantly transported to international markets by air, as the global floriculture trade continues to depend heavily on airfreight. While this mode of transport ensures speed and freshness, it remains an expensive option, and alternatives such as sea freight are still limited in both capacity and reliability for highly perishable commodities. The fragility of this system was highlighted during global crises such as the COVID-19 pandemic, when the grounding of passenger planes many of which also carry cargo, severely disrupted supply chains. With drastically reduced cargo space, freight costs more than doubled, and thousands of tonnes of flowers were lost due to the inability to reach markets in time. This underscored both the vulnerability of the sector to external shocks and the urgent need for investment in more resilient and diversified logistics solutions (Chebet, 2021). The freight cost takes away nearly 40 % of the export value, which is significant even at a time of crisis. The industry is currently looking into ocean freight and hybrid shipping system to save costs and carbon footprint. Flowers can now be transported by sea because of improved refrigerated containers and controlled atmosphere technology that can keep flowers fresh for 30 days. Though shipping by sea has its own problems like infrastructure, long lead times, and the need to ensure product quality is maintained over long distances (Njogu, 2022). The stress on freight logistics remains to be a significant bottleneck for exporters, especially smaller and medium farms which do not possess economies of scale and hence are not able to negotiate for favourable shipping rates.

Export markets, especially the European Union (EU), are now becoming more severe in their enforcement of phytosanitary regulations, pesticide residue limits and quarantine pest controls. Failure to meet the standards such as non compliance raises the cost for exporters as their consignments are rejected. Thereby suffering compliance cost and reputational loss (Wani *et al.*, 2018). The False Codling Moth *(Thaumatotibia leucotreta)* has often led to rejections of cut flowers as well as fruits and vegetables from Kenya. In response to this situation, the Kenya Plant Health Inspectorate Service (KEPHIS) has rolled out stricter measures and a "systems approach" (Kenya Flower Council, 2021). To become adopted on these systems, producers have to be certified by KEPHIS, monitor pest traps and compliant farming records. Therefore, production has to become the more complex. To keep the market accessible, these measures are a must. However, they definitely lead to increased compliance costs for producers. In addition smaller exporters may find it difficult to use advanced pest management technologies (FloralDaily, 2023). The growing customer expectations for traceability, as well as "pesticide-safe" production, add a new level of regulation. Producers have to innovate constantly and lose competitiveness if they do not meet standards.

2.1.7. Competition from other producing countries

Kenya's floriculture industry, while globally recognized as one of the leading exporters of roses, is increasingly challenged by stiff competition from both traditional and emerging players. Countries such as Ethiopia, Colombia, and Ecuador have strategically invested in infrastructure, logistics, and market diversification to capture larger shares of the international flower trade (FreshPlaza, 2025). This competition is not merely based on volume but also on cost structures, government support, product diversity, and innovation in marketing strategies. Ethiopia, for instance, has become a formidable competitor within Africa, benefitting from substantial government incentives such as tax exemptions, subsidized land and low cost energy tariffs. A key competitive advantage is the development of a sea freight corridor via Djibouti, which has drastically reduced export costs and enhanced reliability compared to Kenya's heavy reliance on air freight (Kassa *et al.*, 2025). This not only provides Ethiopian exporters with lower freight charges but also positions them as environmentally friendlier in an era where buyers are increasingly sensitive to the carbon footprint of supply chains.

Similarly, Colombia and Ecuador dominate the American market, particularly the United States, with their long standing reputation for carnations, chrysanthemums and alstroemeria flowers that are less widely produced in Kenya (Thursd, 2023). These Latin American countries have also leveraged bilateral trade agreements that reduce tariffs and ease

customs clearance, further enhancing their global reach (World Bank, 2021). Additionally, they have invested in state of the art post harvest technologies and varietal breeding programs that allow them to meet consumer preferences in terms of color diversity, fragrance, and vase life. In comparison, Kenya's reliance on roses is estimated to account for nearly 70 % of its total flower exports poses a strategic vulnerability. While roses remain highly profitable, overdependence on a single crop exposes the industry to risks from market saturation, fluctuating prices, and shifting consumer preferences (Kenya Flower Council, 2023).

To remain competitive, Kenyan producers must expand their portfolio to include summer flowers, fillers, and niche varieties that are gaining traction in emerging markets in Asia, the Middle East, and Russia (Odongo, 2023). Furthermore, Kenyan exporters face mounting pressure from stringent sustainability demands by European Union supermarkets, which increasingly prioritize suppliers certified under schemes such as GLOBAL G.A.P., Fairtrade and Rainforest Alliance ((Bermudez & Ngige, 2024). Competitor countries, especially Ethiopia, have aggressively aligned their industries with such certifications, often backed by direct government and donor funding. By contrast, Kenyan growers shoulder the costs of compliance independently, creating disparities in production costs.Ultimately, sustaining Kenya's global position requires continuous innovation, investment in new varieties, strengthening logistics competitiveness and proactive government engagement in trade negotiations. Without diversification and stronger policy support, there is a real danger that Kenya could gradually lose market share to countries offering cheaper, more diverse and sustainability certified products (Thursd, 2023).

2.1.8. Environmental and Climate Risks

Environmental and climate related challenges represent one of the most pressing threats to the future sustainability of Kenya's floriculture industry. The sector is highly dependent on stable climatic conditions, abundant water resources, and reliable energy supplies, all of which are increasingly under pressure due to climate change and environmental degradation. Rising global temperatures, unpredictable rainfall patterns, and recurrent droughts are already affecting flower yields and raising production costs (Mulwa *et al*., 2021).

One of the most significant risks is water scarcity, particularly in major production hubs such as Naivasha, Mount Kenya, and Thika. Lake Naivasha, which supplies a large share of irrigation water for rose farms, has been subject to over extraction, pollution, and declining water levels (Saina *et al.,* 2019). This has fueled conflicts with surrounding communities and raised social and political risks for the sector, as local populations compete for limited

resources. Such tensions jeopardize the social license of flower farms to operate and can provoke regulatory interventions. Climate risks extend beyond water shortages to energy costs and cooling systems. Rising fuel and electricity prices have made it increasingly expensive to operate cold rooms and maintain the unbroken cold chain necessary for export of quality flowers (Njeri *et al*., 2014). Additionally, recurrent power outages create risks of post harvest losses, while reliance on fossil fuel based energy add to the industry's carbon footprint. To adapt, farms are experimenting with solar powered irrigation and cold storage, as well as energy efficient greenhouse technologies (Hortifresh, 2025). The impacts of climate change also manifest in higher pest and disease pressures, as warmer and more humid conditions create favorable environments for pathogens like *Botrytis cinerea* and bacterial wilt. This not only increases crop losses but also raises the cost of pest control, particularly as international regulations tighten around pesticide use (Borbaruah, 2023). The combined burden of water stress, pest outbreaks, and energy costs threatens both profitability and long term viability. Beyond climate risks, the sector is hindered by structural and regulatory challenges. Exporters regularly report that bureaucratic delays in licensing, duplications of roles across government departments, and heavy taxation erode competitiveness (Njogu, 2022). These inefficiencies often increase the cost of doing business and slow down the clearance of perishable exports, further exacerbating vulnerability to climatic shocks.

Adopting sustainable adaptation strategies is therefore essential. Practices such as rainwater harvesting, recycling wastewater, precision irrigation, shade netting, and drought tolerant varieties are increasingly being promoted by research institutions and certification bodies (Hortifresh, 2025). Similarly, the integration of renewable energy systems and green technologies could mitigate the combined risks of high energy costs and carbon intensive production. Moreover, global buyers are increasingly insisting on evidence of environmental responsibility as a prerequisite for long term contracts (Kenya Flower Council, 2021). This means that Kenyan exporters must not only adapt to climate risks but also document and communicate their sustainability practices transparently. Failing to do so risks both market access and international reputation.

In summary, environmental and climate risks represent a dual challenge; they directly threaten production costs and yields while simultaneously shaping international market expectations. Unless Kenya's floriculture industry accelerates its climate adaptation and sustainability transition, it risks losing both profitability and competitiveness in an increasingly environmentally conscious global trade environment.

2.1.9. Agronomy of *Rosa hybrida*

Rosa hybrida (rose plant) is one of the most important ornamental plants in the world economically and culturally. The Rosaceae family comprises over 100 genera and some 3,000 species, including other economically important crops such as apples, pears or cherries (Wissemann & Ritz, 2005) Roses are woody perennial shrubs or climbers with erect, trailing, or climbing stems depending on the variety. The genus *Rosa* contains between 100 and 300 species, depending on whom you ask and what you read, according to research published in 2001 (Ertter, 2001). This 100 year old debate is based on the extensive hybridization and selection of cultivars. Because rose types are the leading floricultural crop in the world, they are now very commonly adapted to temperate and subtropical climates. Today, scientists are examining rose breeding and classification using various molecular markers. Botanists today are beginning to resolve some taxonomic problems related to the original source of ancestry and species identification. Some of these studies will contribute to future breeding programs that will increase genetic diversity and horticultural value (Rits *et al.,* 2005). The shoot of a rose plant has 8–15 repeating units of a leaf, prickles, an axillary bud, a node and internodes (Zarif *et al.,* 2025). Roses exhibit considerable variation in morphological traits, including stem height, bud size, petal color, and fragrance. The architecture of these traits is also quite variable, which causes difficulties in rose taxonomic classification (Ertter, 2001). The situation is further complicated by the natural hybridization of the various rose species, which makes it unclear whether a particular form is wild or cultivated (Gudin, 2000). Soil composition, temperature, and light intensity are other environmental factors that influence these morphological characteristics. That's why horticulturalists often mention a genetic environment interaction while assessing the performance of roses. Thanks to modern imaging technology and digital phenotyping, researchers are now able to precisely document this difference in morphology. Consequently, the combination of traditional taxonomy with modern computational tools is helping to redefine rose morphology (Fiasson *et al.,* 2003).

The rose’s evolutionary history dates back at least 34 million years. Fossils show that the rose originated from Asia, spreading to Europe, North America and North Africa later (Fougère-Danezan *et al.,* 2015; Ritz *et al.* 2005). Wild roses are the gene pool for the modern cultivated roses. The plant hybrid debuted in the 18^{th} and 19^{th} centuries through interspecies breeding.The crosses were designed to enhance important floral characteristics like petal colour, fragrance, vase life and disease resistance. Though the trait of interest has improved in most cultivated crops, this has come at the cost of trade cultivars (Zlesak, 2006). Such trade off may happen if one character is improved due to breeding, causing varied expression of a

neighbouring character (Anderson, 2006; Bendahmane *et al.*, 2013). For instance, enhancing the size of the bloom sometimes reduced fragrance intensity as there will never be a complete trade off. Even with these downsides, ongoing raising has resulted in thousands of cultivars suited to diverse flower markets and household gardens. Roses are at once both natural species and cultural artifacts. Consequently, roses are a living archive of plants and people and places. The testing of molecular phylogeny on roses has aided in understanding the hybridization history and the evolution of roses. In addition, roses symbolize many things in worldwide culture. That is why they are important outside of horticulture (Bendahmane *et al.*, 2013)

At the moment, roses are mainly classified in hybrid tea, floribunda, grandiflora, miniature and landscape roses. Each group has its own objective. Hybrid teas are valued for their long stems and large blooms that are useful for the cut flower industry whereas floribundas are used in landscaping due to their clustered blooms and hardiness (Debener & Linde, 2009). Crosses between hybrid teas and floribundas, grandifloras have the elegance of single blooms but the abundance of clustered flowers. Small and landscape roses are especially valued in urban gardening due to their compact growth habit and adaptability. This grouping reflects aesthetic preferences and the broader role of roses in environmental design and sustainable landscape management. In addition, different consumer markets exist for these groups, some cultivars are now global export leaders in cut flowers while others have a more local presence, are used for cultural traditions (Debener & Linde, 2009). The capability of rose breeding must overcome multiple obstacles for various agricultural and ornamental purposes.

Roses also possess significant economic and cultural value. Roses have been grown as cut flowers as far back as 500 BC. Subsequently, they have become widely known as symbols of beauty, romance, and social status (Miller, 2012). Apart from being used ornamentally, roses are also a source of valuable second metabolites. Essential oils including citronellol, geraniol and nerol in the petals are important raw materials for the international perfume industry (Scalliet *et al*., 2008). Rose hips and petals contain vitamins A, C and E. Furthermore, they are used in the food and beverage industry for teas, jams, jellies, and soups (Cinar & Colalegul, 2005). The global trade of products derived from roses significantly contributes to the agricultural economy of the world, especially in places like Bulgaria, Turkey and Kenya where they are grown on a large scale. Roses have different traditions, festivals and rituals in various cultures which highlight their cultural significance other than being sold in the market (Siddiqua, 2024). Roses serve diverse and unique functions in art, harvesting, and human health and well being. People of different cultures have popularized and used this crop in various

ways since ancient times, making it one of the most popular crop When rose petals are treated well, rose water is obtained which is used as medicine (Cuttler, 2003).

Rose propagation is primarily performed by budding or grafting where the bud of the desired variety is joined to a strong rootstock to enhance vigour, disease resistance and hardiness (Ljubojević *et al.,* 2025). There has been ever growing interest in other propagation methods such as micropropagation and cuttings to produce uniform and disease free planting material (Xia *et al,* 2006). Nevertheless, regular cultural practices, such as pruning, bending, and harvesting, often cause wounds and expose rose plants to crown gall disease caused by *Agrobacterium tumefasciens*. The use of disinfectants on tools, propagation houses and planting materials will reduce the pathogen load (Maina *et al.,* 2013). According to research, integrated pest and disease management, which includes the use of biocontrol agents and resistant cultivars, may help lessen the need for chemical pesticides. In Kenya's region of Kericho, a trial conducted in the field showed that Agrowipe (a botanic neem based extract) and Fresh Fri reduced occurrence of crown gall and there was substantial improvement in yield and flower quality compared to untreated controls (Opisa *et al.,* 2020). Crop specialists use integrated farming systems that advantage of the positive attributes of different farm technologies to their greatest benefit.

The *Rosa hybrida* would act as an interesting vehicle between biology, culture, and economics of a globalised world. This makes it one of the most studied ornamental crops worldwide. The production of the species is receiving continuous improvement by molecular breeding, tissue culture, integrated pest and disease management, among others to maintain its economic importance globally. As consumers want flowers produced as sustainably as possible, breeders and growers will be forced to innovate while not losing the aesthetic and symbolic points of roses. People love roses as they can be grown anywhere. Moreover, roses have brought human societies together across time and space (Bernhardt, 1999). In future, the incorporation of genomics and biotechnology will further expedite the enhancement of rose breeding. This will probably improve their economic viability and their overall impact on global environmental change and sustainable development (Arya. 2019).

2.1.10. Crown gall disease in roses

Crown gall disease of roses and other dicots is one of the most destructive bacterial pathogens affecting these plants and currently poses a commercial floral problem throughout the world. The abnormal growth or tumour (gall) is caused by a soil borne bacterium, *Agrobacterium tumefasciens (syn. Rhizobium radiobacter)* which affects the stem, crown and

root of the plant (Schell *et al*., 2009). They increase in size, decay, split and fall releasing the *A. tumefasciens* back into the soil which successfully re gains a new Ti-plasmid from its neighbouring pathogenic *A. tumefasciens* (Shams *et al*., 2012). The presence of tumors in these ornamental crops affects their appearance and the physiology of the ornamental crop plants. It can weaken the architecture of the plants. Such crops also reduce the productivity of field and greenhouse crops. Crown gall is a major problem of rose cultivation as it greatly diminishes plant vigor and flower quality. On cut flowers and nursery stock, crown gall causes a severe drop in market value. Hence, it is a dreaded disease in intensive rose farms (Kado, 2002; Shams *et al.,* 2012). Once established, it is very difficult to control crown gall. The bacterium has a long life in the soil. It can infect through any type of wound, which means common horticultural practice spreads it. Countries like Kenya and Ethiopia exporting roses as a leading commodity have reported crown gall outbreak causing losses of millions of dollars per year thus, reducing competitiveness in the global cut flower industry (Maina *et al.*, 2013).

Agrobacterium tumefasciens is unique among plant pathogens because the bacteria can transfer genes to plants horizontally, which is an unusual mechanism in plant disease biology. In particular, a segment of DNA (T-DNA) from the tumor inducing (Ti) plasmid of the bacterium is transferred into the genome of the host plant, a classic model of plant microbe interaction. Once integrated into the plant DNA, the T-DNA induces uncontrolled cell division and differentiation. These processes are responsible for gall formation, which can be highly variable depending on the age of the plant, cultivar, and environmental conditions (Escobar & Dandekar, 2003; Gelvin, 2003). Galls initially appear as small, soft, and whitish outgrowths, but over time, they develop into dark, woody, and irregular tumor like formations that hinder the regular flow of water and nutrients through the vascular system (Collns, 2001). When the natural physiological processes of a plant or tree tissues are disrupted, the plant's vigor may be reduced. The disruption may also increase susceptibility to drought, salinity, and nutrients deficiency abiotic stresses. Losses in the flower yield and quality may then be aggravated. (Agrios, 2005) Also, these tumors are often associated with secondary pathogens, which aggravate the decline of plants and challenge management in commercial operations because of its DNA transfer ability. Studies conducted regarding *A. tumefasciens* have been largely in the field of biotechnology, though its pathogen role is perhaps one of the main issues faced in floriculture globally (Chilton *et al.,* 1977; Gelvin, 2017)

When a plant is injured due to agricultural activities like cutting, grafting, transplanting, or bending of shoots in the field or nursary, *Agrobacterium tumefasciens* enters through the wound. Injury caused by feeding insects, nematodes, or accidental handling of plants can also

cause entry (Jin *et al.*, 1990). After entry, the bacteria use polysaccharides and fibrils to adhere themselves to the plant cell surface. This helps them attach to and colonize the wound site. After attaching, T-DNA is delivered by a type IV secretion system that is encoded by virulence (vir) genes on the Ti plasmid. This marks an important step in crown gall pathogenesis (Gelvin, 2017). A rose crop can be reinfected multiple times during its life cycle especially in an intensive system where there is frequent manipulation of the plants. Cut flower roses get pruned a lot during harvesting. Because of this, the rose cut flower grows very susceptible. Also, repeated injuries allow easy bacterial entry. The strong relationship between horticultural practices and disease epidemiology explains the persistence of crown gall on high-value crops such as roses (Kado, 2014).

Taxonomically, according to Young *et al*. (2001), *Agrobacterium tumefasciens'* family is Rhizobiaceae, order Rhizobiales and class Alphaproteobacteria. Similarly, other bacteria of the order are symbiotic and pathogenic whose importance is majorly agricultural. The bacterium is a rod-shaped Gram negative (0.6 – 1.0 µm by 1.5 – 3.0 µm) and motile with peritrichous flagella, allowing it to actively infiltrate plant wound sites (Sider, 2009; Sigee, 1993). They grow in the presence of oxygen and do not form endospores. However, its capacity to produce extracellular polysaccharides helps it survive under changing environmental conditions. It is commonly found in soil and the rhizosphere of plants. The colonies are described as being slimy, smooth, whitish to cream coloured on laboratory media. The mucoid appearance of the colonies is relevant to survival and persistence in plant tissues (Young *et al.*, 2001). The bacterium closely resembles and functions similarly to the nitrogen fixing *Rhizobium* species. This indicates that it occupies a position with evolutionary benefits between beneficial and pathogenic lifestyles in plants (Goyal & Habtewold, 2023). Because of this duality, it has become one of the most studied plant associated bacteria.

Crown gall disease was one of the first plant diseases studied at a molecular level, historically speaking. In 1897, scientist Fridiano Cavara noticed issues like abnormal growths in grapevines. He isolated the causal agent and named it *Bacterium tumefasciens*. In the US, Smith and Townsend reported similar symptoms on chrysanthemums in 1907. This confirmed its pathogenic status and was renamed *Phytomonas tumefasciens* (Kado, 1976). The category of the pathogen was altered over the years and it was revised into *Agrobacterium tumefasciens*, which can be found in old literature related to plant pathology and is a name still widely used. These early findings formed the basis of molecular plant pathology and genetic engineering, as *A. tumefasciens* was recognized later as a natural genetic engineer capable of transferring DNA

into the genome of plants (Chilton *et al.*, 1977). After many years of research, crown gall has become a destructive disease as well as a mainstay of plant molecular biology.

The bacterium reduces the market quality and even the productivity of roses, which has a direct impact on its profitability. Plants that have fallen victim to pathogens often shows symptoms like stunting, yellowing, and loss of vigor. They may also fail to produce good quality flowers. Such plants also incur losses for vendors (Schell & van Montagu, 2009). In a serious infection, whole plants may wilt or die altogether. Young plants that have not yet established a foothold will be affected. The pathogen is tough to eliminate once it infects an area, as it can survive in soils for more than ten years. It can either survive in or on plant debris or as a free living organism (Shams *et al.*, 2012). In East Africa, floriculture is the main export sector. An outbreak of crown gall on roses caused high yield loss. Also, it was shackled by replanting, soil sterilisation and strict sanitation (Maina *et al.*, 2013). In order to ensure sustainable commercial, rose production, integrated management strategies should be developed. Recent studies have also revealed the devastating effects that crown gall disease has on the global rose sector, particularly in East Africa, where a substantial amount of cut roses is exported to Europe. Kenya, which brings in almost 40 % of EU rose imports, suffers from crown gall disease in major production centres such as Naivasha and Thika, which greatly lower export quality (Murugu, 2015).

Ethiopia is another blooming floriculture center. It has also been facing similar issues like crown gall which are decreasing its competitiveness. Expenses linked to the management of this ailment comprise direct losses resulting from the death of plants and indirect costs related to greater sanitation labour, larger chemical application, and lesser planted density due to soil contamination (Maina *et al.*, 2013). Moreover, crown gall in production areas can interefere with trade agreements and certification processes because countries that import require strict compliance with phytosanitary measures. It shows that crown gall affects global trade and the farmers' livelihoods that rely on ornamental flowers. Because of its unique DNA transfer mechanism, study of *Agrobacterium tumefasciens*, both pathogen and biotechnology tool has been extensive. Because of its ability to integrate foreign genes stably into the genome of plant, it forms the foundation of modern plant genetic engineering leading to the development of genetically modified (GM) crops showing traits like disease resistance, herbicide tolerance and improved nutritional quality (Zhongging *et al.*, 2022; Stanton, 2021). Roses are being genetically redesigned via *Agrobacterium*-mediated approaches for enhanced resistance to diseases, improved flower longevity and pigment biosynthesis. The use of this bacterium as both a biotechnology tool and a pathogen is a contradiction. The need for

integrated research research which combines plant pathology and biotechnology flowering plants economics for managing impact of crown gall disease is reiterated by this dual identity (Li *et al*., 2000).

2.1.11. Genome structure of *Agrobacterium tumefasciens*

Agrobacterium tumefasciens is special when it comes to the genome of bacteria because it is multipartite, containing both a circular and a linear chromosome, apart from two large plasmids. Many researchers have focused on this unusual genome architecture as it offers important information about bacterial evolution, adaptation to complex environments and pathogenicity mechanism. Unlike many other bacteria that have one circular chromosome. The dual-chromosome system of *A. tumefasciens* allows for functional diversification and genetic plasticity (Goodner *et al.,* 2000). This arrangement is thought to grant the bacterium a broad ecological range, allowing it to thrive in settings ranging from bulk soil to the rhizosphere and as a pathogen in plants. The organism is used in microbial genomics especially for investigating how genome partitioning influences bacterial physiology, host–microbe interactions and evolutionary adaptation (Barton, 2019).

One of the most studied *Agrobacterium tumefasciens* C58 isolates has been sequenced in greater detail. The *Agrobacterium tumefasciens* C58 isolate genome contains approximately 5.7 million base pairs (Mb). A circular chromosome (2.84 Mb), a linear chromosome (2.07 Mb), and two plasmids, namely the tumor-inducing (Ti) plasmid pTiC58 and an accessory or cryptic plasmid, pAtC58 (Goodner *et al.,* 2000; Wood, 2001). The complete genome sequence of this strain, the first from a plant pathogen, has made it a reference genome suitable for comparative studies of plant bacterial interactions. The circular chromosome serves the basic purpose of existence while the linear chromosome will carry the more responsible genes to adapt to various conditions. This kind of configuration was unheard of in bacteria until now and only a few groups like Streptomyces and Borrelia possess linear chromosomes. With the complete genome sequence now available, it is possible to dissect the genetic basis of virulence, opine metabolism and horizontal gene transfer (Wood, 2001).

The TIGR database analysis showed that the major metabolic pathways and protein synthesis in *A. tumefasciens* are very similar to those of *E. Coli* K-12. According to Goodner *et al.,* (2000), it is essential for the bacteria to survive in a broad range of environments. In contrast, the linear chromosome has many genes acquired by HGT (horizontal gene transfer). This includes genes needed for ecological fitness, colonization of hosts and adaptation to plant-associated environments (Lassalle *et al.,* 2011). Research indicates that this chromosome

contains genes that have a role in response to stress, degradation of aromatic compounds, and metabolism of secondary metabolites, and as such contributes to adaptation to their niche (Lassalle *et al.,* 2011). Surprisingly, the linear chromosome carries telomere like caps at the ends which protect the ends of DNA from degradation like eukaryotic genomes. Structural innovation is also present in other actinomycetes such as Streptomyces, indicating a converge evolution of similar mechanisms for linear chromosome maintenance in bacteria (Goodner *et al.,* 2000). *A. tumefasciens* harbors the most distinctive genetic elements, the tumor inducing (Ti) plasmid. The DNA region and the virulence genes of *Agrobacterium tumefasciens* are responsible for crown gall disease occurrence in plants (Wood, 2001).

The T-DNA is cut out and inserted into the genome of the plant host through a type IV secretion system coded by the vir operon, causing unregulated plant cell proliferation and gall formation (Gelvin, 2003; Stanton, 2021). Once T-DNA is integrated, it has been found to encode genes for the synthesis of plant growth hormones auxins and cytokinins which alters the growth of the plant favorably for tumor formation. In addition, the T-DNA encodes opine biosynthesis genes. Amino acids have unusual derivatives, known as opines, made in plant cell galls, which are *A. tumefasciens* metabolizes to get an enhanced ecological advantage (White & Winans, 2007). The Ti plasmid is essential for causing disease and also gives the bacterium a unique metabolic niche or environment. The second plasmid in question, pAtC58, was initially interpreted as being cryptic since it did not appear to be virulent. Later studies, however, showed that it helps bacteria ecology and survival considerably. This plasmid has genes that helps in breaking opines, tolerating to stress and interacting with microbes present in surrounding area (Ge *et al.*, 2023; Veluthambi *et al.*, 2003). It has also been suggested that pAtC58 enhances. *Agrobacterium tumefasciens* is competitive in non-pathogenic environments; for example, when it is in a soil saprophyte. Adding functions of the Ti plasmid, pAtC58 increases the ecological versatility of the bacterium, enabling it to assume saprophytic and pathogenic lifestyles dependent on environmental cues. The interaction of plasmids shows how multipartite genomes help bacteria to grow and last long (Kado, 2002).

Agrobacterium tumefasciens has been inferred from genomic studies. According to Brown *et al.* (2017), *A. tumefasciens* may have had ancesteral plasmid integration events, which became fixed later during evolution. Comparative genomic analysis reveals that related Rhizobiaceae (including symbiotic nitrogen fixing *Rhizobia*) share a similar genome architecture and an evolutionary origin. This dual lifestyle of pathogen and potential symbiont reflects the evolutionary plasticity of Ti plasmid from *Agrobacterium tumefasciens* and its close relation to beneficial root-colonising bacteria. Such genetic versatility explains why *A.*

tumefasciens can survive in many places including agricultural soils and plant tissues. It shows how horizontal gene transfer plays a role in bacterial evolution. The multipartite structure is thus an evolutionary innovation that strikes a balance between essential cellular processes and adaptive flexibility.

The Ti plasmid, on a biotechnological level, is a natural vector which has found applications in plant molecular biology. A binary vector system is obtained by replacing the tumor-inducing genes of the T-DNA region (located in one of the T-DNA regions) with genes of interest for generating transgenic plants. This enables stable incorporation of desired foreign genes into the plant genome (Gelvin 2017; Hoekema *et al*., 1983). The world's most widely used plant transformation technology made possible by this system valid for generation of transgenic crops with herbicide tolerant, pest resistant, drought tolerant and nutrition enriched traits. Researchers have used the *Agrobacterium* mediated transformation of plants not just for agriculture, but also for studying gene function, promoters, and regulatory networks. The Ti plasmid is a great example of how a plant pathogen can be re engineered to become a very important tool in biotechnology. The presence of two chromosomes and fewer large plasmids in *Agrobacterium tumefasciens* is responsible for its adaptability to many environments and its ability to cause diseases in plants. The multiple part organization helps the bacterium tradeoff general metabolic functions with particular ones to enhance plant colonization. Moreover, the Ti plasmid directly causes the bacterium to act as a plant pathogen and biotechnological tool (Hoekema *et al.,* 1983). These insights not only deepen our understanding of bacterial genome evolution but also highlight the paradoxical position of *A. tumefasciens* can be a terrible agriculture pathogen but is a good partner for genetic engineer. As genomic technologies improve, additional comparative analyses will probably uncover new dimensions of its genetic versatility for application in disease management and plant biotechnology.

2.1.12. Pathogenesis *of Agrobacterium tumefasciens*

Agrobacterium tumefasciens' pathogenesis is one of the most studied examples of bacteria plant interactions due to its interesting mechanism of horizontal gene transfer across biological kingdoms. Most pathogens infect plants with the aid of toxin production, enzyme production, and effector proteins (Jin *et al*., 1990). The DNA that is transferred or T DNA participates in the induction of disease by *Agrobacterium*. When plant cells take up the T-DNA of *Agrobacterium* they get genetically integrated into the genome of those cells (Moore *et al*., 1997). This genetic integration reprograms normal cells to proliferate uncontrollably and provide sores for tumorous growth (pathogenesis) called crown galls (COST 873, 2011).

Simultaneously, novel amino acid derivatives called opines are synthesized by the host plant cells. These opines are the only nutrient source for *Agrobacterium* (Escobar & Dandekar, 2003; Gelvin, 2003). This is the only known example of natural inter kingdom DNA transfer and has intrigued a range of plant pathologists, molecular biologists, and biotechnologists. The scientists chose to study *A. thaliana*. *A tumefasciens* because it is a serious agricultural threat that is also a valuable tool for genetic engineering (Li *et al*., 2000).

The bacteria are found mainly in the rhizosphere, where they grow on sugars, amino acids, and organic acids released from plant root exudates (Larebeke *et al.,* 1974). Infection can occur through opportunistic means because of wounds caused by agricultural activities such as pruning, grafting, transplanting, and mechanical tillage, as well as insect feeding and nematode damage (Agrios, 2005; Jin *et al.,* 1990). Plants produce wound derived chemicals, phenolic compounds (acetosyringone), soluble sugars and amino acids, known to act as strong chemoattractants for motile *Agrobacterium* to the injuried site. After being concentrated around the wound site, the bacterium utilizes a two component regulatory system that involves VirA sensor kinase and VirG response regulator to sense the compounds and activate the transcription of virulence genes (Lee *et al.,* 2009) The early step of bacterial infection is critical as it determines infection success, and the pathological relationship the bacterium will establish with the host. Plant cell attachment is necessary after chemical recognition for T-DNA transfer. A. The attachment of *A. tumefasciens* to damaged plant cell walls using polysaccharides, cellulose fibrils and adhesive proteins allow for a stable association with host tissues (Matthysse & White, 1995). This close contact gives the bacterium a secure launching pad for the DNA transfer machinery. Mutations that disturb adhesion greatly lessen infection efficiency, stressing the importance of this step in pathogenesis (Matthysse & White, 1995). In addition to sticking together, binding also helps share signals that control defense reactions. To illutrate, enzymes of the cell wall degrading type that are secreted upon attachment can compromise the strength of plant barriers while the plant attempts to resist this bacterium by launching an oxidative burst (Burr & Katz, 1983). The outcome of this communication between molecules decides whether an infection proceeds successfully or is blocked by the host. Once anchored to a host plant cell, the bacterium takes over the transfer of T-DNA. The specific T-DNA region of the Ti plasmid has 25 base pair border repeats, whose excision is catalyzed by the VirD1/VirD2 endonuclease complex. The resulting single-stranded T-DNA molecule, which becomes coated with VirE2 proteins, forms a nucleoprotein complex (T-complex) which stabilizes it and protects it from degradation (Christie & Cascales, 2005; Gelvin, 2017). A syringe like apparatus referred to as type IV secretion system (T4SS) exports the complex into

the plant cell. The Ti plasmid encodes this T4SS through the virB operon. After entering the host cytoplasm, the T complex is guided into the plant nucleus via nuclear localization signals on VirD2 and VirE2. Illegitimate recombination mediates its integration into random sites of the plant genome (Tzfira *et al.,* 2004). After integration, the host machinery transcribes and translates the T-DNA. The T-DNA oncogenes encode enzymes that disrupt plant hormone metabolism and cause an excess of auxins and cytokinins (Lee *et al*., 2009). This inability to balance hormones leads to uncontrolled division of cells and the formation of tumours at the site of infestation. Meanwhile, other T-DNA genes code for enzymes responsible for the biosynthesis of a number of opines, such as nopaline, octopine, and agropine (Gheysen *et al*., 2001). These opines are catabolized by *A. tumefasciens* and are unable to get benefitted from most other microbes. A dual action of tumor induction and opine production guarantees a situation. The *A. tumefasciens* invades and uses the resources within the newly created home of the plant. As the crown galls get larger, they interrupt the plant's vascular systems. So, it disrupts the flow of water and nutrients (COST 873, 2011). The impact of this interference on haploid and diploid becomes apparent as it manifests growth retardation, leaf chlorosis, reduced flowering, and overall decreased vigor of virulent plants (Schell *et al.,* 2009). After sometime, the tumours can become woody and necrotic. These impair the host further and make it prone to worms as well as secondary infections. Crown gall disease can kill the plant in severe cases especially in young or stressed plants. This can cause substantial economic losses to commercial floriculture and nursery industries (Shams *et al.,* 2012). The long-term survival of *A. tumefasciens* over a decade in soil which causes plant tumors, complicates management and control of this pathogen significantly (Sawada *et al.,* 1995). *Agrobacterium tumefasciens* has capacity to adapt to extreme environmental conditions in a natural way (Patentlens.net, 2014). Scientists developed binary vector systems for genetic transformation by disarming the Ti plasmid which involves removal of oncogenes as well as retaining T-DNA border sequences and virulence machinery (Hoekema *et al.,* 1983; Gelvin, 2017). Genetically Modified Organisms (GMOs) are engineered organisms that exhibit unique characteristics not found in the species originating within nature. Scientists create them using biotechnology. Today, this plant bacterium, *Agrobacterium tumefasciens* has been very useful in producing plants with improved qualities. However, besides plant improvement, it is also used for functional genomics, among other uses. Although *Agrobacterium tumefasciens* causes major agricultural problems, the organism has also been a valuable entry point for biotechnological advances ((Li *et al*., 2000).

2.1.13. Symptoms of crown gall in roses

Roses infected with crown gall disease exhibit some characteristic symptoms, which can, however, vary considerably with the age of the plant, the site of infection, environmental conditions, and bacterial inoculum density. One of the major symptoms of the disease is a gall like tumor that develops on the roots, crowns or stems, usually just above the soil line, but can also occur on aerial parts of the plant that have suffered mechanical injury and/or insect damage (Agrios, 2005; Shams *et al.*, 2012). Galls result from an uncontrolled growth of plant tissue caused by the incorporation of T-DNA into the host genome by *Agrobacterium tumefasciens*. These galls are a result of changes in hormonal balance and overproduction of auxin and cytokinin. The disease appears in both young and mature plants (Schell *et al*., 2009). However, younger and actively growing plants might be more affected. In fact, they sometimes suffer stunting or death. In the case of infection, it is crucial to keep in mind that the expression of symptoms can be delayed by weeks or months. Drought, nutrient deficiencies, and high temperature are environmental stressors that can aggravate gall formation and disease severity, complicating management in commercial fields of roses (Escobar & Dandekar, 2003; Gelvin, 2003). At the early stage of infection, galls appear as tiny, soft, whitish swellings at the wound and crown regions. These early swellings tend to be smooth and can be small, meaning that they are easily missed during regular checks, especially inside dense plantings or in greenhouses (Schell *et al.*, 2009). Galls currently disturb localized vascular tissue; they do not impair the physiology of the plant as of now. The plant looks fine and has healthy leaves and shoots. A few subtle changes may start to occur, like slight reductions in growth rate or localized chlorosis. It is essential to identify these soft galls at an early stage for the effective control of bacterial disease spread by means of removing the infected tissue, before the galls become hardened tumors. Some species of plant can continue to flourish under opportune circumstances, resulting in the increased likelihood of disease epidemics (Maina *et al.* (2013). As the disease continues, the galls grow bigger. They then become hardened and rough. They then take on irregular shapes and whitish or light brown colour as shown in plate 1b. As the disease progresses, they become darker, a shade of brown or black. Inside, the galls toggle a disorganized and hyperplastic parenchyma. These parenchyma cells grow uncontrollably due to the expressions of bacterial oncogenes on T-DNA (Escobar & Dandekar; 2003; Gelvin, 2003). When the xylem and phloem of a plant gets infected by these tissues, it affects normal flow of water and food conducting abilities and cause physiological damage to the host. When

large galls form on a plant, they become woody and fibrous in character. Since they become strongly fixed to the plant body, mechanical removal is not easy (Kado 2002).

Plate 1 (a): healthy rose plants (b): rose plants infected by crown gall

Severe infections that result in gall development interfere with shoot elongation, flowering, and overall architecture of the plant reducing yield and ornament quality. Also, if the gall is present for a long time, it will weaken the plant. This also makes the plant more susceptible to other pathogens. Similarly, such a gall can influence root system architecture. Moreover, the gall can also reduce water uptake efficiency and nutrient absorption (Kado, 2002; Schell *et al.*, 2009). Roses infected with *A. tumefasciens* show other symptoms due to stress. Plants may have diminished growth, yellowing of leaves, slow growth, late flowering, and poor quality flowers. Leaves might become yellow, fall off too early, or show uneven color, while shoots do not usually grow too much (Shams *et al*., 2012). Advanced infections cause wilting and necrosis in young tissues and premature senescence of flowers. Thus, this causes a serious degradation of the aesthetic value and marketability of these plants. The infection of the crown gall makes the plant more susceptible to other invaders (Schell *et al*., 2009). Also, other invaders include opportunistic bacteria, fungi, wood boring insects. The plant will decline faster when the infection occurs. Also, there is an increased risk of total crop loss. A disease management strategy which includes both cultural and chemical control of pathogens to keep the plant healthy and productive is needed (Agrios, 2005). In soil and plant debris, this disease is one of the most persistent pathogens. *A. tumefasciens* could survive for a little over ten years in contaminated soil. Even after the galls slough off or decay, *A. tumefasciens* can be a source of inoculum for new plants (Alsup, 2004; Kado, 2002). The rose can survive for longer periods

of time which contributes to the cycle of infection. This causes problems in the management of rose nurseries and commercial fields. Planting in infested soils repeatedly increases the severity of diseases and reduces the performance of the crop and causes a huge economic loss (Alsup, 2004). Soil sterilisation, crop rotation, and careful sanitation of propagation materials are therefore critical to limit pathogen persistence in soil. Also, the galls that develop in the crops may utilise bacteria which may adapt to the local conditions. This may develop resistance against the measures the farmers take (Maina *et al.,* 2013).

Crown gall diagnosed in the nursery and greenhouse is traditionally done by looking for tumors, especially in the area of the crown or graft union. Many series of abnormal growth like those caused by herbicides, mechanical damage, or hormonal imbalance could mimic gall symptoms (Pulawska, 2010). Molecular and serological diagnostic tools such as PCR-based detection and ELISA are increasingly being used to confirm the presence of *A. tumefasciens*. (Sawada *et al.,* 1995). Hasty and accurate diagnosis can permit targeted interventions such as removal of infected plants, sterilisation of propagation materials, and isolation of disease-free stock necessary to control the spread of crown gall in commercial rose production.

Crown gall disease severely impacts the cut flower sector of the industry. Plants that are infected with illness produce fewer flowers of lower quality. This reduces the vase life and the value of the flower (Maina *et al.,* 2013). Galls are unsightly blemishes that can lead to rejection of nursery stock in the domestic and export markets. The economic impacts mainly occur in major rose exporting nations like Kenya and Ethiopia where crown gall disease can hinder substantial revenues, lessen competitiveness in the international market and spike management costs of disease (Maina *et al.,* 2013). Apart from the economic losses directly linked with disease outbreaks, producers and the national floriculture industry's reputation can also be affected. Integrated disease management and breeding for resistant cultivars will ensure sustainable rose production (Thomas & David, 2014).

2.1.14. Host range and distribution

Agrobacterium tumefasciens is the cause of crown gall disease. It is one of the most widely affecting bacterial plant pathogens. It makes management difficult in various agricultural and horticultural systems. The pathogen has been found to infect more than 93 families of dicotyledonous plants including woody perennials and herbaceous annuals showing it is quite adaptable to different plant tissues and environmental conditions (Kado, 2002; Pulawska, 2010). The broad host range of *Agrobacterium tumefasciens* is due to the Ti plasmid. Furthermore, the Ti plasmid is responsible for the transfer of T-DNA, virulence factors and

opines synthesis. The opines are plant metabolites that are released upon wounding. The ability of *A. tumefasciens* to detect molecules like phenolic compounds, which are produced by infected plants is not unique to this agrobacterium. It also detects nopaline, produced by certain tumor shell plants, as well as other plant wound compounds. It is likely that various agrobacteria or root nodulating bacteria detect such phenolic compounds. This organism's ability to adapt to different environments affects the global floriculture and agriculture industries and genetic engineering studies. Furthermore, its global existence makes it difficult to contain and manage (Jin *et al*., 1990).

Crown gall disease is very damaging in woody ornamental crops like roses, chrysanthemums and Marguerite daisy as tumor formation adversely affects plant vigor, ornamental value and commercial value of the plants (Escober & Dandekar, 2003; Rhouma *et al.,* 2006). The bacterium *A. tumefasciens* is a serious disease of fruit crops. The disease affects stone fruits (cherries, peaches, apricots) and pome fruits (apples, pears) and nut crops (almonds, walnuts). Furthermore, it reduces growth and fruit set and reduces quality and yield (Burr *et al.,* 1999; Ophel & Kerr, 1990). Grapevines *(Vitis* spp.*)* are very susceptible to crown gall infections that harm vine physiology, reduce the yield of grapes, and decrease the life of the vineyard most importantly in young or intensively managed vineyards. Due to the increased management costs for monitoring, eradicating and maintaining disease free propagation material, the economic impact of *Agrobacterium tumefasciens* in these crops has been shown. The diversity in host range shows that the pathogen can adapt to both herbaceous and woody tissues. This has been made possible by the flexible genome of the pathogen, along with horizontal gene transfer and specialized virulence mechanisms which contribute to interexploitant and interspecific adaptability (Rhouma *et al.,* 2006). Crown gall disease was also described on a range of herbaceous crops including sugar beet, tomato, beans, alfalfa, and cotton. Illness caused on these crops, most often does not lead to huge economic consequences, in contrast to perennials (De Cleene & De Ley, 1976; Maarten *et al.,* 1987). These incidental infections show that the pathogen has the power to adapt to and survive in agricultural soils that have mixtures of cropping systems. Even mild infections on these crops may act as an inoculum reservoir for *Agrobacterium tumefasciens*. This can lead to a build up of *Agrobacterium tumefasciens* in soil and a greater risk of disease in neighbouring high value or other susceptible crops. The pathogen's ability to colonize a wide range of ecological niches indicates its evolutionary success and complicates common crop rotation or sanitation strategies, especially in the high-value ornamental and fruit production system areas (Gelvin, 2003).

Certain plants are widely used as assay hosts, in laboratory studies for virulence and pathogenicity of different *Agrobacterium* strains. For example, *Datura stramonium* (Jimsonweed), *Kalanchoe daigremontiana*, and *Helianthus annuus* (sunflower) (Kado, 2002). These plants are very sensitive and form tumors quickly so they are a good bioassay system for research and breeding. They can also be used for testing other biocontrol agents or disinfectants under controlled condition before applying them onto commercial crops. When indicator plants are used, it gives insight on strain variability, environmental adaptability, and host pathogen interactions to help in commercial production management decision making (Kado, 2002). The pathogen is also identified using conventional disease diagnosis methods based on the isolation of this bacterium on selective or general media, and pathogenicity tests on alternative plants including carrots and tomatoes (Chen *et al.*, 1991; Lopez *et al*., 1991). In addition molecular methods such as using primers designed from virulence (vir) genes or T-DNA sequences to detect and identify the pathogen from cultures, soils, and plant tissues (Chen *et al.*, 1991). Previous research indicates that *A. tumefasciens* can be detected in plant or soil samples using qPCR to target the chvE gene. This protein is encoded by the chvE gene and is responsible for a sugar induced increase in the expression of vir genes (Guo *et al*., 2019; Jailani *et al*., 2022; Petrovichev *et al*., 2017).

Agrobacterium tumefasciens is found in many places with famous crops. These include temperate, tropical and subtropical areas (Pulawska, 2010). There are many ways it can be moved around. These can happen via contaminated nursery stock. It can also be transmitted through soil, irrigation water, tools, and agricultural equipment. After establishing in a production system, the bacterium can persist for years in soil, plant debris, and contaminated propagation materials making it practically impossible to eradicate (Moore *et al.*, 1997). This species can withstand a wide range of ecological conditions as well as environmental stressors like drought, extreme temperatures and nutrient deficiency. Thus, it continues to thrive and reproduce. The worldwide prevalence of this disease highlights the need for quarantine restrictions, certification of pathogen free stock, and international vigilance against crown gall outbreaks (Kado, 2002).

Crown gall disease ranks as one of the most serious constraints to rose production in East Africa, especially in Kenya and Ethiopia, a leading export industry for the two countries (Maina *et al.*, 2013). Plants can have poorer growth, early dying, stunted growth and poor flower quality when infected. In turn, these lead to economic loss to growers and exporters. The constant presence of *Agrobacterium tumefasciens* in soil and the species' cyclomorphic nature makes it a high risk situation and it also poses a threat to the rose industry. In addition,

the visible galls cause aesthetic issues with cut flowers and may impact their marketability and acceptance. This may further affect international standards and trust for Kenyan and Ethiopian market (Rikken, 2012). Disease free planting material can be used, sanitation of soil, resistant cultivars, and biocontrol along with other integrated approaches to safeguard the viability of the industry (Julliete *et al.*, 2020).

2.1.15. Biotypes of *Agrobacterium tumefaciens*

The variety of strains of *Agrobacterium tumefasciens* has been known to affect the host range, pathogenicity and, epidemiology. Initial phenotypic and biochemical analysis revealed considerable differences among these isolates obtained from different plant species, geographical locations and farming systems. Because of these variations, a biotype classification was developed. Biotypes were classified according to physiological properties, biochemical activity, and host specificity. This information can be used for fundamental research and useful in controlling the disease (Lindström & Young, 2010; Kado, 2002). This classification also helped to reveal *Agrobacterium tumefasciens's* phylogenetic separation. *Agrobacterium tumefasciens* has a wide host range and kingom. It can infect other plants, particularly monocots, leading to a rapid adaptation of the bimodal pathogen.

Traditionally, *Agrobacterium tumefasciens* was based on their biochemical characteristics, tumor shape, and range of hosts they infect, 17 strains of *A. tumefasciens* were divided into 3 major biotypes. Biotype I strains are the predominant type and have a broad host range. They infect a wide range of dicotyledonous plants, which include several economically important ornamentals. Grapes, stone fruit and rose are also affected. Some strains, known to produce 3-ketosugars, are often isolated from commercial nurseries and vineyards, causing considerable yield loss and economic damage (Moore *et al.*, 1997). Typically, the strains of biotype II were associated with *Agrobacterium rhizogenes* that causes hairy root disease. They show different metabolic properties. For example, they prefer L-tartaric acid instead of glucose and produce polygalacturonase. Biotype II strains are frequently found in grapevines and in some woody ornamentals, they have a limited host range but are pathogenic on certain crops (Costechareyre *et al.,* 2010). Biotype III, initially included in *Agrobacterium*. Later they named it *Agrobacterium vitis*, a species specialised for grapevine infection. The genetic examination of Biotype III isolates established them as being genetically distinct. They had a narrow host range, restricted primarily to members of the genus *vitis*. This is of great significance to the worldwide wine industry (Ophel & Kerr (1990). Other specialized strains, such as those

infecting *Rubus* (raspberry and blackberry), which have been described as *Agrobacterium rubi*, though they are closely related to other *A. tumefasciens* (Young *et al.*, 2001).

The classical biotype system has undergone considerable refinement owing to the advent of molecular techniques such as DNA - hybridization, 16S rRNA gene sequencing, multilocus sequence analysis (MLSA) and whole genome comparisons. The research findings indicated that *Agrobacterium* was a group with high genetic diversity. Many lineages previously considered biotypes were elevated to species based on phylogenetically based evidence (Young *et al.*, 2001). For instance, Biotype ll isolates are now known as *Rhizobium rhizogenes* and Biotype lll isolates have been classified as *Allorhizobium vitis*, owing to their limited host specificities and lineage. Many of the species formerly classified as *Agrobacterium tumefasciens* are now placed on the genus *Rhizobium* based on their genetic similarity with nitrogen fixing symbionts. Even with these changes in classification, the name *Agrobacterium tumefasciens* is still used in many plant pathology and biotechnology publications for strains that induce crown galls and transfer T-DNA (Lassalle *et al.* (2011).

Understanding *Agrobacterium* at the biotype and species level is practically and theoretically useful for disease management and plant biotechnology. Some biotypes are specific to the hosts. Strains of tumor inducing bacterium that infect ornamental plants require different monitoring and sanitation measures than those infecting grapevines (Gelvin, 2017). In plant biotechnology, the correct choice of strain is important because, with the variability of individual biotypes, it affects efficiency of T-DNA transfer. Their compatibility with different classes of plants and transformation success also varies (Lassalle *et al.* (2011). A range of biotypes as well as derived strains have been engineered to serve as efficient vectors for plant transformation displaying how the plant pathologist has harnessed their biological properties to enhance biotechnology. Diseases are reported to develop in specific regions and are primarily caused by region specific biotypes. An example of this is the grape and stone fruit disease outbreak in the 90s, which warranted a survey and quarantine (Burr *et al.*, 1998). Furthermore, the creation of opines and other metabolites has improved ecological fitness and competition of certain biotypes in the plant rhizosphere due to greater genetic and metabolic diversity. In summary, *Agrobacterium* biotypes research is very useful for controlling the disease and exploiting this organism for biotechnology.

2.2. Management of *Agrobacterium tumefasciens*

It is quite difficult to manage crown gall disease in roses and other susceptible crops due to the features of *Agrobacterium tumefasciens*. The pathogen is very resistant and survives in the soil and plant debris for over 10 years without a host, which makes it very difficult to eradicate once it becomes established in a production system (Kado, 2002; Shams *et al.*, 2012). Additionally, the bacterium cannot be routinely eliminated from infected plants or infected soils by any bactericides or other chemical treatments since crown gall disease has a quite wide host range. Further, the disease infects over 90 dicots families (Pulawska, 2010). As a result, management relies heavily on integrated strategies that combine preventive cultural practices, biological agents and other chemical agents where appropriate. Also, it is possible to devise strategies that minimize chances of infection while not causing harm to the plants nor reducing productivity through knowledge of the pathogen's biology of plant wounding and T-DNA tranfer. A mix of management strategies can not only cut down disease incidence but also safeguards long term sustainability of rose production systems and other affected high end crops (Kenya Flower Council, 2021).

Crown gall disease occurs when three factors are present; (1) host plant that is capable of being infected, (2) virulent strain of *A.tumefasciens* and (3) environmental conditions that favour survival and access to wounded plant tissues such as the presence of plant nutrients (Agrios, 2005; Moore *et al.*, 1997). To successfully manage a disease, one or more components must be targeted. Using resistant or less susceptible cultivars can greatly reduce the occurrence of disease for example, *Pythium* in nurseries and greenhouses where young seedlings are less vulnerable. In transplanting, pruning and handling, prevention of wounding is also essential since entry of bacteria into the host is wound dependent. Environmental managing by preventing too much irrigation that creates favorable conditions in the soil for the survival of the bacteria helps remove chances of infection (Maina *et al.*, 2013; Pulawska, 2010). Cultural methods are key in the management of crown gall. Regularly cleaning and disinfecting pruning shears, grafting knives, and propagation benches will stop the disease from being spread from one plant to another mechanically (Alsup, 2004). Rose cultivators who rely on disease free planting material, ideally, certified clean stock, reduce inoculum pressure. Crop rotation by the use of non host species, removal and destruction of infected plants and avoiding the replanting of susceptible crops in soils formerly infested are other cultural practices that lessen bacterial persistence and reduce secondary infection. Also, proper nutrition and healthy plants are better equipped to resist infections. For example, vigorous plants are less likely to produce galls than stressed plants or those with nutrient deficiencies (Shams *et al*., 2012).

Managing *A. tumefasciens* has become easier with the emergence of biological control. The population of *A. tumefasciens* thrives in organic or eco-friendly production systems. Many people have used *Agrobacterium radiobacter* strain K84 that does not cause diseases as a biocontrol agent. Agrocin 84 is an antibiotic produced by it. Also, it prevents *A. tumefasciens* strain from inducing tumor. Strains of *AgrobacteriuAm tumefasciens*. K84 has been shown to reduce the incidence of crown gall in the field and in greenhouses when used as a soil drench, a seedling dip, and a wound protectant. The efficacy of biocontrol agents may be affected by environmental conditions, soil pH, and compatibility with the species of host plants, thus requiring site-specific management (Pulawska, 2010; Shams *et al.,* 2012). Not many chemical options are available, but some treatments can lessen inoculum in propagation material. Cuttings and trimming tools can disable *A. tumefasciens*, for example with hot water treatment on insides of plant tissues with tissue integrity. Broad-spectrum agents have been tested in some production systems, but soil fumigation rarely is a sustainable practice due to environmental impacts and regulation (Moore *et al.,* 1997). Methods of physical control are also used for reducing pathogen load in the topsoil. For example, infested soils can be solarised or deep-plowed. However, these methods are labour-intensive. In any case, these methods may not be suitable for large-scale operations. Since no individual approach is completely successful, integration of approaches is the most successful management programme. A more elaborate program includes the use of disease free propagation material, cleaning and disinfectant, use of biological means with non-pathogenic strains, cultural management to prevent wounding and monitoring for early detection of infection. When nursery staff and growers are educated on disease recognition, hygiene and preventive practices, integrated management becomes more effective. For long term sustainability, the affected crops including rose production should be subjected to these measures continuously. These measures should be monitored and modified according to local environmental and economic conditions (Agrios, 2005; Maina *et al.,* 2013). Dealing effectively with crown gall requires knowledge of the pathogen, their host susceptibility and environmental factors. Despite the challenges posed by its persistence and wide host range, an integrated approach offers the greatest opportunity to diminish disease incidence, protect plant health and ensure the viability of commercial rose and ornamental production systems.

2.2.1. Cultural control

Cultural control methods are the first and foremost important method of control against crown gall disease due to *Agrobacterium tumefasciens*. The first array of practices seeks to protect against the introduction and spread of the pathogen through sanitation, hygiene and management of plant wounds. The latter is the main route of entry for bacterium (Agrios, 2005). For the cultural methods to become effective, they need to be applied consistently. Furthermore, it has to be done carefully and in conjunction with other management practices. Additionally, cultural practices improve the health of rose plants and enhance their resistance against secondary infections and environmental stresses. Unlike interventions that involve chemicals or biologicals, cultural practices are cheap, environmentally safe, and appropriate for both smallholder and commercial rose production systems, thus very practical and sustainable in the long run (Murugi, 2015).

Keeping the production environment clean minimizes spread of pathogens. Cutting tools, pruning shears, grafting knives and other such equipment disinfected regularly and kept well. Transfer of *Agrobacterium tumefasciens* from infected to healthy plants (Maina *et al.*, 2013). To lessen bacteria surviving on the surfaces in greenhouses and nurseries, benches, propagation trays, and irrigation systems. Tools (nails and scissors) are commonly disinfected with 70 % ethanol, sodium hypochlorite solution and quaternary ammonium compound. A study reported that sanitizing cutting tools lowered gall formation in *Datura stramonium* caused by *A. tumefasciens* (Kado, 2002). However, some studies have indicated that, even if all of the plant material with visible crown galls is removed from a nursery, growers may still purchase and plant apparently healthy plants that are actually infected without showing any symptoms, because the pathogen can still be present in the plants. In this way, pathogens can easily spread which might cause difficulties in managing crown gall. Also, removing plant debris and fallen leaves as these can harbor the pathogen reduces the chances of secondary infections and limits inoculum build up in the soil (Pulawska, 2010). Routine sanitization stops other pathogens in soil from growing too. This makes the whole nursery healthier. Preventing mechanical damage to the plants is essential as infection is wound dependent. When plants are carefully handled during transplanting, pruning, bending or harvesting the potential infection sites are reduced (Agrios, 2005). Staff should be trained to safeguard plants against rough handling, sharp impacts and avoid unnecessary manipulation of their stems and roots. Using clean gloves and sterilized trays, plus not stacking or crowding, will also help reduce the injury of the cuttings. Also, performing these operations in periods of low humidity or on less stressed plants can reduce the risk of bacterial colonization and gall formation. (Shams *et al.*, 2012). By

providing plants with the best water, food, and heat, they will be less stressed. A reduction in stress means animals will attack them less. More importantly, the plants' natural defence against nasty pathogens will improve.

Getting rid of those plants with a severe infection is a removal that can help lower the inoculum levels in their area. Plants with large, old galls and severely stunted growth should be uprooted and destroyed either by burning or burying so that the bacterium will not survive (Kado, 2002). If infected individuals are identified quickly and monitored continuously, disease spread will be limited while keeping overall nursery healthy. It's usually not a good idea to partially remove the galls because cutting by itself will make the wounds worse and will allow for the entry of more bacteria (Maarten *et al.,* 1987). Also closely monitoring new plantings for early gall formation will allow for quick intervention to prevent widespread outbreaks. The promotion of *A. tumefasciens* infection is significantly aided by biotic vectors such as root chewing insects, nematodes and other wounding agents. Controlling these vectors lowers the chance of wounds from which bacteria could enter and limits pathogens' dissemination (Maarten *et al.,* 1987; Pulawska, 2010). Strategies like biological control of nematodes, deployment of insect proof nets and careful monitoring of insect population are incorporated in integrated pest management to reduce the risk of infection. Healthy soil structure and moisture levels can help to stop the population growth of vectors. Furthermore, planting resistant varieties, crop rotation and avoiding planting susceptible hosts into previously infested fields can help break the pathogen vector cycle and limit long term incidence of disease (Maina *et al.,* 2013).

New localised treatments were being used by several growers other than the established cultural techniques to lessen gall formation. Use of fresh cow dung at the base of the plant as a microbial amendment, regulation of copper nutrition to improve plant defence, and cutting of small galls followed by corn oil application to reduce bacterial spread has been reported (Maina *et al.* 2013). Though not scientifically verified to any great extent, these methods are believed useful when used along with basic hygiene and sanitation. If these techniques continue to be experimented on, field tested or trialed, they may offer effective and cheap options for small-scale growers unable to afford commercial biocontrol agents. Combining these efforts with the best practice of regular monitoring and early rogueing, can form a more comprehensive cultural management system that reduces primary and secondary infections.

The use of cultural control is essential for controlling crown gall in roses. Repetitive use of the same will not trigger the pathogen in a more independent area of the field, and it will neither duplicate itself at the field level. Rose growers can ensure their crops are healthy and

high quality, while reducing the economic impacts caused by crown gall through careful sanitation, wound prevention, rogueing, vector management and grower innovations. The long term sustainability and effectiveness of these practices will be ensured through proper documentation, staff training and continuous monitoring (Murugi, 2015).

2.2.2. Biological control using *Agrobacterium radiobacter*

One of the most successful and scientifically validated biological approaches to crown gall management is the use of *Agrobacterium radiobacter*, particularly strain K84. This bacterium is a naturally occurring, non-pathogenic relative of *A. tumefasciens* and has been widely adopted because of its ability to suppress crown gall pathogens (Murugi, 2015). Unlike chemicals that may cause phytotoxic effects or disrupt soil microflora, K84 provides a targeted, environmentally sound solution. Its action is preventive rather than curative, offering rose growers a sustainable method to protect their crops against infection.The biocontrol efficacy of strain K84 is largely due to its production of a highly specific antibiotic like compound called agrocin 84. This compound is unique in that it selectively targets crown gall pathogens without affecting beneficial or neutral microorganisms in the soil. Agrocin 84 inhibits the replication of pathogenic *A. tumefasciens* DNA, thereby halting bacterial proliferation and gall development. By acting in a highly selective manner, agrocin 84 safeguards the soil microbiome while effectively suppressing disease pressure (Ryder & Jones, 1990).To achieve this effect, strain K84 must successfully colonize plant wound sites, which are the natural infection courts for *A. tumefasciens*. Once established at these sites, K84 competes for resources and ecological niches with pathogenic strains. Importantly, K84 metabolizes opines specialized compounds secreted by tumor cells that normally serve as food sources for pathogenic *Agrobacterium*. By consuming opines, K84 effectively deprives the pathogen of energy, thereby reducing its capacity to initiate infection. Furthermore, the ability of strain K84 to persist in the rhizosphere and on plant surfaces for extended periods provides long term protection, even after initial application (Chen *et al*., 2007). However, the success of K84 is strongly influenced by environmental factors. Soil pH, moisture, temperature, and the physiological state of the host plant can all affect colonization and biocontrol efficiency (Maarten *et al*., 1987). For this reason, K84 tends to perform more reliably in controlled environments such as nurseries and greenhouses compared to open fields, where conditions fluctuate more widely. To improve its practicality, commercial formulations such as Dygal**l** have been developed. These products are widely used for roses and other susceptible crops, and they can be applied as soil drenches, sprays, or dips on seedlings and cuttings (Pulawska,

2010). The timing of application is critical. K84 is most effective when applied preventively, before infection has occurred. Once galls are already established, the bacterium cannot eliminate the pathogen. Thus, integration into early nursery practices, such as dipping cuttings before planting, ensures the best results (Ryder & Jones, 1990).

Despite its effectiveness, limitations exist. Over time, pathogenic *A. tumefasciens* populations may develop resistance to agrocin 84 through horizontal gene transfer of the agrocin plasmid. Field studies in Kenya, for instance, have reported the presence of galls on K84 treated roses, demonstrating that resistance can occur and reduce effectiveness (Maina *et al.,* 2013). This underscores the risks of relying solely on a single biocontrol method. To address this challenge, researchers recommend combining K84 with other management strategies, particularly cultural methods such as sanitation, wound prevention, and vector control. Periodic rotation with other biocontrol agents also helps reduce resistance build-up. Continuous monitoring of pathogen populations is equally essential to detect resistance early and adjust management approaches (Pulawska, 2010). In conclusion, *Agrobacterium radiobacter* strain K84 represents a landmark in the biological control of crown gall disease. Its specificity, persistence, and ecological safety make it an invaluable tool for integrated management systems. Recombination DNA techniques have been used to construct a new biological strain, K1026, identical to the K84 strain.The K1026 strain does not have the ability to transfer its mutant agrocin 84 plasmid to other bacteria (Burr *et al*., 1999; Chen *et al*., 2007). The *A. radiobacter* strain K1026 was suggested as a safer organismthan the K84 strain to control crown gall (Eastwell *et a*l., 2006). Several studies have indicated that other bacterial strains, including *Rahnella aquatilis* HX2（Rosenberg *et al.,* 2016), *Pseudomonas fluorescens*, *Bacillus subtillis*, *Curtobacterium* spp. *Pseudomonas* spp. Sn48, *Pseudomonas* spp. Ba35, *Pantoeasp*. Sa14 and *B. velezensis* CLA178 (Kawaguchi *et al.,* 2012), have various effects in reducing crown gall development. No chemical products have been effectively suggested for the control of crowngall disease. The biological control of crown gall disease using the *A. radiobacter* strains K84 and K1026 is a significant achievement in helping propagation nurseries battle with this disease worldwide. However, alternative products should be researched as chemical products used to control crown gall whenever serious failures or unexpected deleteriouseffects of the suggested biological control products are considered. Because of resistance risks and environmental influences, it should not be used in isolation but rather as part of a broader integrated disease management program. For rose growers, this approach offers a reliable, sustainable, and cost-effective method to limit crown gall and protect economic returns (Maina *et al.,* 2013).

2.2.3. Biological Control Using *Trichoderma* spp.

Trichoderma species are known as beneficial fungi, which have strong potential in suppressing *Agrobacterium tumefasciens,* a soilborne pathogen by various mechanisms. This fungus can control diseases using antibiosis, mycoparasitism, competition for nutrients and induction of host plant systemic resistance, and could thus be used as a component in integrated disease management programs (Howell & Stipanovic, 1995). *Trichoderma* has many types of actions by which it controls and improves the pathogen population. It also promotes plant growth by enhancing root development and nutrient uptake. These fungi can grow in the soil around a plant's roots and on the plant's surfaces. Fungi create a protective barrier that helps prevent a pathogen from establishing disease in plant roots or leaves (Hermosa *et al.,* 2012). The substance produced by virens can be used as an antifungal agent on a broad range of pathogenic fungi. Usage of virens produces a number of bioactive metabolites like gliovirin, peptaibols, polyketides, among others. Antifungal agent virens are produced by fungi of order Hypomyces. These antifungal virens affect a variety of plants in the environment, the metabolites can directly inhibit or Stop the growth of *A. tumefasciens* and killing it. In addition, *Trichoderma* will stimulate the activity of defence related enzymes in plants, like chitinases, peroxidases and glucanases. As a result, systemic resistance improves the capacity of plant to resist pathogen attack. In addition, systemic resistance contributes to disease suppression (Akashi *et al*., 2018). Commercial *Trichoderma* formulations, like Tricotech, have found increasing use as biocontrol agents in nursery and field situations in Kenya and rose producing countries (Harman *et al.,* 2004) These products may be applied as spores or mycelial suspensions to soil, or as seedling dips to colonize root surfaces, and they act both prophylactically and curatively against crown gall (Evalon *et al.,* 2024). Research indicates that utilizing *Trichoderma* along with cultural operations, like sanitation and careful pruning, significantly reduces gall incidence and makes the plants more vigorous. Using *Trichoderma* is good for the environment and is compatible with organic farming systems. They can also minimize the need for chemicals. This is good for sustainable rose production. In the future, we will select better strains and application times so that they can work well under different conditions (Evalon *et al.,* 2024).

2.2.4. Biological Control Using *Bacillus subtilis*

Bacillus subtilis and each other related species are among the most commonly used soil microbes for biological control in agriculture owing to their capabilities of suppressing a range of plant pathogens. Bacteria are capable of producing a variety of secondary metabolites inclusive of the antibiotics, lipopeptides (surfactin, iturin, fengycin), siderophores and hydrolytic enzymes that directly inhibit the growth of pathogens and damaging their cell structures (Backman *et al.,* 2018). By competing for nutrients and colonization sites, *B. subtilis* helps to stop nasty microbes. They stop their attack in the rhizosphere. They create a microbiome. On top of that, these bacteria also form endospores that help them survive unfavourable environmental conditions and protect the crops over a long period. Research in a field has shown that multiple soil or foliar applications enhance the colonization and persistence to maximize biocontrol efficacy. Mixing *B. subtilis* with other good microorganisms can help stop crown gall from developing (Alvarez *et al.,* 2020). The bacteria from the sulfur reducing genus *Bacillus* have been shown to promote plant growth and development. This refers to the ways in which one organism can reduce stress levels or induce better growth and development in others (Glick, 1995). The organism also activates systemic acquired resistance (SAR) in the plant. This activates a flow of defence related genes that prepare the host for invasion by pathogens (Backman *et al.,* 1997). This kind of resistance can enhance tolerance not only to *Agrobacterium tumefasciens* but also to other soil borne pathogens. Some strains also produce VOCs that enhance plant growth and suppress pathogenic microbes. These features make *B. Subtilis* a sustainable, low cost, organic and eco friendly solution for rose nursery, commercial floriculture systems (Evalon *et al.,* 2024). Plant growth promoting rhizobacteria (PGPR) play a crucial role in biocontrol. PGPR can colonize the roots of plants, competing with harmful microorganisms and soil borne pathogens (Meena *et al*., 2020; Nagrale *et a*l., 2023; Santoyo *et al*., 2021). This process often involves various antibiotics, which protect the host plant from pathogen infection (Liu *et al*., 2024; Paterson *et al.,* 2017). Many strains in the genus *Bacillus* are typical PGPR with a broad spectrum of antimicrobial activity. Due to their ability to produce endospores that are highly tolerant to environmental conditions, *Bacillus* strains have a significant advantage in the preparation and application of products (Ben *et al*., 2021). As a result, many *Bacillus* strains have been successfully developed as commercial biocontrol agents (Cochrane *et al*., 2016; Fira *et al*., 2018; Pan *et al.,* 2023). Many PGPR also inhibit soil borne diseases through triggering induced systemic resistance (ISR) in plants (Farag *et al*., 2013; Grady *et al*., 2016). In this process, PGPR provide preemptive protection for plants when exposed to pathogens

(Khan *et al.*, 2022).The jasmonic acid (JA) and ethylene (ET) pathways playimportant roles in ISR, which are to salicylic acid (SA) pathways involved in the systemic acquired resistance (SAR) (Beneduzie *et al.*, 2012; Meena *et al.*, 2020; Zhu *et al*., 2022), PGPR including *Bacillus* strains have also been used in the biocontrol of crown gall disease. For example, researchers isolated PGPR strains from the rhizosphere of roses, which reduced gall size by 26 % (Kavutu *et al*., 2022), In another study, twelve bacterial strains isolated from the rhizosphere were tested in combination with commonly used antagonistic agents K84 and K1206 against crown gall in stone fruit nurseries (Rhouma *et al*., 2008) and several strains significantly reduced the incidence of the disease. In addition, it was found that the strain *B. albus* JKXZ3 exhibits a strong antagonistic effect against crown gall in cherry trees (Yan *et al*., 2020), while *B. amyloliquefaciens* JK10 can effectively control crown gall in blueberries（Kim *et al*., 2011; Yao *et al* 2022). Although there are studies reporting that isolated PGPR strains are effective in inhibiting crown gall（Sunita *et al*., 2006) only few focused on the mechanisms by which PGPR strains protect host plants from crown gall disease（Kuzmanovi´ *et al*., 2018).

In countries around the world, diseases like powdery mildew, fire blight, root rots, and bacterial wilts in crops have been controlled using *Bacillus* based bioproducts (Lastochkina *et al.,* 2019). The ability of *Bacillus subtilis* to reduce gall formation caused by *Agrobacterium tumefasciens* on rose plants depends on strain selection, formulation, and time of application. Practical application can be done through seed treatment, soil drenching as well as foliar sprays depending on the crop and conditions (Mnif, 2015). Also, *Bacillus species* comply with organic certification standards suitable for environmental rose production. Researchers continue to look for improved strains and new formulation technologies to maximize shelf life, field persistence and disease suppression (Evalon *et al.*, 2024).

2.2.5. Chemical Control

The use of chemicals for controlling crown gall disease is limited and unreliable as *A. tumefasciens* is not very sensitive to common bactericides and antibiotics and quickly develops resistance. Copper hydroxide, which is Copper Oxychloride, combats the growth of bacteria but is usually ineffective (Agrios, 2005). Also, if these substances are continuously employed, phytotoxicity may occur. In addition, it inflicts burning on the leaf, stunted growth, or poor quality of flower on the susceptible cultivar of specific rose classes (Arim, 2011). Furthermore, they clear and inhibit soil microbial community and also lower soil fertility. In addition, copper residues could stick to the plant surfaces leading to regulatory and ecological problems. Previously, some chemicals like 2-methoxyethyl mercury chloride, copper oxychloride and an

aqueous solution of cuprammonium were tested as pre planting dips for apple seedlings, and copper oxychloride and an aqueous solution of cuprammonium were shown to offer good possibilities for the control of crown gall in apple seedlings (Grim, 1987). Similarly, Utkhede & Smith (1993), found that using copper oxychloride as a root dip treatment effectively reduced crown gall infection in apple trees, but also resulted in phytotoxicity issues. In the other study, copper oxychloride was used to control crown gall disease on roses, but it was not successful in inhibiting crown gall growth (Opisa *et a*l., 2021).

Farmers often have to alternate or combine their chemical treatments with others in order to achieve marginal control. Antibiotics like streptomycin and tetracycline can sometimes lower bacterial numbers. Nevertheless, they are not yet widely used in agriculture due to cost, regulation and the risk of generating antibiotic resistant strains of the pathogen (Brent & Hollomon, 1998). As public concerns increase about health, residues in food as well as the environment, there is limited use of copper oxychloride. Because crown gall infections affect the entire plant, antibiotics will not get to the pathogen in the tumors after being applied to the leaves or the soil. If some bacteria are able to survive and produce new colonies, infection can again occur even if not totally killed (Kado, 2014). As a result, they usually do not get prescribed or used for a long time. They are mostly used in experiments or in laboratories. Temporary disinfection of surfaces, tools, or growing media can be performed using oxidizing agents like sodium hypochlorite or hydrogen peroxide (Maina *et al.*, 2013). But they cannot help with infections that have already spread throughout the rose plant. The bacterium resides in the vascular tissues and within the galls (Kado, 2002). These chemicals prevent cross-contamination in propagation or transplanting but do not cure existing infections. When plants need too much care, it is tough to manage as it causes damage. Chemical treatments are considered secondary controls for that reason. Because of these limitations, this chemical application is usually seen as a final option and rarely recommended alone (Maina *et al.*, 2013). Biological and cultural practices are the best management measure for crown gall disease. Farmers should be encouraged to use resistant varieties, biological inoculants and hygiene measures rather than chemicals for preventing any damage. Scientists are studying eco friendly chemicals which, if used in management strategies, they do not cause phytotoxicity and help to keep the environment safe (Evalon *et al.*, 2024).

2.2.6. Future Perspectives in Crown Gall Management

While current management strategies provide varying degrees of success, the future of crown gall management lies in developing more sustainable, precise, and scientifically advanced methods. The complexity of *Agrobacterium tumefasciens* interactions with host plants, soil environments, and microbial community demands continued research and innovation. Several promising directions are already emerging (Barasarathi *et al*., 2025)**.** One area of focus is genetic resistance. Advances in molecular biology and plant breeding are paving the way for roses and other ornamentals to be developed with enhanced resistance to crown gall. Through conventional breeding, marker assisted selection, and more recently, gene editing technologies such as CRISPR-Cas9, researchers are identifying resistance genes and engineering plants with reduced susceptibility to *Agrobacterium* infection (Gelvin, 2017). While commercial deployment of genetically resistant roses has not yet occurred, this approach holds long term promise, particularly for high value ornamental crops where crown gall can cause substantial economic losses. Another frontier lies in microbial ecology and the manipulation of soil microbiomes (University of Adelaide). Increasing evidence shows that soil microbial communities can significantly suppress or enhance disease outcomes. Research is now focusing on the use of microbial consortia complex mixtures of beneficial bacteria and fungi that can establish protective microbiomes around plant roots. Such probiotic soils may provide broad spectrum suppression of *A. tumefasciens* while also promoting overall plant health and resilience (Compant *et al*., 2019). Advances in the next generation sequencing and metagenomics are accelerating these discoveries, enabling scientists to identify beneficial microbes and design custom formulations for commercial use. Nanotechnology is another emerging tool in plant disease management. Nanoparticles, due to their unique properties, can act as carriers for antimicrobial compounds or as direct antibacterial agents. For example, silver nanoparticles have demonstrated activity against a wide range of plant pathogens, including *Agrobacterium*. While still in the experimental stage, the integration of nanotechnology into crown gall management could lead to novel, highly targeted treatments that minimize environmental impacts compared to conventional chemicals (Compant *et al*., 2019). Future management will also emphasize digital agriculture tools. Remote sensing, artificial intelligence and machine learning are increasingly being applied to disease monitoring and prediction. By analyzing environmental data, growers may soon be able to forecast crown gall risk under specific field conditions, enabling more precise timing of preventive measures such as biocontrol applications or sanitation practices. Such precision agriculture approaches enhance efficiency and reduce unnecessary interventions, making crown gall management

more sustainable and cost effective. Finally, future perspectives highlight the need for stronger integration of research, policy, and farmer participation. Effective adoption of new technologies requires not only scientific advances but also policies that support safe commercialization, farmer education, and equitable access. Strengthening extension services, establishing participatory research programs, and providing incentives for sustainable practices will be critical in ensuring that innovations reach growers in both large scale and smallholder systems. **I**n summary, the future of crown gall management is shifting toward a multifaceted paradigm that integrates genetic resistance, microbiome engineering, nanotechnology, digital tools, and participatory extension systems. Together, these advances promise to enhance the resilience of rose production systems while reducing reliance on environmentally harmful chemicals (Gelvin, 2017).

2.3. Induced systemic resistance

Various approaches can be used to induce resistance which is one of the hope for management of diseases. Induced resistance is significantly different from direct chemical control, which tackles the pathogen. Rather, it activates the innate immune system of the plant for a more efficient immune response in the future. This method provides a green, low-cost, disease management method that is sustainable. Apart from the low cost, an important benefit of this method is chemical residue and environment safety in floriculture (Pieterse *et al.*, 2014). Inducing resistance reduces the cost of chemical applications. In addition the overall plant health and production is increased by stimulating the plant's defense mechanism before infection. Plants may not have immune systems like animals (Spoel *et al*., 2003). But they can fine tune their defense mechanisms by responsponing to changes in the environment. Induced systemic resistance (ISR) refers to the defensive response which is initiated against a broad-spectrum of pathogenic organisms in plant roots by groups of microorganisms. For instance, when PGPR colonize the roots, a defence is triggered in aerial parts as well as roots (Shah, 2003; Van Loon *et al.*, 1998). Systemic acquired resistance (SAR) is induced following pathogen infection or when specific signalling molecules are applied. The result can be broad spectrum resistance against a range of pathogens (Glazebrook, 2001). The first recorded experiment demonstrating induced resistance occurred in the 1960s when salicylic acid (SA) or its analogs were shown to induce resistance of apples to apple scab *(Venturia inaequalis)* (Williams & Kuc, 1969). A number of studies since then have shown that resistance can be induced chemically or biologically in a wide variety of crops with useful applications in horticulture and agriculture.

Three primary phytohormones are essential in inducing resistance. Salicylic acid (SA) has been connected to resistance against biotrophic and hemibiotrophic pathogens. It also induces systemic acquired resistance (SAR), which is characterized by the accumulation of pathogenesis related (PR) proteins that strengthen the cells defense (Grant & Lamb, 2006). On the other hand, jasmonic acid (JA) and ethylene (ET) are linked with resistance to necrotrophic pathogens and herbivores and are often associated with ISR, particularly after the application of beneficial microbes (Pieterse *et al*., 2014). According to Thaler *et al.* (2004), the interaction of these signaling pathways allows plants to adjust their defense against different pathogens so that a balance can be achieved without wasting resources. Induced Systemic Resistance is a potential alternative to chemical control of crown gall disease. The use of SA and its synthetic analogue, benzo (1,2,3)-thiadiazole-7-carbothioic acid (BTH), was shown to activate SAR in roses and other crops resulting in a notable reduction in the severity of bacterial diseases (Ryals *et al.,* 1996; Wang *et al.,* 2005). Plants that can't store SA, like the sid2-2 group in *Arabidopsis thaliana,* are more vulnerable to *A.tumefasciens* infection. Studies showed that constitutive accumulation of SA can confer resistance in tomato against *A. rhizogenes* or *A. tumefasciens*. Also, helpful rhizobacteria like *Pseudomonas fluorescens* and *Bacillus subtilis* have been shown to cause ISR on host plants. Likewise, they provide better protection against crown gall and many other bacterial pathogens. It is because rhizobacteria compete for nutrients, secrete antimicrobial compounds, and stimulate the plant's systemic defence (Calvo *et al.,* 2014; Lastochkina *et al.,* 2019).

The advantages of induced resistance are many. It provides multiple protection by allowing plants to defend against various pathogens without more chemicals. This method cuts down pesticide use and helps floriculture become more sustainable while lowering production costs and environmental impact (Evalon *et al.,* 2024). Moreover, resistance responses that are induced are long lasting, they can last weeks to months and provide permanent protection as opposed to chemical sprays that last for a short time (Pieterse *et al*., 2014). However, there are limitations and challenges. Induced resistance does not always confer complete protection and its effectiveness varies. The impact of the plant genotype, the pathogen strain, and environmental conditions play a key role in this effectiveness. When plants activate their defense pathways too much, it happens at the cost of growth and reproduction and this potentially reduces crop productivity (Walters & Heil, 2007). To use them in the rose growing business, cost effective inducers are required that are effective under field growing conditions. Even though there are many challenges, integrated disease management (IDM) of roses can be a good plan combined with drip irrigation (Evalon *et al.,* 2024).

Application of SA extenally or its functional analogs, such as 2, 6-dichloroisonicotonic acid and benzo-(1, 2, 3)-thiadiazole-7-carbothioic acid S-methyl ester (BTH), induces SAR in plants, resulting in resistance to certain pathogens (Ryals *et al*., 1996; Wang *et al.*, 2005). In addition, the plants expressing the bacterial NahG gene (encoding salicylate hydroxylase, which converts SA to catechol) are more susceptible to several pathogens (Gaffney *et al.*, 1993). Evidence on the role of SA in plant defense comes from the identification and characterization of an Arabidopsis (*Arabidopsis thaliana*) isochorismate synthase (ICS) mutant (sid2-2) which does not biosynthesis SA (Wildermuth *et al*., 2001). Endogenous SA levels in plants can also affect their interaction with symbiotic microorganisms. This is demonstrated by increased root nodulation and infection upon inoculation with *Mesorhizobium loti*, in transgenic *Lotus japonicus* and *Medicago truncatula* expressing NahG (Stacey *et al*., 2006). A limited number of studies have demonstrated the direct effects of SA on microbes including *Pseudomonas aeruginosa*, *Staphylococcus aureus* (Prithiviraj *et al.*, 2005), *Sinorhizobium meliloti* (Martínez-Abarca *et al.*, 1998), and more recently, *A. tumefasciens* (Yuan *et al*., 2007).

2.4. Use of Biostimulants

Biostimulants are new things used in farming. They help plants grow better. They also help the plant deal with tough weather, they also help fight disease. Among the rose production systems under threat from *Agrobacterium tumefasciens*, the use of biostimulants could provide a beneficial added tool to improve plant health and reduce diseases (Opisa, 2021). Biostimulants, in contrast to standard nutrients and fertilizers, help enhance the plant's physiology and metabolic processes. Besides, these help in building the plant's defence mechanism, if necessary, making them more resistant to biotic and abiotic stress. Incorporating them in floriculture management programs as per sustainable agricultural principles minimises chemical use and ensures ecofriendly production (Percival, 2010; Thomson, 2004). Biostimulants can also enhance the quality and marketability of ornamental plants, helping to meet rising local and international demand for sustainably produced roses (Azcon-Aguilar *et al.*, 2002). Biostimulants improve the health of plants, making them less likely to be infected by pathogens. Therefore, they also make yields more stable, even when weather conditions are bad and make it challenging for the plants to grow. These products can support the role of biological control agents which can work together to suppress plant diseases. Biostimulants are an essential part of integrated crop management for sustainable production of flowers (Justine *et al.*, 2023). Biostimulants are formulations of natural origin applied to plants, microbes or the rhizosphere that stimulate naturally occurring processes to improve the uptake of nutrients,

tolerance to stress and crop quality (du Jardin, 2015). Some common categories of biostimulants include microbial biostimulants, which include nitrogen fixing bacteria, plant growth promoting rhizobacteria (PGPR), mycorrhizal fungi and *Trichoderma* spp., which enhance root colonization, nutrient cycling, and induce systemic resistance. Non microbial biostimulants which include seaweed extracts, humic and fulvic acids, amino acids, vitamins, protein hydrolysates, mixed biostimulants which combine microbial and organic compounds adapted for specific crops and environmental conditions. They also often act as signaling molecules to activate plant defense pathways and stimulate secondary metabolite production. (Calvo *et al.,* 2014). These products can be used for normal fertilization, integrated pest control and also provide extra benefits apart from nutrition.Therefore, growers can tailor applications for maximum efficiency and improve crop productivity. They also enhance crop quality and stress resistance. Microbial biostimulants can enhance root colonization and nutrient cycling. In contrast, non-microbial extracts usually function as signalling molecules that activate plant defence pathways (Burr *et al*. 1998). By combining these formulations crop growth and disease resistance is greatly improved. In addition, using biostimulants reduces reliance on costly chemical fertilizers and pesticides which cuts down on costs and pollution of the environment. (Justine *et al.,* 2023). Biostimulants offer various options for improving plant functioning with relevance to diseased rose production. Numerous biostimulants when applied induce ISR and SAR notably enhancing the plant fortification against the pathogens such as *A. tumefasciens* (Battacharyya & Jha, 2012). Humic acids and mycorrhizal fungi help plants take in more nutrients such as nitrogen, combining these formulations provides synergistic effects, promoting robust growth and enhanced disease resistance simultaneously phosphorus and trace elements that improve overall plant health, making them more resistant to infection (Ferrini & Nicese, 2002). Producers designed nutrient formulations using seaweed extracts and amino acids as key components to improve biophysical and biochemical processes in plants for overcoming different abiotic stresses. (Khan *et al.,* 2009). Additionally, beneficial microbes in biostimulant formulations modify the rhizosphere, compete with pathogenic bacteria and fungi, promote root growth and reduce the chance of crown gall infection (Pieterse *et al.,* 2014). Biostimulants impact the hormonal balance of the plant. This improves the ratio of roots to shoots and promotes photosynthetic efficiency. The production of secondary metabolites and phenolic compounds is stimulated enhancing the immunity of the plant. Using biostimulants all the time improves microbial diversity in soil. Thus, it creates a resilient rhizosphere ecosystem that aids the crops in the long term. Together, these methods suggest the use of biostimulants to boost the productivity of modern rose production and protect plants (Du

Jardin, 2015). With their potential to cut down on pesticide application while maintaining high quality of plants, biostimulants are increasingly being adopted in floriculture. Using seaweed extracts can help ornamental crops develop better roots and flower more abundantly. It can also make plants better able to fight off diseases and pests (Craigie, 2011). Protein hydrolysates and humic substances enhance chlorophyll content, promote shoot growth, and increase the strength of plants. In roses, these formulations prove effective (Cola *et al*, 2017). Research on the use of microbial inoculants including *Bacillus subtilis* and *Trichoderma* spp. showed a reduction of severity in crown gall, probably through triggering induced resistance and competitive exclusion with *A. tumefasciens* in the rhizosphere (Hermosa *et al.*, 2012; Lastochkina *et al.,* 2019). The biostimulants help the plants use water more efficiently. They also ensure that the plants incorporate the most amount of nutrients. The timing and the method of application of biostimulant is important as this will affect its efficacy, which means it can be targeted at times of particular stress by foliar sprays or soil drenches. Using biostimulants together with cultural practices such as proper irrigation, pruning and sanitation can amplify their defence activities. Biostimulants are tools that can be used to prevent diseases and help growing along with being highly profitable and environmentally friendly.The advantages of biostimulants are numerous. This makes them ecologically friendly, biodegradable, and environmentally safe for eco-friendly floriculture. These are already in compatible with the existing integrated pest management (IPM). Also, their multifunctional properties facilitate simultaneous assistance in growth, yield, stress tolerance, and disease suppression (Justine *et al.*, 2023). Despite their potential, effectiveness can vary depending on the formulation, dosage, and environmental conditions. There may also be a lack of standardization of commercial products, which can lead to variability (Du Jardin, 2015). Another consideration for adoption is cost effectiveness, as rose producers will require clear economic advantages resulting from this to compete effectively in export markets, particularly Kenya and Ethiopia. Further research is needed to determine the optimal application protocols in terms of frequency and concentration and combinations of microbial and non microbial products. To ensure that biostimulants perform consistently and farmers can rely on them, production must be regulated and controlled for quality. Teaching growers how to use biostimulants correctly and what to expect from their use will encourage adoption and efficacy. Furthermore, assessing the long-term impacts on soil health and microbial communities is necessary in order to integrate biostimulants sustainably in commercial floriculture systems.

The use of biostimulants is expected to grow in the future due to restrictions on synthetic agrochemicals. Current research is being conducted on the preparations of synergistic

formulations of microbial consortia and organic extracts for plant growth and disease development (Rouphael & Colla, 2020). Biostimulants are a viable and sustainable alternative for increasing plant tolerance, improving the overall production quality, and reducing reliance on chemical control for crown gall management. Combining these with cultural, biological, and induced resistance strategies will help maintain healthy, high quality roses under unfavourable production conditions. In the impending future, there may be the development of custom made biostimulant mixtures for different rose types, local formulations adapted to the native soils along with combination products that simultaneously promote stress tolerance and pathogen resistance. Studying how plants react to biostimulants on a molecular level will help in making models that can assess predicted crop performance. If production of these biostimulants increases, it will become more accessible to farmers and growers. Ultimately, biostimulants can become useful aids in the development of sustainable, resilient and profitable floriculture when combined with other integrated management practices (Pieterse *et al.*, 2014).

CHAPTER THREE
MATERIALS AND METHODS

3.1. Study site

The experiments were conducted at Finlay flower farm in Kericho County, Tarakwet farm in Kenya located at 0 ° 27 south / 35° 27 east and 2100 meters above the sea level shown in Figure 1. Average maximum and minimum temperatures range from 19 to 22 ° C, respectively with a total annual rainfall ranging from 2000 to 2500 mm. The main soils are classified as Nitosols characterized with high levels of Aluminum, Iron and Manganese accompanied with low pH ranging from 5.7 – 6.0 and good drainage. These soils are typical of Kenya's floriculture zones around Lake Naivasha and Thika which accounts for over 70 % of national rose production (Kenya Flower Council 2021). The greenhouse used for the study was a polythene covered structure with controlled irrigation and fertigation systems. Environmental conditions; temperature, relative humidity and light intensity were monitored using data loggers to ensure uniformity across treatments.

Legend

Kericho County

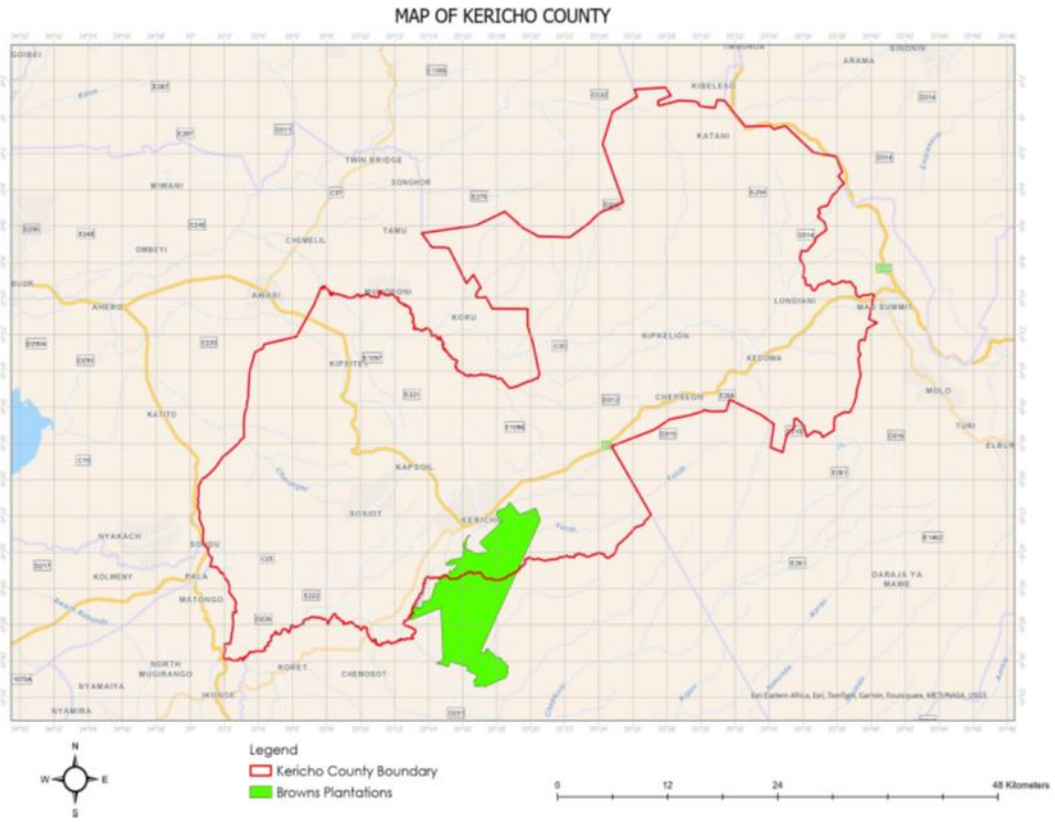


Figure 1. Map of Kenya showing Kericho County and Browns Plantations Kenya LTD where the study was conducted. Source; Applied Research Department, Browns Plantations Kenya LTD former James Finlay Kenya LTD. (GPS coordinates 0.415 º S – 35.3082 º E)

3.2. Testing effect of various biostimulants in suppressing A. tumefasciens in roses

3.2.1. Green house experiment

This study employed a Randomized Complete Block Design (RCBD**)** to evaluate the efficacy of different biostimulants in suppressing *Agrobacterium tumefasciens* infections in roses. RCBD was selected because it is one of the most robust designs for agricultural and horticultural trials, as it effectively minimizes experimental error caused by environmental heterogeneity (Gomez & Gomez, 1984). Greenhouse environments, although controlled, often exhibit micro variations in temperature, humidity, and light intensity across different locations. By blocking these sources of variation and replicating treatments across blocks, the design enhanced the reliability of treatment comparisons and increased the precision of statistical estimates. The methodology was adapted from (Biondi *et al*., 2009), with modifications to fit the greenhouse context of commercial rose production in Kenya. The trial was conducted using the rose variety Tropical Amazon, which is known to be highly susceptible to crown gall. The use of a susceptible cultivar ensured consistent disease pressure across treatments, providing a reliable framework for testing biostimulant efficacy under real world conditions. Each treatment replicate consisted of ten crown galls, all located approximately 15 cm above the pumice based growing medium, which were tagged and monitored throughout the study. Baseline gall diameters were measured using a precision Vernier caliper to establish starting values before treatment application. The tagged galls were then cut using a sterilized roll cut tool. To maintain aseptic conditions and prevent cross contamination, the tool was sterilized after every cut by dipping into a disinfectant solution containing didecyldimethyl ammonium chloride. Such stringent sterilization procedures were essential given the high transmissibility of *A. tumefasciens* through mechanical injury.The experimental period lasted twelve months, allowing observation of both short term and long term effects of biostimulant treatments across seasonal cycles. Weekly measurements were taken to record the diameter of newly developing crown galls on previously tagged stems, using Vernier calipers for accuracy. At the end of each growth cycle, galls were excised and weighed using a servo balance to quantify disease progression in terms of biomass accumulation. In addition, the number of fresh galls emerging on each treatment replicate was recorded monthly. These were also excised, measured, and weighed, ensuring that the dataset captured both incidence (number of new galls) and severity (size and weight of galls).

To assess the agronomic relevance of biostimulant treatments, the trial also monitored yield and quality parameters of harvested rose stems. The total yield of marketable stems was

recorded daily for each treatment replicate over the 12 month period. In addition, a weekly subsample of 30 stems per replicate was subjected to quality assessment. Key parameters included: Stem length, measured with a 60 cm ruler, as longer stems command higher market value, Stem weight, measured using a servo balance, which reflects robustness and postharvest performance, flower head dimensions (length and width), measured with a Vernier caliper, as these characteristics determine visual appeal and vase life. Treatments, as summarized in Table 1, were applied either as foliar sprays or soil drenches depending on the mode of action, at manufacturer-recommended dose rates. Applications were conducted once per month under strictly controlled conditions. The study included 44 plots, each measuring 2 × 1 m^2 and containing 40 rose plants. Guard rows of 1 m were maintained between plots to prevent spray drift and cross-contamination as shown in Figure 2. The Randomised Completely Block Design layout ensured four replicates per treatment, thus enhancing statistical robustness and reducing error variance. The experimental area was physically separated from the rest of the greenhouse to reduce interference and ensure that treatments remained confined to the trial plots. Standard agronomic practices such as weeding, desuckering, pest control were carried out uniformly across all treatments. Other pests and diseases encountered during the trial were controlled with suitable crop protection agents to prevent confounding effects on plant growth and yield. Nutrient supply was standardized across treatments using fertigation, as outlined in Table 2. Macronutrients (N, P, K, Ca, Mg, S) were prepared in Tank A, while micronutrients (Mn, Zn, Cu, B, Mo) were prepared in Tank B, with the exception of iron, which was applied separately due to its tendency to bind with sulfates. Each tank had a capacity of 500 liters, and fertigation was delivered daily at an electrical conductivity (EC) of 1.9 and a pH range of 5.5–6.2. These ranges were maintained to optimize nutrient uptake and simulate commercial greenhouse production standards.

Strict biosafety and containment protocols were observed throughout the experiment to avoid unintentional release of *A. tumefasciens*. Infected plant material was sterilized by autoclaving at 121° C for 20 minutes before disposal. Workers handling diseased material were required to wear gloves, disinfect tools with 70 % ethanol between tasks, and adhere to established phytosanitary guidelines. These measures were necessary to safeguard both experimental integrity and environmental safety. Overall, this experimental setup was designed to provide a comprehensive assessment of biostimulant performance, not only in suppressing crown gall disease but also in sustaining the yield and quality of roses under greenhouse production. By integrating both pathological and agronomic measurements, the study ensured

that findings would be relevant to both scientific research and the practical needs of the floriculture industry.

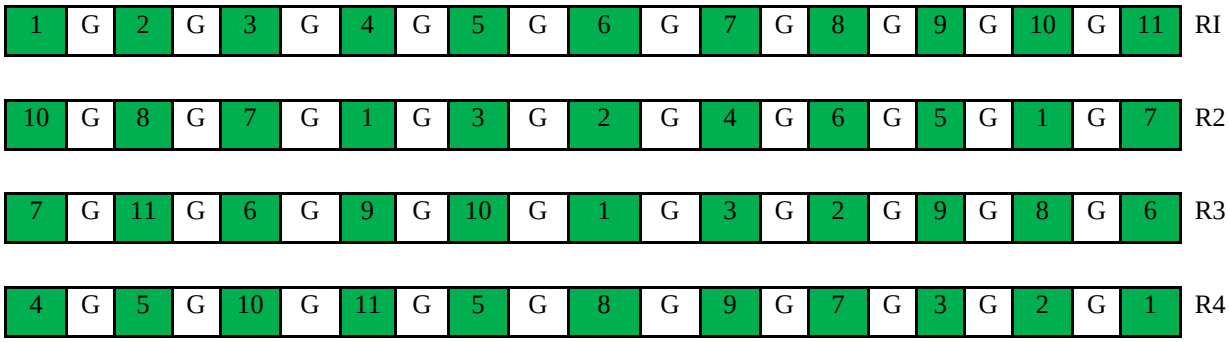


Figure 2: Biostimulants experimental lay out; R1 – R4 replicates, G guard rows, 1-11 randomized plots

Table 1: Various biostimulant treatments, their composition and mode of application

Treatments and Method of application	**composition of various biostimulants**
Control (water only:	spray
Hicure 2.5 ml / L	-Inorganic nitrogen, amino acids, and ashes (drench)
Hicure 2.5 ml / L	- spray
Biozyme 2.5 ml / L	-organic matter from plants, Zinc sulphate and sodium octoborate (spray)
Biozyme 2.5 ml / L	- drench
Foltron 2.0 ml / L	- macro and micronutrients and folycystein (spray)
Foltron 2.0 ml / L	- drench
Codamine radicular 2.0 ml / L	- nitrogen phosphorus, potassium and free amino acids (spray)
Codamine radicular 2.0 ml/ L	- drench
Alexin 2.5 ml / L:	calcium, magnesium, boron, potassium, and salicylic acid (spray)
Alexin 2.5 ml / L	- drench

Table 2: Roses feeding program for macro and micro elements

Fertilizer type	**Quantity (Kg / 500 L)**
Tanks A (per 500 L tank)	
Calcium nitrate	72 kg
Magnesium nitrate	10 kg
Potassium nitrate	10 kg
Micrel iron	1.6 kg
Tank B (per 500 L tank)	
Potassium nitrate	18 kg
Magnesium nitrate	25 kg
MKP	11kg
Potassium sulphate	10 kg
MAP	8 kg
Copper 140 EDTA chelate	60 g
Sodium molybdate	395 g
Manganese 130 EDTA chelate	300 g
Zinc sulphate	100 g
Potassium borate	150 g

3.2.2. Pot trials

A modification of the method described by (Artur *et al*., 2012) was employed for the experimental setup, with adjustments made to suit the local conditions and specific objectives of the study. Two month old rose seedlings, grafted on natal briar (Rosa × natalensis) rootstock and already infected with crown gall disease, were selected as experimental material. The use of grafted seedlings ensured uniformity of growth and increased susceptibility to infection, which provided a reliable model for testing treatments. Before planting, seedlings were visually inspected to confirm the presence of gall symptoms and to exclude any with mechanical injuries or abnormal growth that might introduce bias.The seedlings were grown in plastic containers measuring 30 × 15 × 30 cm, chosen to provide adequate root space and drainage. Each container was filled with a standardized mixture of pumice and tea compost at a ratio of 1:1. This growing medium was selected because pumice offers excellent aeration, drainage, and root support, while tea compost contributes organic matter and beneficial microorganisms that improve soil fertility and microbial balance. The combination was expected to support both healthy plant growth and realistic disease expression, simulating natural production conditions. The media were sterilized before use to minimize contamination by other soilborne pathogens that might interfere with the experiment.

Initial measurements of gall diameter were taken at planting to establish baseline values for each seedling, as described in Section 3.2.1. This step ensured that growth of crown gall tumors during the trial could be accurately monitored and treatment effects reliably compared. A digital caliper was used for tumor diameter measurements to enhance precision, while tumor weight was determined after excision and drying, following standard phytopathological protocols.Treatments, as detailed in Table 1, were applied either as soil drenches or foliar sprays, depending on the mode of action of the respective compounds. Application rates followed manufacturer recommendations or previously validated experimental studies, with modifications where necessary. Treatments were administered consistently to avoid variability, and care was taken to prevent cross contamination between experimental units. Each treatment consisted of three plants, replicated four times, resulting in a total of twelve plants per treatment. Replications were arranged in a Complete Randomized Design (CRD) to minimize positional effects within the greenhouse, such as light intensity or airflow variation, and to strengthen the validity of statistical analysis. Observations were made at regular intervals, and tumor diameter and weight were recorded according to the procedures outlined in Section 3.2.1. Gall size was measured every two weeks to monitor disease progression, while tumor weights

were determined at the conclusion of the experiment to assess cumulative effects of treatments. This dual approach provided both dynamic and final evaluations of treatment efficacy.

Environmental conditions in the greenhouse were monitored to maintain consistency. Temperature was maintained between 22 – 26 ° C during the day and 15–18 ° C at night, while relative humidity was kept at approximately 70 %. These ranges were chosen to mimic the favorable growing conditions for roses in Kenya's highland floriculture zones and to promote consistent disease expression. Supplemental irrigation was provided through drip lines, ensuring uniform water distribution and minimizing leaf wetness, which could otherwise encourage opportunistic infections. Nutrient management followed the rose feeding program outlined in Table 2. The program included balanced supplies of nitrogen, phosphorus, potassium, calcium, magnesium, and micronutrients, which are critical for healthy vegetative growth, flowering, and resilience against stress. By maintaining uniform nutrition across all treatments, differences in tumor development could be more confidently attributed to the experimental treatments rather than nutritional imbalances. Fertilizers were applied through fertigation at recommended intervals to simulate commercial rose production systems. In addition, measures were taken to reduce experimental error. Regular monitoring ensured early detection of any anomalies unrelated to crown gall, such as insect infestations, which were controlled manually to avoid confounding effects of pesticides. Overall, the pot trial methodology was designed to provide a controlled yet realistic system for evaluating the effects of treatments against crown gall disease in roses. The combination of standardized plant material, controlled growing media, rigorous measurement, and systematic design provided a strong foundation for generating reliable and reproducible results.

Plate 2: Pot trial experimental layout for testing biostimulants

3.3. Testing effect of sterilizing agents and pesticides in suppressing crown gall growth

3.3.1. Green house trials

A method described by (Biondi *et al*., 2009) was used. The experiment followed a Randomized Complete Block Design (RCBD**)**, replicated four times to account for variability across the greenhouse microclimate. Randomised Completelyb block Design is considered appropriate in horticultural disease suppression studies because it distributes treatments randomly within blocks, thus minimizing the influence of environmental gradients such as light, humidity, and temperature (Gomez & Gomez, 1984). A total of 28 plots were established, each measuring 2 × 1 m ² and containing 40 rose plants. Guard rows of 1 m were maintained between plots to prevent treatment interference and spray drift, ensuring the integrity of individual treatment responses. Within each treatment replicate, ten crown galls were tagged for longitudinal monitoring. Baseline measurements of gall diameter were taken using a precision Vernier caliper, ensuring accuracy to the nearest 0.1 mm. Tagging specific galls allowed for consistent monitoring of disease progression over time, enabling a more reliable assessment of treatment impact on both gall suppression and regrowth dynamics. To initiate treatment, each gall was carefully cut using a sterilized roll cut tool. Cross contamination was prevented by disinfecting the roll cut in a spore kill solution containing didecyldimethyl ammonium chloride after every cut. This procedure was critical because *A. tumefasciens* is highly transmissible through contaminated equipment, and sterilization maintained experimental rigor while preventing artificial spread of the pathogen

The trial compared seven treatments, chosen to represent a combination of conventional, botanical, and alternative approaches to crown gall suppression:

1. Isacop 50 WP (copper oxychloride, 1.0 g / L) – a widely used fungicide-bactericide with known efficacy against bacterial pathogens. Copper based formulations are standard in crown gall management but raise concerns regarding phytotoxicity and soil accumulation.
2. Fresh Fry (vegetable oil, undiluted) – applied as a physical barrier treatment. Oils can disrupt pathogen colonization on wound sites and may also reduce secondary infections
3. Agrowipe (botanic neem extract, undiluted) – neem is recognized for its broad-spectrum antimicrobial and insecticidal properties, attributed to active compounds such as azadirachtin and nimbin.
4. Dettol (chloroxylenol 4.8 %) – tested at 0.5 ml / L and 1.0 ml / L dilutions. Chloroxylenol is a widely used disinfectant with antibacterial properties, included here as a low-cost alternative treatment option.

5. Hydrogen peroxide (50 %, 1.0 ml / L) – an oxidative agent capable of disinfecting wound sites by destroying bacterial cells. Its rapid breakdown into water and oxygen makes it attractive for sustainable use but requires careful handling due to phytotoxic risks.
6. Control (water only) – untreated wounds were included to provide baseline comparisons against natural disease progression.

To ensure precise and consistent application, treatments were applied monthly following wounding. The method of application varied depending on the physical properties of the treatment: Vegetable oil and neem extract were viscous in nature and were therefore applied using a paintbrush, which ensured direct coverage and adhesion onto wound sites. Hydrogen peroxide, chloroxylenol (both concentrations), copper oxychloride, and water were applied using a 1.5 L hand sprayer (Hardi Ltd., Kenya). The sprayer allowed for uniform distribution of liquid treatments across wound surfaces and minimized operator bias. Greenhouse conditions such as temperature, humidity, and ventilation were monitored daily to maintain an environment conducive to crown gall development, ensuring sufficient disease pressure for treatment evaluation. The use of a greenhouse setting also reduced the influence of external weather variability, thereby increasing experimental precision. Overall, this greenhouse trial was carefully designed to balance scientific rigor, biosafety, and commercial relevance. By including both synthetic and botanical treatments, the study aimed to generate insights into sustainable disease management strategies for roses, providing a foundation for future field trials and potential integration into integrated pest management (IPM) systems.

3.3.2. Pot experiment

To monitor the growth of crown gall tumours closely, a pot trial was conducted. A method described by (Artur *et al*., 2012), was used for the experiment. Rose seedlings grafted on natal briar root stock variety tropical amazon already infected with crown gall were used for the experiment. The seedlings were grown in plastic containers measuring (30 x 15 x 30 cm) filled with pumice, inert material as the growing media. The initial diameter of the galls was measured using a Vernier calliper. The galls were cut as described above in the greenhouse experiment and similar treatments applied at the rates shown. Treatments comprised of 3 plants replicated four times with replicates arranged in a complete randomized design (CRD). Tumour diameter was measured as described in section 3.3.1 (a).

3.3.3. Data collection

Fresh crown gall growths were counted and removed once a month for a period of twelve months. Tumour diameter of the tagged galls was also measured once a month for a period of twelve months. Mean tumour weight of the fresh galls was determined after 12 months by cutting and weighing them using servo weighing scale model SB 3000 from Servo Balans Ltd. Yield was determined by counting total number of marketable stems from each treatment replicate daily for a period of twelve months. The quality of stems produced was determined by sampling 30 stems once a week from each treatment replicate and the following parameters measured; stem length (using ruler), stem weight (using servo weighing balance), flower heads size was measured using a Vernier calliper. Other pests and diseases were controlled when encountered and other agronomic operations such as weeding, removal of blind shoots and bloomers were carried throughout the experimental period.

3.4. Testing effect of *Bacillus subtilis* and *Trichoderma asperellum* in suppressing *Agrobacterium tumefasciens*

3.4.1. Green house trial

A modification of the method described by (Biondi *et al.*, 2009), was used where a susceptible rose variety Tropical amazon already infected with crown gall tumours was used for the experiment. Ten galls were tagged on each treatment replicate and the initial diameter of the galls measured using a Vernier caliper. The galls were cut as described in section 3.1.1. The following treatments were drenched after every two weeks; *Trichoderma asperellum* at 0.25 g / L and 0.5 g / L (provided by Dudutech Kenya LTD in Kenya, application rates also recommended by Dudutech K LTD) and *Bacillus subtilis* at 0.2 ml / L and 0.4 ml / L (provided by Real IPM Company LTD in Kenya, application rates also recommended by Real IPM Company LTD). The treatments were compared with control plots treated with water only. Treatments were replicated four times with replication arranged in a randomized complete block design (RCBD) giving a total of 25 plots. Each plot comprised 40 plants measuring 2 x 1 m^2 with a guard row of 1 m^2 between plots. The number and size of fresh galls was measured as described in section 3.2.1.Total yield and quality of rose stems were measured as described in section 3.2.1.

3.4.2. Pot trials

To monitor the growth of crown gall tumours closely, a pot trial was conducted. A method described by (Artur *et al*., 2012), was used for the experiment. Rose seedlings grafted on natal briar root stock variety tropical amazone already infected with crown gall were used for the experiment. The seedlings were grown in plastic containers measuring (30 x 15 x 30 cm) filled with pumice. The initial diameter of the galls was measured using a Vanier calliper. The galls were cut as described in the greenhouse trial. The following treatments were applied; *Trichoderma asperellum* at 0.25 g / L and 0.5 g / L and *Bacillus subtilis* at 0.2 ml / L and 0.4 ml / L. The treatments were compared with control plots drenched with water only. Treatments comprised of three plants replicated four times with replicates arranged in a complete randomized design (CRD). Tumour diameter was measured as described in section 3.4.1

3.5. Testing effect of selected pesticides in controlling *Agrobacterium tumefasciens*

3.5.1. Pot trials

The method described by (Biondi *et al*., 2009), was used for the experiment. Rose seedlings from a variety of tropical amazon grafted on natal briar root stock artificially inoculated with *A. tumefasciens* was used for the experiment. The seedlings were grown in plastic containers measuring (30 x 15 x 30 cm) filled with a mixture of pumice and tea compost as the growing media. The following treatments were drenched twice a week; enrich BM (bronopol 27 % w / w) 2 g / L and 4 g / L, previcur energy (propamocab hydrochloride plus fosetyl aluminum) 2.0 ml / L and 4.0 ml / L twice a week. Treatments were compared with control plots treated with water only. Mean tumour diameter was measured as described in section 3.2.1. Total number of fresh galls were also counted once a month for a period of 12 months. Agronomical parameters such as stem length, weight and head size were measured as described in section 3.2.1.

3.5.2. Green house experiment

A method described by (Biondi *et al*., 2009) was used with a greenhouse planted with rose variety tropical amazon already infected with crown gall tumour. Ten galls were tagged on each treatment replicate and were measured as described in section 3.2.1. Treatments described in experiment 3.4.1 were used and data collected as described in experiment 3.5.1.

3.6. Data analysis

All the data obtained from various experiments was subjected to analysis of variance (ANOVA) and means separated using Duncan's multiple range test at 5 % level. Mstat statistical package version 2.10 developed by Prof. Russell Freed of Michigan State University, Crop and Soil Science department was used for analysis.

CHAPTER FOUR

RESULTS

4.1. Effect of various biostimulants in suppressing *Agrobacterium tumefasciens* in roses

4.2. Green house experiment

4.2.1. Yield of roses

The results of this experiment are summarized in Table 3. Statistically, significant differences ($P \leq 0.05$) were observed in the yield of marketable roses between the plots sprayed with biozyme (2.5 ml / L**)** and the control plots that were only sprayed with water. Application of biozyme at 2.5 ml / L resulted in a notably higher yield of marketable flowers, representing an overall increase of 14 % compared to the untreated control. This demonstrates the effectiveness of biozyme in enhancing plant productivity and resilience under experimental conditions. In addition to biozyme, other biostimulants were also tested, and although the differences among treatments were not statistically significant ($P \leq 0.05$), notable trends were observed. For instance, plots treated with hicure (2.5 ml / L drench**)** showed a 7.7 % increase, while those treated with hicure (2.5 ml / L spray) exhibited a 4.4 % increase in yield compared to the control. Similarly, biozyme drench (2.5 ml / L) contributed a 3.2 % increase, whereas **f**oltron spray (2.0 ml / L) resulted in yield increments of 10.3 % in one trial and 2.6 % in another. Treatments with codamine radicular spray (2.0 ml / L) and codamine drench improved yields by 2.0 % and 6.5 %, respectively. The most remarkable effects, apart from biozyme, were recorded for alexin, where spraying at 2.5 ml / L improved yields by 12.5 %, while drenching enhanced yields by 13.4 %.Although not all treatments reached levels of statistical significance, the general trend across all the biostimulant applications indicated positive effects on rose yield relative to the untreated control.

Table 3: Mean yield of roses and percentage increase on plots treated with various biostimulants (pooled data for one year)

Treatments	Mean Yield (no)	Percentage increase
Control (water only (spray)	441.8 b*	0.0
Hicure 2.5 ml / L (drench)	475.8 ab	7.7
Hicure 2.5 ml / L (spray)	460.3 ab	4.0
Biozyme 2.5 ml / L (spray)	514.8 a	14.0
Biozyme 2.5 ml / L (drench)	456.5 ab	3.2
Foltron 2.0 ml / L (spray)	492.5 ab	10.3
Foltron 2.0 ml / L (Drench)	455.3 ab	2.6
Codamine radicular 2.0 ml / L (spray)	450.3 ab	2.0
Codamine radicular 2.0 ml/ L (drench)	470.3 ab	6.5
Alexin 2.5 ml / L (spray)	496.8 ab	12.6
Alexin 2.5 ml / L (drench)	500.3 ab	13.4
LSD value	61.5	

*Means within a column followed by the same letter are not significantly different by Duncan's multiple range test at 5 % level

4.2.2. Fresh galls, diameter and weight of crown gall tumours

Results on the efficacy of various biostimulants on *A. tumefasciens* that is biozyme 2.5 ml / L, hicure 2.5 ml / L, codamine radicular 2.0 ml / L, foltron 2.0 ml / L and alexin 2.5 ml / L either sprayed or drenched on fresh galls, gall weight, and diameter are represented in Table 4. No significant differences ($P \leq 0.05$) were observed in the number new galls, gall weight and gall diameter on plots treated with biozyme 2.5 ml / L, hicure 2.5 ml / L, codamine radicular 2.0 ml / L, foltron 2.0 ml / L and alexin 2.5 ml / L either sprayed or drenched and control plots sprayed with water only.

Table 4: Mean fresh galls, gall diameter and gall weight on plots treated with various biostimulants (pooled data for one year)

Treatments	Mean New galls (no)	Mean gall diameter (cm)	Mean Gall weight (g)
Control (water only (spray)	246.0 a	4.2 a	23.4 a
Hicure 2.5 ml / L (drench)	179.8 a	3.8 a	23.0 a
Hicure 2.5 ml / L (spray)	171.3 a	5.1 a	26.1 a
Biozyme 2.5 ml / L (spray)	206.3 a	4.0 a	23.5 a
Biozyme 2.5 ml / L (drench)	245.8 a	4.1 a	28.5 a
Foltron 2.0 ml / L (spray)	166.3 a	3.4 a	26.6 a
Foltron 2.0 ml / L (Drench)	170.5 a	4.1 a	25.2 a
Codamine radicular 2.0 ml / L (spray)	222.8 a	5.3 a	29.0 a
Codamine radicular 2.0 ml/ L (drench)	216.0 a	3.9 a	25.3 a
Alexin 2.5 ml / L (spray)	219.8 a	3.0 a	27.2 a
Alexin 2.5 ml / L (drench)	223.8 a	3.4 a	26.1 a
LSD value	64.9	1.12	1.54

*Means within a column followed by the same letter are not significantly different by Duncan's multiple range test at 5 % level

4.2.3. Flower quality- stem length, weight head length and width

The outcomes of this experiment are presented in Table 5. Significant differences ($P \leq 0.05$) were recorded in several parameters that define flower quality, including stem length, stem weight, and the dimensions of the flower head. Plots treated with biozyme (2.5 ml / L), hicure (2.5 ml / L), codamine radicular (2.0 ml / L), foltron (2.0 ml / L), and alexin (2.5 ml / L) whether applied through foliar spray or drenching consistently outperformed the untreated control plots. Compared to roses treated with water only control conditions, treated plots produced stems that were on average longer by at least 8 cm. Similarly, treated plants yielded stems that were heavier by approximately 7 g, suggesting an improvement in biomass accumulation and structural robustness. The flower heads also exhibited measurable improvements, with head length and width increasing by about 3 mm relative to the control. Such improvements are of considerable commercial significance, since stem length and flower head size are key determinants of market value in the floriculture industry, particularly for export oriented rose varieties. The results further indicate that biostimulants enhance physiological processes linked to vegetative growth and reproductive development. Longer and heavier stems may be attributed to improved photosynthetic efficiency, better nutrient uptake, and enhanced hormonal balance induced by biostimulant application. Larger flower heads, on the other hand, suggest an influence on cell expansion and flower meristem development. Interestingly, no significant differences were observed between the two methods of application i.e. spraying and drenching. This suggests that the active ingredients in the tested biostimulants are effective regardless of the application method, providing flexibility for growers to choose the technique that best fits their production system. For instance, drenching may be more suitable where root zone stimulation is desired, while spraying can be more practical for foliar absorption and canopy-level protection.

Table 5: Mean stem length, head length, head width and stem weight on plots treated with various biostimulants (pooled data for one year)

Treatments	Mean Stem Length (cm)	Mean head length (cm)	Mean head width	stem weight (cm) (g)
Control (water only (spray)	50.0 b*	4.13 c *	2.3 b *	21.4 b*
Hicure 2.5 ml / L (drench)	58.2 a	4.21 ab	2.7 a	28.5 a
Hicure 2.5 ml / L (spray)	63.0 a	4.26 a	2.8 a	28.3 a
Biozyme 2.5 ml / L (spray)	60.0 a	4.23 ab	2.8 a	28.8 a
Biozyme 2.5 ml / L (drench)	57.1 a	4.25 a	2.6 a	28.7 a
Foltron 2.0 ml / L (spray)	57.4 a	4.23 ab	2.6 a	28.5 a
Foltron 2.0 ml / L (Drench)	58.1 a	4.23 ab	2.7 a	29.6 a
Codamine 2.0 ml / L (spray)	57.5 a	4.16 bc	2.8 a	28.8 a
Codamine 2.0 ml/ L (drench)	57.4 a	4.25 a	2.7 a	28.5 a
Alexin 2.5 ml / L (spray)	58.3 a	4.21 ab	2.9 a	28.6 a
Alexin 2.5 ml / L (drench)	58.4 a	4.26 a	2.8 a	28.5 a
LSD value	1.5	0.1	0.08	1.77

*Means within a column followed by the same letter are not significantly different by Duncan's multiple range test at 5 % level

4.3. Pot experiment

The results of the pot experiment are presented in Table 6, and they provide additional insights into the potential role of biostimulants in mitigating crown gall disease. Significant differences ($P \leq 0.05$) were observed in the number of newly formed galls across treatments. Specifically, plots treated with hicure drench (2.5 ml / L) exhibited the lowest incidence of new gall formation, while treatments with foltron drench (2.0 ml / L) and codamine radicular drench (2.0 ml / L**)** also reduced gall formation, though to a lesser extent. The ability of hicure to reduce the number of new galls indicates that it may possess properties that limit pathogen proliferation or enhance the plant's natural defense mechanisms. Biostimulants such as hicure are known to contain amino acids, peptides, or signaling molecules that can act as elicitors of plant defense responses, priming the plant to resist further infections.However, no significant differences ($P \leq 0.05$) were observed in gall diameter or gall weight across treatments, including biozyme, foltron, codamine radicular, alexin (both sprayed and drenched), and the untreated control. This suggests that while biostimulants may help reduce the initiation of new infections, they have limited impact on the growth of existing galls once they are established. From a biological perspective, this aligns with the nature of *Agrobacterium tumefacien***s**, the pathogen responsible for crown gall disease, which integrates T-DNA into the plant genome and permanently alters cellular behavior, making it difficult to reverse the tumorous growth once initiated.

Table 6: Mean new galls, gall diameter and gall weight on plots treated with various biostimulants (pooled data for one year)

Treatments	Mean New galls (no)	Mean gall diameter (cm)	Mean Gall weight (g)
Control (water only (spray)	6.0 bc	2.3 a	33.4 a
Hicure 2.5 ml / L (drench)	5.0 c	4.3 a	33.0 a
Hicure 2.5 ml / L (spray)	9.5 abc	4.9 a	36.1 a
Biozyme 2.5 ml / L (spray)	6.8 abc	3.1 a	33.5 a
Biozyme 2.5 ml / L (drench)	11.3 ab	5.5 a	38.1 a
Foltron 2.0 ml / L (spray)	5.5 bc	2.1 a	36.0 a
Foltron 2.0 ml / L (Drench)	12.0 a	4.4 a	32.2 a
Codamine radicular 2.0 ml / L (spray)	8.3 abc	3.9 a	39.0 a
Codamine radicular 2.0 ml/ L (drench)	8.0 abc	3.4 a	35.3 a
Alexin 2.5 ml / L (spray)	8.0 abc	3.9 a	37.2 a
Alexin 2.5 ml / L (drench)	7.8 abc	2.3 a	33.1 a
LSD value	5.22	2.97	2.51

*Means within a column followed by the same letter are not significantly different by Duncan's multiple range test at 5 % level

4.4. The effect of Agrowipe (botanic neem extract), Fresh fri (vegetable oil) sterilizing agents and pesticides in controlling *Agrobacterium tumefaciens*

Results on the effect of copper oxychloride, Fresh fri (vegetable oil), Agrowipe (botanic neem extract), Dettol (chloroxelenol 4.8 %) and hydrogen peroxide 50 % on new galls, tumour weight and tumour diameter are represented in Tables 8, 9, 10 and 11. There were significant differences ($P \leq 0.05$) on the yield and quality of marketable rose stems on plots treated with vegetable oil (undiluted), botanic neem extract (undiluted) and copper oxychloride at 1.0 g / L. Plots treated with the botanic neem extract and the vegetable oil had a higher yield of marketable roses as shown in Table 7 with better quality as shown in Table 9. They were longer and heavier compared to plots treated with copper oxychloride 1.0 g / L and untreated control. No significant differences were observed in the yield of roses on Plots treated with

chloroxylenol 4.8 % 0.5 ml / L, 1.0 ml / L and hydrogen peroxide 1.0 ml / L, vegetable oil, botanic neem extract, copper oxychloride at 1.0 g / L and untreated control. Results also show that undiluted vegetable oil and botanic neem extract effectively inhibited growth of crown gall tumours and fresh galls compared to copper oxychloride 1.0 g / L and untreated control plots at ($P \leq 0.05$).

4.4.1. Yield of marketable stems

There were significant differences ($P \leq 0.05$) on the yield of marketable rose stems on plots treated with vegetable oil (undiluted), botanic neem extract (undiluted) and copper oxychloride at 1.0 g / L. Plots treated with the botanic neem extract and the vegetable oil had a higher yield of marketable roses as shown in Table 7.

Table 7: Mean yield of marketable stems on plots treated with copper oxychloride, vegetable oil, botanic neem extract, chloroxylenol 4.8 %, hydrogen peroxide and untreated control on yield of roses (pooled data for one year)

Treatments	Yield of marketable stems (no)
Isacop 50 WP (copper oxychloride) 1.0 g/L	446.0 b*
Fresh fri (vegetable oil) –undiluted	510.8 a
Agrowipe (botanic neem extract –undiluted	508.0 a
Dettol (chloroxylenol 4.8 %) 0.5 ml / L	491.3 ab
Dettol (chloroxylenol 4.8 %) 1.0 ml / L	471.5 ab
Hydrogen peroxide 50 % 1.0 ml / L	500.5 ab
Untreated control	495.0 ab
LSD value	61.6

*Means within a column followed by the same letter are not significantly different by Duncan's multiple range test at 5 % level

4.4.2. Diameter, weight and fresh crown gall tumours

Results showed that, undiluted vegetable oil and botanic neem extract effectively inhibited growth of crown gall tumours and fresh galls compared to copper oxychloride 1.0 g / L and untreated control plots at ($P \leq 0.05$) as shown in Table 8. Dettol and hydrogen peroxide moderately inhibited growth of crown gall.

Table 8: Effect of copper oxychloride, vegetable oil, botanic neem extract, chloroxylenol 4.8 %), Hydrogen peroxide and untreated control on crown gall tumours (pooled data for one year)

Treatments	Mean gall diameter (cm)	Mean gall weight (g)	Mean new galls
Isacop 50 WP (copper oxychloride) 1.0 g/L	10 a*	36.0 a	415.3 a
Fresh fri (vegetable oil) –undiluted	0.5 e	1.5 b	272.2 b
Agrowipe (botanic neem extract –undiluted	0.2 e	0 b	240.0 b
Dettol (chloroxylenol 4.8 %) 0.5 ml / L	7.8 bc	16.5 ab	360.8 ab
Dettol (chloroxylenol 4.8 %) 1.0 ml / L	9.1 bc	4.3 b	465.0 ab
Hydrogen peroxide 50 % 1.0 ml / L	7.0 bc	2.8 b	328.3 ab
Untreated control	11.7 a	10 ab	434.0 a
LSD value	3.25	25.8	228.1

*Means within a column followed by the same letter are not significantly different by Duncan's multiple range test at 5 % level

4.4.3. Quality of rose stems

Significant differences ($P \leq 0.05$) were observed on the quality of marketable rose stems on plots treated with vegetable oil (undiluted), botanic neem extract (undiluted) and copper oxychloride at 1.0 g / L. Plots treated with Agrowipe - botanic neem extract and Fresh fri - the vegetable oil had a better quality as shown in Table 9. They were longer and heavier compared to plots treated with copper oxychloride 1.0 g / L and untreated control. No significant difference ($P \leq 0.05$) was observed on the quality of rose stems treated with Agrowipe - botanic neem extract and Fresh fri - the vegetable oil, Dettol at 0.5 ml / L, 1.0 ml / L and hydrogen peroxide at 1.0 ml / L.

Table 9: Effect of copper oxychloride), vegetable oil), botanic neem extract, chloroxylenol 4.8 %), hydrogen peroxide and untreated control on quality of roses variety tropical amazone (pooled data for one year)

Treatments	Mean Stem length (cm)	Mean head size (cm)	Mean stem weight (g)
Isacop 50 WP (copper oxychloride) 1.0 g/ L	54.0 a *	3.4 b	23.0 a
Fresh fri (vegetable oil) –undiluted	59.0 b	4.1ab	28.5 b
Agrowipe (botanic neem extract –undiluted	59.0 b	4.1ab	28.3 b
Dettol (chloroxylenol 4.8 %) 0.5 ml / L	59.0 b	4.3a	27.3 b
Dettol (chloroxylenol 4.8 %) 1.0 ml / L	57.5 b	4.3 a	28.3 b
Hydrogen peroxide 50 % 1.0 ml / L	58.8 b	4.3 a	28.5 b
Untreated control	55.2 a	3.3 b	23.3 a
LSD value	2.18	0.2	3.15

*Means within a column followed by the same letter are not significantly different by Duncan's multiple range test at 5 % level

4.4.4. Pot experiment - mean tumour diameter and tumour weight

Results for the pot experiment are shown in Table 10. Results show that fresh fri - undiluted vegetable oil and agrowipe - botanic neem extract effectively inhibited growth of crown gall tumours and fresh galls compared to copper oxychloride 1.0 g / L, Dettol 0.5 ml / L, 1.0 ml / L and untreated control plots at ($P \leq 0.05$). Results are also shown in Plates 3, 4, 5, 6, 7, 8 and 9.

Table 10: Effect of copper oxychloride), vegetable oil, agrowipe botanic neem extract, chloroxylenol 4.8 %, hydrogen peroxide and untreated control on crown gall tumours rose variety tropical amazone

Treatment	Mean gall diameter (cm)	Mean gall weight (g)
Isacop 50 WP (copper oxychloride) 1.0 g/L	14.7 a*	36.0 a*
Fresh fri (vegetable oil) –undiluted	3.1 bc	1.5 b
Agrowipe (botanic neem extract –undiluted	0.9 c	0 b
Dettol (chloroxylenol 4.8 %) 0.5 ml / L	8.1 ab	16.5 ab
Dettol (chloroxylenol 4.8 %) 1.0 ml / L	8.9 ab	14.3 ab
Hydrogen peroxide 50 % 1.0 ml / L	6.2 ab	12.8 ab
Untreated control	8.3 ab	10.0 ab
LSD value	8.67	25.9

*Means within a column followed by the same letter are not significantly different by Duncan's multiple range test at 5 % level

Plate 3: (a) Crown gall treated (b) after cutting gall (c) gall at 12 months.
With copper oxychloride
1.0 g / L before cutting gall.

Plate 4: (a) Crown gall treated with (b): after cutting gall (c) gall at 12 months
fresh fri before cutting gall

Plate 5: (a) Crown gall treated with (b) after cutting gall (c) gall at 12 months
Agrowipe before cutting gall .

Plate 6: (a) Crown gall treated with dettol (b) after cutting gall (c) gall at 12 months.
0.5 ml / L before cutting gall



Plate 7: (a) Crown gall treated with dettol 1.0 ml / L before cutting gall. (b) after cutting gall (c) gall at 12 months.



Plate 8: (a) Crown gall treated with hydrogen peroxide 1.0 ml / L before cutting gall (b) after cutting gall (c) gall at 12 months.



Plate 9: (a) crown gall treated With water only control before cutting gall (b) after cutting gall (c) gall at 12 months

4.5. Effect of selected fungicides in controlling *Agrobacterium tumefasciens*

4.5.1. Field experiment – flower quality (stem length and head size)

Results on the efficacy of previcur energy 2.0 and 4.0 L / Ha and enrich BM 2.0 and 4.0 Kg / Ha and untreated control in controlling *Agrobacterium tumefasciens* are shown in Table 11. No significant differences at ($P \leq 0.05$) were observed in the flower quality (stem length and head size) on plots treated with previcur energy at 2.0 and 4.0 L / Ha, enrich BM 2.0 and 4.0 Kg / Ha and untreated control.

Table 11: Mean stem length, head length and head width on plots treated with previcur energy, enrich BM and control (water only)

Treatments	Mean Stem Length (cm)	Mean Head length (cm)	Mean width (cm)
Previcur energy 2.0 L / Ha	57.5 a	2.20 a	2.19 a
Previcur energy 4.0 L / Ha	57.7 a	2.19 a	2.24 a
Enrich BM 2.0 Kg / Ha	57.2 a	2.22 a	2.21 a
Enrich BM 4.0 Kg / Ha	56.9 a	2.21 a	2.21 a
Control (water only)	56.7 a	2.20 a	2.22 a
LSD value	1.7	0.14	0.08

*Means within a column followed by the same letter are not significantly different by Duncan's multiple range test at 5 % level

4.5.2. Mean yield and stem weight

Results on the efficacy previcur energy 2.0 and 4.0 L / Ha, enrich BM 2.0 and 4.0 Kg/ Ha and untreated control in controlling *Agrobacterium tumefasciens* are shown in Table 12. No significant differences at ($P \leq 0.05$) were observed in the yield of roses and stem weight among these treatments.

Table 12: Mean Yield and mean stem weight on plots treated with previcur energy, L Enrich BM and control (water only)

Treatments	Mean Yield (no)	Mean stem weight (g)
Previcur energy 2.0 L / Ha	316.5 a	27.5 a
Previcur energy 4.0 L / Ha	367.2 a	28.3 a
Enrich BM 2.0 Kg / Ha	302.2 a	28.5 a
Enrich BM 4.0 Kg / Ha	303.2 a	27.0 a
Control (water only)	328.8 a	27.7 a
LSD value	2.1	1.84

*Means within a column followed by the same letter are not significantly different by Duncan's multiple range test at 5 % level

4.5.3. Mean fresh galls and gall diameter

Results on the efficacy of previcur energy 2.0 L / Ha and 4.0 L / Ha, enrich BM 2.0 and 4.0 Kg / Ha and untreated control in suppressing crown gall growths in the field are shown in Table 13. No significant differences at ($P \leq 0.05$) were observed in the fresh galls and gall diameter among these treatments.

Table 13: Mean fresh gall and gall diameter on plots treated with Previcur energy, Enrich BM and control (water only)

Treatments	Mean Fresh galls (no)	Mean gall diameter (g)
Previcur energy 2.0 L / Ha	89.0 a	8.6 a
Previcur energy 4.0 L / Ha	61.8 a	5.0 a
Enrich BM 2.0 Kg / Ha	79.8 a	4.3 a
Enrich BM 4.0 Kg / Ha	68.5 a	6.3 a
Control (water only)	76.5 a	5.1 a
LSD value	1.92	1.48

*Means within a column followed by the same letter are not significantly different by Duncan's multiple range test at 5 % level

4.6. Pot experiment

4.6.1. Mean fresh galls and gall diameter

Results on the efficacy of previcur energy 2 and 4 L / Ha, enrich BM 2.0 and 4.0 Kg / Ha and untreated control in suppressing crown gall growths in the field are shown in Table 14, Plates 10,11,12,13 and 14. No significant differences at ($P \leq 0.05$) were observed in the fresh galls and gall diameter among these treatments in the pot experiment.

Table 14: Mean fresh gall and gall diameter on plots treated with Previcur energy, Enrich BM and control (water only) – pot experiment

Treatments	Mean Fresh galls (no)	Mean gall diameter (g)
Previcur energy 2.0 L / Ha	12.0 a	5.7 a
Previcur energy 4.0 L / Ha	10.0 a	5.6 a
Enrich BM 2.0 Kg / Ha	9.5 a	3.8 a
Enrich BM 4.0 Kg / Ha	7.8 a	3.8 a
Control (water only)	7.0 a	3.6 a
LSD value	6.167	1.86

*Means within a column followed by the same letter are not significantly different by Duncan's multiple range test at 5 % level

Plate 10: (a) Crown gall treated with (b) after cutting gall (c) gall at 12 months previcur energy 3 ml / L before cutting gall

Plate 11: (a) Crown gall treated with (b) after cutting gall (c) gall at 12 months Previcur energy 4 ml / L before cutting gall

Plate 12: (a) Crown gall treated with (b) after cutting gall (c) gall at 12 months enrich bm 2 g / L before cutting gall

Plate 13: (a) Crown gall treated with enrich BM 4 g / L before cutting gall (b): after cutting gall Plate (c) gall at 12 months

Plate 14: (a) crown gall treated with water only before cutting gall (b) after cutting gall (c) gall at 12 months

4.7. Effect of selected biological agents in controlling *Agrobacterium tumefasciens*

4.7.1. Field trial - mean production and stem weight

Results on the efficacy of *B. subtilis*, 0.2 L / Ha and 0.4 L / Ha and *T. asperellum* 0.25 Kg / Ha and 0.5 Kg / Ha on yields and stem weight in the field are shown in Table 15. No significant differences at ($P \leq 0.05$) were observed among these treatments.

Table 15: Mean production and stem weight in plots treated with *B. subtilis, T. asperellum* and control (water only)

Treatments	Mean Production (no)	Mean stem weight (g)
Bacillus subtilis 0.2 L / Ha	485.8 a	27.4 a
Bacillus subtilis 0.4 L / Ha	502.5 a	27.7 a
Trichoderma asperellum 0.25 Kg / Ha	528.8 a	27.2 a
Trichoderma asperellum 0.5 Kg / Ha	512.2 a	27.4 a
Control (water only)	491.8 a	26.4 a
LSD value	127.5	1.29

*Means within a column followed by the same letter are not significantly different by Duncan's multiple range test at 5 % level

4.7.2. Fresh galls and gall diameter

Results on the efficacy of *B. subtilis* 0.2 L / Ha and 0.4 L / Ha, *T. asperellum* 0.25 Kg / Ha and 0.5 Kg / Ha and control (water only) in suppressing crown gall growths are shown in Table 16. No significant differences at ($P \leq 0.05$) were observed among these treatments.

Table 16: Mean fresh galls and gall diameter in plots treated with *B. subtilis*, *T. asperellum* and control (water only)

Treatments	Mean number of Fresh galls (no)	Mean gall diameter (cm)
Bacillus subtilis 0.2 L /Ha	111.5a	4.1 a
Bacillus subtilis 0.4 L / Ha	104.5 a	3.3 a
Trichoderma asperellum 0.25 Kg / Ha	103.5 a	4.0 a
Trichoderma asperellum 0.5 Kg / Ha	106.5 a	4.6 a
Control (water only)	105.0 a	3.6 a
LSD value	1.51	1.19

*Means within a column followed by the same letter are not significantly different by Duncan's multiple range test at 5 % level

4.7.3. Stem length and head size

Results on the efficacy of *B. subtilis*, 0.2 L / Ha and 0.4 L / Ha and *T. asperellum* 0.25 Kg / Ha and 0.5 Kg / Ha on the quality of roses harvested are shown in Table 17. No significant differences at ($P \leq 0.05$) were observed in the quality of roses among these treatments.

Table 17: Mean stem length and head size in plots treated with *B. subtilis*, *T. asperellum* and control (water only)

Treatments	Mean Stem length (cm)	Mean head length (cm)	mean head width (cm)
Bacillus subtilis 0.2 L /Ha	56.6 a	4.2 a	2.4 a
Bacillus subtilis 0.4 L / Ha	55.3 a	4.1 a	2.4 a
Trichoderma asperellum 0.25 Kg / Ha	56.4 a	4.1 a	2.3 a
Trichoderma asperellum 0.5 Kg / Ha	56.2 a	4.0 a	2.2 a
Control (water only)	55.1.a	4.1 a	2.3 a
LSD value	1.36	0.08	0.08

*Means within a column followed by the same letter are not significantly different by Duncan's multiple range test at 5 % level

4.8. Pot trials

Results on the effect of *B. subtilis*, 0.2 L / Ha and 0.4 L / Ha, *T. asperellum* 0.25 Kg / Ha and 0.5 Kg / Ha and control (water only) in suppressing crown gall growths are shown in Table 18, Figure 15, Plates 15,16, 17, 18 and 19. Significant differences at $P \leq 0.05$ were observed on plots treated with these treatments. Plots treated with *B. subtilis*, 0.2 L / Ha and 0.4 L / Ha, *T. asperellum* 0.25 Kg / Ha and 0.5 Kg / Ha suppressed crown galls compared to control plots treated with water only. However, no significant differences ($P \leq 0.05$) were observed on the new crown gall growths after 12 months.

Table 18: Mean initial gall diameter, gall diameter at 12 months and fresh galls on plots treated with *B. subtilis*, *T. asperellum* and control (water only)

Treatments	Initial Gall diameter (cm)	Gall diameter at 12 months (cm)	Fresh galls (no)
Bacillus subtilis 0.2 L / Ha	4.6 a	0.0 b	0.0 a
Bacillus subtilis 0.4 L / Ha	4.1 a	3.0 b	0.6 a
Trichoderma asperellum 0.25 Kg / Ha	3.1 a	0 b	1.6 a
Trichoderma asperellum 0.5 Kg / Ha	2.1 a	0.0 b	0.3 a
Control (water only)	4.0 a	16.7 a	1.7 a
LSD value	1.19	10.92	2.43

*Means within a column followed by the same letter are not significantly different by Duncan's multiple range test at 5 % level.

Plate 15: (a) Crown gall treated with *B.subtilis* 2 ml / L before cutting gall (b) after cutting gall (c): gall at 12 months

Plate 16: (a) Crown gall treated with *B subtilis* 4 ml / L before cutting gall (b) after cutting gall (c) gall at 12 months

Plate 17 (a) Crown gall treated With *T. asperellum* 0.25 g / L before cutting gall (b) after cutting gall (c) gall at 12 months

Plate 18: (a). Crown gall treated With T. *asperellum* 0.5 g / L before cutting gall (b) after cutting gall (c) gall at 12 months

Plate 19: (a) Crown gall treated With control (water only) before cutting gall (b) after cutting gall (c) gall at 12 months

CHAPTER FIVE
DISCUSSIONS

The findings of this study can be placed in a broader agronomic context through earlier work on different crops in different situations, which show the multifaceted roles of these agents. Not only do these have direct biological effects on pathogens, they impact soil structure, nutrient cycling, and plant–microbe interactions which ultimately elevate their role in integrated pest and disease management systems. The application also contributes to international initiatives aimed at reducing dependence on synthetic agrochemicals, cutting back on environmental contamination, health risks and resistance development, while furthering productivity with lower ecological costs. With climate change we need cropping systems that are resilient, and the systems should preferably be of native crops. The short term outcomes of reduced incidence of diseases are certainly great, but the long term ones are even greater. These include environmental sustainability, economic viability of farmers, and increased resilience of agricultural systems. Hence, making the case for biological and ecological approaches of sustainable agriculture.

5.1. Effect of selected biostimulants in suppressing *Agrobacterium tumefasciens* in roses

The application of biostimulants such as Biozyme (2.5 ml / L), Hicure (2.5 ml / L), Codamine Radicular (2.0 ml / L), Foltron (2.0 ml / L), and Alexin (2.5 ml / L), either as foliar sprays or soil drenches, demonstrated a significant improvement in both the yield and quality of roses even when plants were challenged with *Agrobacterium tumefasciens* infections. These findings provide strong evidence that the incorporation of biostimulants into rose cultivation systems can play a dual role: not only in reducing the negative effects associated with crown gall disease but also in promoting the general health and vigor of rose plants (Rouphael & Colla, 2015). The benefits of these products stem largely from their multifaceted mechanisms of action, which include enhancing nutrient use efficiency, stimulating hormonal pathways involved in growth and defense, and increasing plant tolerance to both biotic stresses such as pathogens and abiotic stresses such as drought, salinity, or temperature extremes (Du Jardin, 2015). Biostimulants are known to enhance physiological processes at the cellular and biochemical levels. They function by improving the uptake of essential nutrients, increasing chlorophyll content, and activating plant enzymatic systems. These processes collectively enhance photosynthetic activity, energy assimilation and biomass accumulation. For example, in leafy vegetable crops, the application of biostimulants has been reported to increase pigment

composition, including chlorophylls and carotenoids, which are critical for efficient light capture and photosynthetic activity. This increase translates into greater biomass accumulation and improved overall productivity (Justine *et al*., 2023). In floricultural systems, similar trends have been documented, where biostimulant use has been shown to accelerate both vegetative and reproductive growth phases. This acceleration is particularly important for commercial cut flower industries because it shortens production cycles, ensures a more predictable harvest and optimizes resource use efficiency in greenhouse production systems (Rouphael & Colla, 2020).

Among the biostimulants tested, Biozyme is one of the most extensively studied and widely used. Research evidence highlights its ability to enhance plant growth and productivity across different crop categories. For instance, in guava, Biozyme application improved plant vigor, fruit set, overall yield and postharvest fruit quality, particularly in terms of sugar content and physicochemical properties (Sayan *et al*., 2015). Similarly, in pears, Biozyme treatment significantly enhanced sugar accumulation in fruits (Gasanova & Gyul-Aakhmedov, 1992). Earlier studies in apples (*Malus domestica*) also reported the pro yield effects of micronutrient based biostimulants on both fruit yield and quality, further validating the consistency of Biozyme's effects across diverse fruit crops (Inomata *et al*., 1992). The utility of biostimulants extends beyond fruit crops into the domain of ornamentals, where they are gaining increasing prominence. Research on ornamental species such as *Antirrhinum majus* (snapdragon), *Capsicum annuum* (ornamental pepper), and *Eustoma grandiflorum* (lisianthus) revealed that biostimulant application resulted in greater vegetative vigor, extended stem length, and superior flower quality (Mondal *et a*l., 2015; Nahed *et al.,* 2009; Paradikovic *et al.*, 2011). Moreover, integrated treatments combining organic amendments like bioslurry with biostimulants such as Hicure were particularly effective in carnations, leading to improved stem length and larger flower heads, two critical parameters determining the commercial value of cut flowers (Niyokuri *et a*l., 2017). These findings underline the wide applicability and effectiveness of biostimulants across horticultural systems.

Importantly, biostimulants are increasingly recognized as sustainable alternatives or complementary tools to conventional fertilizers and pesticides. Unlike chemical inputs that often provide immediate but short lived benefits, biostimulants operate through long term modulation of plant physiological processes. They strengthen plant growth and reproduction while enhancing stress resilience and reducing the ecological footprint of crop production. This aligns with global agricultural strategies that are steadily moving away from heavy dependence on synthetic agrochemicals and embracing ecological or agroecological approaches to farming (Calvo *et al.,* 2014). In the context of roses affected by *Agrobacterium tumefasciens*,

biostimulants can improve plant resistance by positively influencing soil plant interactions. These interactions facilitate nutrient cycling, enhance soil microbial diversity and improve soil health, creating a more balanced agroecosystem that suppresses pathogen proliferation.

In conclusion, the results of this study emphasize that biostimulants significantly improve rose performance under pathogen stress conditions, particularly crown gall disease. The improvements may be linked to the activation of both physiological and biochemical defense mechanisms, enabling plants to tolerate infections more effectively. While these results are promising, there is still a need for further research to clarify the precise mechanisms of action of different biostimulants and to determine whether their effects are sustained over multiple growth cycles. Additionally, the economic feasibility and cost effectiveness of incorporating biostimulants into large scale commercial rose production must be carefully assessed. Nevertheless, the integration of biostimulants represents an important step toward creating floriculture systems that are not only economically viable but also environmentally sustainable and resilient to biotic challenges such as crown gall disease.

5.2. Effect of sterilizing agents, agrowipe, Fresh fri and fungicide in suppressing crown gall growth

Considerable research attention has been directed towards evaluating sterilizing agents and botanical alternatives as potential substitutes for synthetic agrochemicals in the management of crown gall disease in roses. Crown gall, caused by *Agrobacterium tumefasciens*, is notoriously difficult to manage due to the pathogen's ability to integrate its DNA into plant genomes, leading to the development of tumor like growths on stems and roots. Traditional reliance on chemical pesticides has proven to be costly, environmentally hazardous, and in some cases ineffective, thus creating a pressing need for alternative, sustainable disease management strategies. One of the products tested in this study, Agrowipe, demonstrated high effectiveness against *Agrobacterium tumefasciens*. Agrowipe is formulated from a mixture of African herbal trees, seeds, shrubs, and weeds, which are known to contain diverse bioactive compounds with antimicrobial activity (Mitali *et al.,* 2012). These bioactive compounds include alkaloids, terpenoids, flavonoids, and phenolic compounds that inhibit the growth and proliferation of bacterial and fungal pathogens. In particular, neem (*Azadirachta indica*) extracts, which form a key ingredient in many biocontrol formulations, have long been recognized for their rich repertoire of secondary metabolites such as azadirachtin, nimbin, and salannin. These metabolites are potent inhibitors of microbial growth and disrupt the metabolic

pathways of pathogens, thereby reducing disease incidence (Isman, 2006). Similarly, vegetable oils such as Fresh Fri were observed to have inhibitory effects on crown gall development. Their mode of action is largely physical, with oils creating a coating around nematodes or microbial cells, leading to suffocation and disruption of respiration (Maina *et al.*, 2013). This mechanical mode of action makes them relatively low risk in terms of resistance development compared to synthetic chemicals. In the present study, vegetable oils prevented gall formation by interfering with crown gall activity, which in turn reduced secondary infections and tumor initiation.

Contrary to these promising results, the application of copper oxychloride, a widely used copper based fungicide, not only failed to inhibit gall formation but paradoxically enhanced both the size of tumors and the number of galls. This outcome suggests that certain fungicides may create conditions that inadvertently favor pathogen activity or weaken plant defense responses. Such paradoxical effects are not unique to roses. For example, Kairu *et al.*, (1984) reported that captafol sprays enhanced the growth of *Pseudomonas syringae* on coffee, while Oniang'o *et al.,* (2005) demonstrated that copper oxychloride increased the growth of *Phomopsis thea* on tea under laboratory conditions. These findings underscore the complexity of plant pathogen chemical interactions and highlight the risk of unintended consequences when broad spectrum fungicides are applied to sensitive cropping systems like floriculture. The study also revealed that treatments with hydrogen peroxide (H_2O_2) at 1.0 ml / L and chloroxylenol at concentrations of 0.5 ml / L and 1.0 ml / L provided moderate inhibition of crown gall development. Hydrogen peroxide is well documented in the literature for its dual role as both an antimicrobial compound and a signaling molecule in plant defense responses (Alvarez *et al.,* 1998; Barker & Orlandi, 1995). As a reactive oxygen species (ROS), H_2O_2 can directly kill pathogenic microbes through oxidative stress while simultaneously acting as a signal to activate systemic acquired resistance (SAR) in plants. It promotes the strengthening of plant cell walls through the modification of hydroxyproline rich glycoproteins, thereby making tissues less permeable to pathogen invasion (Anderson *et al.,* 1998). Additionally, hydrogen peroxide is known to trigger localized programmed cell death or hypersensitive responses (HR), which confine the pathogen to infected cells and prevent systemic spread (Auh & Murphy, 1995). However, the erratic performance of hydrogen peroxide in this study suggests that the concentration and timing of application are critical determinants of its effectiveness. Excessive levels of H_2O_2 may cause phytotoxicity, while suboptimal levels may fail to suppress pathogen activity effectively.

The adoption of botanical formulations and sterilizing agents is increasingly driven by concerns about the environmental and health impacts of synthetic pesticides. Chemical pesticides are often associated with high economic costs, toxicity risks to non-target organisms, and long term soil degradation. In contrast, plant based formulations such as neem extracts and vegetable oils offer safer, eco friendly alternatives that can be incorporated into integrated disease management programs (Reimers *et al.*, 1993; Varshney, 2001). Over the last three decades, research has confirmed the utility of botanicals as part of sustainable pest and disease management strategies, aligning with global efforts to reduce reliance on synthetic chemicals. Nevertheless, while antibiotics and synthetic bactericides may provide short term suppression of bacterial diseases, their widespread use is discouraged due to the high risk of resistance development in pathogen populations (McManus & Stockwell, 2001). This makes botanicals, essential oils, and natural oils an attractive alternative for long-term use. Neem-based formulations such as Agrowipe and edible oils like Fresh Fri represent practical tools for integrated pest management (IPM), combining safety, efficacy, and sustainability. However, further work is required to standardize the concentration of active ingredients, evaluate their efficacy under diverse agroecological conditions, and assess cost-effectiveness in commercial floriculture systems.

In conclusion, the findings of this study reinforce the potential of sterilizing agents and plant based formulations in suppressing crown gall disease in roses. Products such as Agrowipe and Fresh Fri demonstrated promising results by reducing gall development through biochemical and physical mechanisms, respectively. Meanwhile, hydrogen peroxide and chloroxylenol provided moderate but significant inhibition by leveraging plant defense pathways. However, the paradoxical effects observed with copper oxychloride underscore the importance of careful evaluation of chemical inputs in disease management programs. Overall, integrating sterilizing agents and botanicals into rose production systems offers a safer, more sustainable, and environmentally friendly pathway toward managing crown gall and reducing dependency on synthetic fungicides.

5.3. Effect of *Bacillus subtilis* and *Trichoderma asperellum* in controlling *Agrobacterium tumefasciens*

The application of beneficial microorganisms, particularly *Bacillus subtilis* and *Trichoderma asperellum*, has emerged as a promising component of sustainable crop protection strategies. These organisms belong to a class of biological control agents that function through diverse mechanisms, ranging from direct antagonism against pathogens to the

enhancement of plant defense responses. Despite their potential, the results of field trials in this study showed no statistically significant differences in rose yield, stem quality, or gall suppression between microbial treatments and untreated controls. This limited effectiveness under commercial greenhouse conditions can be partly attributed to the reduced viability and colonization potential of biopesticides when used concurrently with chemical pesticides, which are commonly applied for managing other pests and diseases (Bashan *et al.*, 2014). The performance of microbial inoculants is highly variable and influenced by several environmental factors such as temperature, humidity, soil microbiome composition and the persistence of pesticide residues (Mala *et al.,* 2012). These factors often reduce the consistency of microbial biocontrol outcomes in open-field and greenhouse conditions.

Interestingly, results from controlled pot experiments indicated that both *B. subtilis* and *T. asperellum* were effective in suppressing crown gall growth. This finding highlights the fact that under favorable conditions, where external interfering factors such as pesticide residues and fluctuating environmental parameters are minimized, microbial inoculants can exert strong biocontrol activity. Thus, optimization of application conditions is crucial to unlocking the full potential of microbial agents in rose production systems. *Bacillus subtilis* is among the most extensively studied plant growth promoting rhizobacteria (PGPR) and is widely recognized for its antagonistic activity against plant pathogens. Its biocontrol mechanisms are multifaceted. One of its key features is the production of cyclic lipopeptides such as surfactin, iturin, and fengycin, which have strong antifungal and antibacterial properties (Chen *et al.*, 2009; Ongena & Jacques, 2008). These metabolites disrupt the cell membranes of pathogens, thereby inhibiting their growth. In addition, *B. subtilis* competes effectively for ecological niches by colonizing root surfaces, where it forms biofilms that provide plants with a protective shield against pathogens while simultaneously promoting root development (Wang *et al.*, 2018).

Multiple studies have validated the efficacy of *B. subtilis* against diverse plant diseases. For example, Abbasi and Weselowski (2015) and Tan *et al*., (2013) demonstrated that *B. subtilis* strains could suppress bacterial wilt in tomatoes caused by *Ralstonia solanacearum* and bacterial spot in tomatoes caused by *Xanthomonas* species. These effects were achieved through biofilm formation and the production of antimicrobial metabolites. Moreover, *B. subtilis* strains have consistently shown antagonistic activity against a wide range of soilborne fungal pathogens, including *Fusarium* spp., *Rhizoctonia solani*, *Sclerotinia rolfsii*, and *Verticillium dahliae*, in both greenhouse and field settings (De Curtis *et al.*, 2010; Kumar *et al.*, 2012; Li *et al*., 2013). Beyond disease suppression, *B. subtilis* is also known to improve

soil health by enhancing microbial diversity and nutrient cycling, thereby creating more resilient agroecosystems (Mercado-Flores *et al.,* 2014).

Trichoderma species are among the most commercially significant fungal biocontrol agents, accounting for more than 60 % of biofungicides registered worldwide (Verma *et al*., 2007). *Trtchoderma asperellum* is particularly noted for its diverse modes of action. These include antibiosis, achieved through the secretion of secondary metabolites that inhibit pathogen growth; mycoparasitism, where *Trichoderma* hyphae coil around those of pathogens and secrete hydrolytic enzymes to degrade them; competition, where it outcompetes pathogens for nutrients and space; and induction of systemic resistance, where it activates the host plant's innate defense mechanisms (Harman *et al.,* 2004; Vinale *et al.,* 2008). Beyond direct pathogen suppression, *Trichoderma* spp. also confer additional agronomic benefits, including improved nutrient uptake, enhanced plant vigor, and increased tolerance to abiotic stresses such as drought and salinity (Vessey, 2003). These attributes make *Trichoderma* not only a biocontrol agent but also a plant growth promoter. Recent studies reinforce its broad utility. For instance, Pandey *et al*., (2016) reported that *Trichoderma* formulations were effective against powdery mildew (*Erysiphe pisi*) in peas, while Kumar *et al.,* (2015) demonstrated suppression of stem gall disease in coriander through seed treatment and foliar sprays of *Trichoderma* suspensions. The current study supports these findings by confirming the suppressive effect of *T. asperellum* against crown gall in roses under controlled conditions.The observed discrepancies between pot experiments and field trials underscore a common challenge in the commercialization of microbial biocontrol agents. While promising under laboratory and controlled conditions, their performance often declines in field or greenhouse environments where multiple uncontrolled variables interfere with microbial viability and activity. Key areas requiring attention include the development of improved formulation technologies that extend shelf life, compatibility studies with commonly used pesticides, and the refinement of application timing to ensure optimal colonization and activity (Harman *et al.,* 2004).

In conclusion, the combined use of *Bacillus subtilis* and *Trichoderma asperellum* presents a promising approach to sustainable crown gall management in roses and other floricultural crops. Both organisms exhibit strong antagonistic activity and additional plant growth promoting benefits. However, realizing their full potential requires addressing critical limitations such as formulation stability, chemical compatibility, and environmental variability. With further refinement and integration into Integrated Disease Management (IDM) systems, microbial inoculants like *B. subtilis* and *T. asperellum* can become powerful tools for reducing dependence on chemical pesticides and promoting resilience in rose production systems

5.4. Effect of Previcur energy and enrich BM in controlling *Agrobacterium tumefasciens*

The results of this study showed that *Agrobacterium tumefasciens* infections in roses were not effectively controlled by Enrich BM at concentrations of 3.0 g / L and 4.0 g / L, nor by Previcur Energy at 2.0 ml / L and 4.0 ml / L. This negative result implies that despite their efficacy against other classes of pathogens, these chemicals exhibit little or no activity against the crown gall pathogen. Enrich BM is marketed as a broad spectrum immunomodulator and plant defense activator with a primarily prophylactic mode of action. Its beneficial effects are thought to occur through stimulation of systemic resistance in plants rather than through direct antimicrobial activity. According to product literature from Osho Chemicals (2018), Enrich BM "enhances the plant's resistance to a broad spectrum of bacterial and fungal pathogens by activating its natural defense pathways." These defense pathways resemble those triggered by systemic acquired resistance (SAR), mediated by signaling molecules such as salicylic acid, jasmonic acid, and ethylene, which lead to upregulation of defense related genes and production of pathogenesis related (PR) proteins and phytoalexins (Durrant & Dong, 2004; Pieterse *et al.*, 2014). However, the persistence of *A. tumefasciens* infection indicates that such induced resistance may not be sufficiently robust to counteract the unique tumor-inducing strategy of the pathogen. *A. tumefasciens* causes disease by transferring T-DNA (tumor inducing DNA) from its Ti-plasmid into host plant cells, permanently reprogramming cell metabolism and growth to form galls (Escobar & Dandekar, 2003). Because this transformation is stable and internal, external defense activation may not be enough to prevent or reverse tumor formation. Recent reviews and trials reinforce the general challenge of managing crown gall once infection is established. A recent evaluation of chemical and biological products for crown gall control highlights the limited efficacy of many chemical agents and emphasizes that preventive measures and biological antagonists are usually more successful (Öksel *et al.*, 2024).

Previcur Energy is a fungicide produced by Bayer, containing two active ingredients: propamocarb hydrochloride (530 g / L) and fosetyl-aluminum (310 g / L). Propamocarb is primarily active against oomycete pathogens such as *Phytophthora*, *Pythium*, by inhibiting spore germination and mycelial growth (Bayer Crop Science, 2019). Fosetyl-aluminum (FAL) displays a dual function; a modest direct action on pathogen invasion and an indirect enhancement of plant defenses via apoplastic release of phosphonate derivatives, which trigger defense signaling pathways within the host (Petre *et al.,* 2015). Because *A. tumefasciens* is a bacterial pathogen with a distinct mode of infection and disease development (i.e., DNA integration and tumor formation), these fungicidal modes of action are not well suited to control it. The poor performance of Previcur Energy observed here aligns with findings in other pathosystems. For instance, Emil *et al*., (2007) demonstrated that propamocarb hydrochloride failed to control *Verticillium* wilt in pepper, reflecting its restricted effectiveness to particular pathogen classes. Furthermore, though fosetyl aluminium has, in some contexts, shown protective effects against bacterial pathogens such as lowering susceptibility to *Erwinia amylovora* or limiting *Pseudomonas syringae* infection), these outcomes are highly dependent on pathogen species, crop, and mode of action compatibility.A broad review of crown gall management notes that most chemical and biological agents tested for crown gall control exhibit inconsistent results across different host species, and their field performance is often limited (Bayer Crop Science, 2019).

In the face of limited chemical efficacy, recent work has emphasized biological control and antagonistic strains as more reliable options. For example, a 2024 study on endophytic bacteria showed that cell free culture supernatants from certain bacterial isolates were able to suppress crown gall development in plants, pointing to novel microbial metabolites with potential activity against *A. tumefasciens* (Etminani *et al.*, 2024). Moreover, in grapevine systems, nonpathogenic *Allorhizobium vitis* (synonym of some *Agrobacterium* types) strains such as ARK-1 have been highly effective in reducing crown gall incidence in field trials, outperforming the traditional K84 strain in meta analyses (ARK-1 risk reduction to at least 6 % of control. A new biopesticide based on ARK-1 is currently being developed and has shown positive results in field trials in mixed crop settings (Kawaguchi *et al.,* 2023). Additionally, a recent news release describes a novel biocontrol strain, GTI-5813, derived from modification of *A. vitis* F2/5, being trialed in vineyards as a dip treatment to suppress crown gall infection, representing a potential next-generation biological control agent. Another relevant line of work is in exploring the "gallobiome" or the microbial communities associated with crown gall tumors. A 2024 study profiled the bacterial microbiota in rose galls, offering insights into how

the local microbiome might influence disease progression or suppression (University of Adelaide, 2024)

Given the lack of effectiveness of Enrich BM and Previcur Energy against *A. tumefasciens*, relying on these products alone is unjustified in the context of crown gall disease, especially considering added costs and potential non target ecological disturbances (e.g., suppression of beneficial microbial communities). As McManus *et al.*, (2002) argued, indiscriminate use of chemicals can drive resistance and harm beneficial organisms. Rather than discarding these products entirely, future research should explore whether their application is more effective as preventive treatments for instance applied before pathogen invasion rather than curative. Combining them with robust biological control agents such as ARK-1, *B. subtilis* and *Trichoderma* spp. or botanical compounds (neem, essential oils) may produce synergistic effects, particularly in integrated pest management (IPM) frameworks. Given the advances in antagonistic strain development, such as ARK-1 and GTI-5813, these biological tools may surpass chemical elicitors in managing crown gall under realistic field conditions. In summary, while Enrich BM and Previcur Energy retain value for managing certain fungal or bacterial diseases, they are not viable solutions for crown gall disease in roses. The future lies in refining microbial biocontrol agents, improving formulation and delivery methods, understanding microbiome interactions and integrating cultural practices to sustainably manage *A. tumefasciens* in floriculture systems.

CHAPTER SIX
CONCLUSIONS AND RECOMMENDATIONS

6.1. Summary

The floriculture sector is seriously constrained by crown gall disease, which is caused by *Agrobacterium tumefasciens*. In Kenya, the most important export crop is roses. The illness charecterised by swelling like tumours in stems and roots. Which, causes loss of vigour and poor stem quality. In severe cases, it may cause 100 % loss in the yield of affected rose varieties. A study of integrated management practices was carried out at James Finlay Kenya Ltd in Kericho for three years. These included biostimulants such as (Foltron, Biozyme, Alexin, Hicure, Codamine radicular), sterilizing agents (Dettol, hydrogen peroxide, neem extract, vegetable oil), biological agents (*Trichoderma asperellum* and *Bacillus subtilis*) and selected chemical pesticides like copper oxychloride, Previcur Energy, and Enrich BM.

The results indicated that biostimulants significantly improved yield and stem quality, being more effective when sprayed rather than drenched. Experiments have shown that the extract of neem and vegetable oil effectively suppressed crown gall (which is caused by the bacteria *Agrobacterium tumefasciens*). Also, neem extract and vegetable oil proved far superior to a few fungicides such as copper oxychloride which not only could not suppress gall but promoted gall under certain conditions. Biological control agents such as *B. subtilis* and *T. Asperellum* showed strong suppression of *A.tumefasciens*. The research showed effectiveness of biocontrol using *Agrobacterium tumefaciens* under controlled conditions, but less effective under field conditions probably interfacing pesticides and environmental factors. On the other hand, Previcur Energy and Enrich BM were not able to control crown gall in both field and pot tests, these results suggest limited relevance for bacterial disease. In conclusion, the utilization of biologicals, biostimulants and plant extracts can be beneficial in reducing diseases sustainably. The results align with global trends advocating for lower use of chemicals in the flower and agricultural industry for environmentally safe and effective long terms (Lamichhane *et al.,* 2017; Pretty & Bharucha, 2015).

6.2. Conclusions

i. All tested biostimulants enhanced rose yield and stem quality, with foliar sprays performing better than drenches. This underscores the role of biostimulants in improving plant vigor, resilience, and productivity beyond their traditional use as supplements.

ii. Agrowipe (neem extract) and Fresh Fri (vegetable oil) effectively suppressed *A. tumefasciens,* offering eco friendly alternatives to copper based fungicides. The

ineffectiveness of copper oxychloride and its observed stimulation of gall formation confirm reports from other crops that copper may induce pathogen proliferation under certain conditions.

iii. Previcur energy at 2.0 and 4.0 ml / L and enrich BM at 2.0 and 4.0 g / L did not control *Agrobacterium tumefasciens*. There was no increase in the yield and quality of roses under both the field conditions and controlled environment.

iv. *Bacillus subtilis* at 0.2 ml / L and 0.4 ml / L and *Trichoderma asperellum* 0.25 g / L and 0.5 g / L effectively controlled crown gall in controlled environments but not under field conditions. Their performance confirms their potential as sustainable alternatives, though application strategies must address compatibility with agrochemicals and environmental variability.The study demonstrates that no single treatment provides complete control of crown gall. Instead, integrated use of plant extracts, microbial agents, and biostimulants provides the best outcomes in terms of yield, quality, and disease suppression

6.3. Recommendations

i. Biozyme should be applied as a spray and not drenched to achieve good results. Alexin can either be sprayed or drenched on roses. Both methods are suitable. Foltron should be sprayed and not drenched. Both Codamine radicular and hicure should be drenched and not sprayed. More research is needed to elucidate which mechanism is involved in improving the quality and yield when various biostimulants are applied.

ii. Agrowipe and fresh fri should be adopted for use to control *Agrobacterium tumefasciens* in roses. The use of copper oxychloride in controlling *A. tumefasciens* should be discouraged because of its iatrogenic effects (increases incidence of crown gall). Further research should be conducted to establish if agrowipe and fresh fri can still control *A. tumefasciens* at diluted levels. This will help to reduce the overall cost of application. Further research on hydrogen peroxide and dettol should be done test various application rates since they are known to be effective anti bacterial agents.

iii. For previcur energy and enrich BM, more trials should be done to determine if they can be used as preventive applications instead of curative application of *Agrobacterium tumefasciens*. Applications can start early immediately after planting.

iv. *Bacillus subtillis* and *Trichoderma asperellum* should be applied with compatible pesticides which do not reduce their efficacy. Their use should be encouraged since they are also safe to the user and environment compared to chemical pesticides. They can also be used in organic farming. More research should also be done to identify the

mechanisms involved in controlling / suppressing plant disease and effective timing of application to enhance their efficacy.

6.4. Suggestions for further research

i. **Mechanistic and molecular investigations of biostimulants**

Transcriptomic, proteomic, or metabolomic studies should be conducted to elucidate how biostimulants such as Biozyme, Foltron, Alexin, Codamine radicular, and Hicure influence plant defense pathways, nutrient uptake, or hormonal signaling in roses under *Agrobacterium tumefasciens* infection or attack. Further research should also be conducted on whether combinations of biostimulants with microbial agents provide synergistic effects, this is informed informed by recent work showing enhanced outcomes from microbial consortia in plant health management.

ii. **Extended field trials of plant extracts over multiple seasons and regions**

Neem extract (Agrowipe) and vegetable oil (Fresh Fri) should be tested in a variety of rose cultivars and across different agroecological zones in Kenya and East Africa for consistency, phytotoxicity risks, and cost benefit analysis under commercial farm settings. In addition diluted levels tested and alternative formulations or carriers investigated such as nano emulsions to reduce costs while maintaining efficacy.

iii. **Broader screening of natural antimicrobial agents**

Hydrogen peroxide, Dettol, and other low toxicity antimicrobials should be evaluated over a gradient of concentrations and application timings. Also impacts on beneficial soil microbiota should be studied and monitored. The use of novel or under utilized biochemical such as oligosaccharides, fine proteins as reported in other rose-crown gall studies

iv. **Preventive versus curative use of chemical and biological agents**

Experiments should be designed comparing preventive application at planting and early vegetative stage as compared to curative treatments of Previcur Energy and Enrich BM. For example, recent work on *Agrobacterium radiobacter* strains in rose suggests strong preventive suppression of crown and root galls at least 85 % reduction in trials (Oksel *et al.,* 2024).

v. **Improvement of biological control under field conditions**

There should be focus on carrier formulations such as encapsulation, biochar, polymer matrices and stabilization strategies to enhance survival and persistence of *Bacillus*

subtilis, *Trichoderma asperellum*, or other antagonists in field soil. Combinations of biocontrol agents should be tested to broaden spectrum and reduce competition or interference. The biocontrol of crown gall in grapevine using *Bacillus* strains is a promising. Induction of induced systemic resistance (ISR) should be evaluated or priming of plant immunity by microbial agents, as demonstrated in cherry tree–*Agrobacterium* systems where *Bacillus strains* upregulated jasmonic acid, ethylene, and salicylic acid gene patways (Li *et al*., 2025). Integrated management systems should be developed and tested combining biostimulants, plant extracts, biocontrol agents, and cultural practices such as sanitation and soil amendments. A cost benefit analysis should be done and scalability assessments for smallholder versus large-scale rose farms, modeling adoption thresholds and risks.

vi. **Pathogen diversity, genomics, and resistance screening in Roses**

Molecular and genomic profiling of *A. tumefasciens* strains isolated from Kenyan rose farms should be conducted to identify virulence genes, population structure, and variation in resistance to treatments. The information should be used to guide targeted biocontrol strategies and breeding for resistance. In silico approaches for identifying quorum sensing inhibitors (QSIs) have also been Propsed as novel mitigation strategies

vii. Newly engineered biocontrol strains or gene edited variants such as modified *Agrobacterium* strains lacking necrosis or pathogenicity, as is being trialed in grapevines in the University of Adelaide. Quorum sensing disruptors, such as phage therapy, or CRISPR-based suppression of pathogenic genes should also be investigated.

REFERENCES

Abbasi, P. A., & Weselowski, B. (2015). Efficacy of *Bacillus subtilis* QST 713 formulations, copper hydroxide, and their tank mixes on bacterial spot of tomato. *Crop Protection*, *74*, 70 - 76. https://doi.org/10.1016/j.cropro.2015.04.009.

Ádám, A., Bestwick, C., Barna, B., & Mansfield, J. (1995). Enzymes regulating the accumulation of active oxygen species during the hypersensitive reaction of bean to *Pseudomomas syringae*, *Pseudomonas pv.phaseolicola*, *Planta*, *197*(2). https://doi.org/10.1007/bf00202643

Adeola, O., Meru, A. K., & Kinoti, M. W. (2019). Kenya's blooming flower industry: Enhancing global competitiveness. *Africa's Competitiveness in the Global Economy*, 331-349. https://doi.org/10.1007/978-3-319-67014-0_13

African Development Bank Group. (2013). *Annual report*. African Development Bank. https://www.afdb.org

Agriculture Institute. (2025). *Rose cultivation: Best practices for growing roses for cut flowers*. https://agriculture.institute/floriculture-and-landscaping/rose-cultivation-best-practices/

Agricultural Marketing Resource Center. (n.d.). *Floriculture*. Agricultural Marketing Resource Center | Agricultural Marketing Resource Center. https://www.agmrc.org/commodities-products/specialty-crops/floriculture

Agrios, G. N. (1997). *Plant pathology* (4th ed.). Academic Press.

Agrios, G. N. (2005). *Plant pathology* (5th ed.). Academic Press.

Ahmad, M., Nadeem, S. M., Naveed, M., & Zahir, Z. A. (2016). Potassium-solubilizing bacteria and their application in agriculture. *Potassium Solubilizing Microorganisms for Sustainable Agriculture*, 293-313. https://doi.org/10.1007/978-81-322-2776-2_21

Alan, W. (2021). *Rose Diseases: Identification and Management*. Department of Plant Sciences. https://plantsciences.tennessee.edu/wp-content/uploads/sites/25/2021/11/UTExtension-Rose-diseases-Identification-and-management-W833.pdf

Akansha, S., Nandini, S., Kabadwal, B., Tewari, A., & Kumar, J. (2018). Review on plant-*Trichoderma*- Pathogen interaction. *International Journal of Current Microbiology and Applied Sciences*, *7*(2), 2382-2397. https://doi.org/10.20546/ijcmas.2018.702.291

Alsup, C. M. (2004). Screening for active ingredients in plant extracts that inhibit the growth of *Agrobacterium tumefasciens*. Plant Health Instructor.*The Plant HealtInstructor*. https://doi.org/10.1094/phi-i-2004-

Alvarez, M. E., Pennell, R. I., Meijer, P., Ishikawa, A., Dixon, R. A., & Lamb, C. (2020).

Reactive oxygen intermediates mediate a systemic signal network in the establishment of plant immunity. *Cell*, *92* (6), 773-784. https://doi.org/10.1016/s0092-8674(00)81405-1

Amayi, M. K. (2021). Flower growing firms' contribution to community water use management in Naivasha Sub County, Kenya. *Environmental Challenges*.*5*, 100330.

Anderson, N. O. (Ed.). (2006). Flower breeding and genetics: issues, challenges and opportunities for the 21st century.

Anderson, M. D., Chen, Z., & Klessig, D. F. (1998). Possible involvement of lipid peroxidation in salicylic acid-mediated induction of PR-1 gene expression. *Phytochemistry*, *47*(4),555-566. https://doi.org/10.1016/s0031 9422(97)00726-7.

Apostol, I., Heinstein, P. F., & Low, P. S. (1989). Rapid stimulation of an oxidative burst during Elicitation of cultured plant cells. *Plant Physiology*, *90*(1), 109-116. https://doi.org/10.1104/pp.90.1.109.

AP News. (2025, February 14). Thorny problems with Valentine's roses: Invasive bugs and toxic pesticides hurt Kenya's exports. https://thehill.com/homenews/ap/ap-business/ap-thorny- problems-with-valentines-roses-invasive-bugs-and-toxic-pesticides-hurt-kenyas-exports/

Arim, K. (2011). The impact of *Agrobacterium tumefasciens* and other soil-borne disease-causing agents of economic importance in the production of roses. In *Proceedings of the Video Conference on Global Competitiveness of the Flower Industry in the East African Region*. Kenya Development Learning Centre.

Artur, A., Piotr, S., & Stanisław, B. (2012). Efficacy of fungicides and essential oils against bacterial diseases of fruit trees. *Journal of Plant Protection Research*, *52*(4), 467-471. https://doi.org/10.2478/v10045-012-0075-7

Auh, C. K., & Murphy, T. M. (1995). Plasma Membrane Redox Enzyme Is Involved in the Synthesis of O2- and H2O2 by Phytophthora Elicitor-Stimulated Rose Cells. *Plant physiology*, *107*(4), 1241–1247. https://doi.org/10.1104/pp.107.4.1241

Azcón-Aguilar, C., Jaizme-Vega, M. C., & Calvet, C. (2002). The contribution of arbuscular mycorrhizal fungi to the control of soil-borne plant pathogens. *Mycorrhizal Technology in Agriculture*, 187-197. https://doi.org/10.1007/978-3-0348-8117-3_15

Backman, P. A., Wilson, M., & Murphy, J. F. (2018). Bacteria for biological control of plant diseases. *Environmentally Safe Approaches to Crop Disease Control*, 94-110. https://doi.org/10.1201/9781351071826-7

Bahadur, I., Meena, V. S., & Kumar, S. (2014). Importance and application of potassic biofertilizer in Indian agriculture. *International Resource Journal Biological Science, 3*(12),80-85. https://www.academia.edu/download/98864800/14.ISCA-IRJBS-2014-170.pdf.

Baker, C. J., & Orlandi, E. W. (1995). Active oxygen in plant pathogenesis. *Annual Review Of Phytopathology*, *33*(1), 299-321. https://www.annualreviews.org/content/journals/10.1146/annurev.py.33.090195.001503

Barasarathi, J., Perveen, K., Khan, F., Muthukumaran, M., Debnath, A., Behera, M., Pongen, M., Sayyed, R., & Mastinu, A., (2025). Targeting *Agrobacterium tumefasciens*: A Computational Study on Quorum Sensing Inhibition. *Journal of Basic Microbiology 65, (7):*70041.doi: 10.1002/jobm.70041. Epub 2025 Apr 22. PMID: 40264335; PMCID: PMC12216800.

Barton, I. S. (2019). *Genome Stability, Evolution, and Dynamics in the Cooperative Plant Pathogen, Agrobacterium tumefaciens* (Doctoral dissertation, Indiana University).

Bhattacharyya, P. N., & Jha, D. K. (2012). Plant growth-promoting rhizobacteria (PGPR): emergence in agriculture. *World Journal of microbiology and biotechnology*, *28*(4), 1327-1350.

Bayer Crop Science. (2019). *Sustainability Report*. Site maintenance https://www.bayer.com/sites/default/files/2024-03/bayer-sustainability- report-2023.pdf.

Ben. G. H., Bouri, M., Mougou H., A., Schuster, C., Leclerque, A., & Rhouma (2021) A. *Bacillus velezensis* strain MBY2, A potential agent for the management of crown gall disease. *Plant Disease 14*, 120 – 125.

Bendahmane, M., Dubois, A., Raymond, O., & Bris, M. L. (2013). Genetics and genomics of flower initiation and development in roses. *Journal of Experimental Botany*, 64(4), 847–857.

Berg, van den, M. (2013). Importing, productivity and SMEs: firm-level evidence from the Netherlands. *Discussion Paper Series, nr 13–07.* Tjalling C. Koopmans Research Institute

Bermudez, S., & Mumbi, J. (2024). The Kenyan flower subsector. *International Institute for Sustainable Development. https://www. iisd. org/publications/report/sustainability-standards-kenyan-flower-subsector*.

Bermudez, S., & Ngige, J. M. (2024). *The Kenyan flower subsector: A model of enhanced*

competitiveness through mandatory and voluntary sustainability standards (Case study). International Institute for Sustainable Development.

Bernhardt, P. (1999). *The rose's kiss: A natural history of flowers*. Island Press.

Bika, R., Baysal-Gurel, F., & Jennings, C. (2020). *Botrytis cinerea* management in ornamental production: A continuous battle. *Canadian Journal of Plant Pathology*, *43*(3), 345-365. https://doi.org/10.1080/07060661.2020.1807409

Biondi, E., Bini, F., Anaclerio, F., & Bazzi, C. (2009). Effect of bioagents and resistance inducers on grapevine crown gall. *Phytopathologia Mediterranea*, *48*(3), 379-384. https://www.jstor.org/stable/26463361

Bloom, T. J., & Tsujita, M. J. (2003). Cut rose product In: Av Roberts, Debener T., Gudin S., Editions. *Encyclopedia of Rose Science, 2*, 594-600. https://www.researchgate.net/publication/285816557_Cut_rose_production

Bosmans, L., Van Calenberge, B., Paeleman, A., Moerkens, R., Wittemans, L., Van Kerckhove, S., De Mot, R., Lievens, B. & Rediers, H. (2016). Efficacy of hydrogen peroxide treatment for control of hairy root disease caused by *Rhizogenic agrobacteria*. *Journal of applied microbiology*, *121*(2), 519–527. https://doi.org/10.1111/jam.13187

Brent, K. J., & Holloman, D. W. (1998). *Fungicide resistance: The assessment of risk* (Monograph No. 2, pp. 1–48). FRAC, Global Crop Protection Federation.4

Brown, P. J., Chang, J. H., & Fuqua, C. (2023). *Agrobacterium tumefasciens*: a transformative agent for fundamental insights into host-microbe interactions, genome biology, chemical signaling, and cell biology. *Journal of Bacteriology*, *205*(4), e00005-23.

Burr, T. J., Bazzi, C., Süle, S., & Otten, L. (1999). Crown gall of grape: Biology of *Agrobacterium vitis* and the development of disease control strategies. *Plant Disease*, *82* (12), 1288-1297. https://doi.org/10.1094/pdis.1998.82.12.1288

Calvo, P., Nelson, L, & Kloepper, J. W. (2014) Agricultural uses of plant biostimulants. *Plant soil* (283) 3 - 41https://link.springer.com/content/pdf/10.1007/s11104-014-2131-8.pdf

Central Bank of Kenya. (2021, January 2). *MPC Flower Farms Survey of January 2021*. CBK | Central Bank of Kenya. https://www.centralbank.go.ke/2021/02/02/mpc-flower-farms-survey-of-january-2021/

Chatterjee, R. (2025). *How Kenya is fuelling global romance with fresh flower exports*. Africa Aviation News, Supply Chain & Railway Updates | Logistics

Insights. https://www.logupdateafrica.com/trade/how-kenya-is-fuelling-global-romance-with-fresh-flower-exports-1354684

Chebet, K. E. (2021). The fragility of Kenya's cut flower sector and impacts as exposed by COVID-19. *Unpublished master's thesis, Ghent University*.

Chepoghisho, L. L. (2019). *Performance of The Kenyan Cut Flower Exports; An Empirical Investigation (1986-2018)* (Doctoral dissertation, University of Nairobi). https://erepository.uonbi.ac.ke/handle/11295/109380

Chen, F., Guo, Y. B., Wang, J. H., Li, J. Y., & Wang, H. M (2007). Biological control of grape crown gall by Rahnella aquatilis HX2. *Plant Disease. 91*, 957‑963

Chilton, M. D., Drummond, M. H., Merlo, D. J., Sciaky, D., Montoya, A. L., Gordon, M. P., & Nester, E. W. (1977). Stable incorporation of plasmid DNA into higher plant cells: the molecular basis of crown gall tumorigenesis. *Cell*, *11*(2), 263-271.

Christie, P. J., & Cascales, E. (2005). Structural and dynamic properties of bacterial type IV secretion systems. *Molecular membrane biology*, *22*(1-2), 51-61.

Çınar, İ., & Çolakoğlu, A. S. (2005). Potential health benefits of rose hip products. In *International Rose Hip Conference 690* (pp. 253-258) https://www.actahort.org/books/690/690_39.htm

Cochrane, S. A., Vederas, J. C. (2016). Lipopeptides from *Bacillus* and *Paenibacillus* spp. A Gold Mine of Antibiotic Candidates. *Medical. Resource.Revision*, 36, 4‑31.

Coetzee, J. H., & Hoffman, E.W. (2018). Social and environmental responsibility of the floriculture industry in South Africa. Acta Hortic. 1201, 479-484 DOI:10.17660/ActaHortic.2018.1201.64https://doi.org/10.17660/ActaHortic.2018.1201.64

Colla, G., Hoagland, L., Ruzzi, M., Cardarelli, M., Bonini, P., Canaguier, R., & Rouphael, Y. (2017). Biostimulant action of protein hydrolysates: Unraveling their effects on plant physiology and microbiome. *Frontiers in plant science, 8*, 2202.

Collins, A. (2001). *A class project for soil-borne plant pathogens* (PP 728). North Carolina State University, Department of Plant Pathology.

Compant, S., Samad, A., Faist, H., & Sessitsch, A. (2019). A review on the plant microbiome: ecology, functions, and emerging trends in microbial application. *Journal of advanced research*, *19*, 29-37.

COST Action 873 Training School. (2011). *Agrobacterium classical and molecular phytobacteriology* [Training school held 5–9 September 2011, University Lyon 1, Villeurbanne, France]. Yumpu. https://www.yumpu.com/en/document/view/35391163/cost-action-873-training-school-agrobacterium-cost-873

Costechareyre, D., Rhouma, A., Lavire, C., Portier, P., Chapulliot, D., Bertolla, F., Boubaker, A., Dessaux, Y., & Nesme, X. (2010). Rapid and efficient identification of *Agrobacterium* species by recA allele analysis. *Microbial Ecology*, *60*(4), 862-872. https://doi.org/10.1007/s00248-010-9685-7

Cutler, R. (2003). Secondary metabolites | Culinary uses and nutritional value. *Encyclopedia of Rose Science*, 707-716. https://doi.org/10.1016/b0-12-227620-5/00200-7

Daughtrey, M., & Buitenhuis, R. (2020). Integrated pest and disease management in greenhouse ornamentals. *Integrated Pest and Disease Management in Greenhouse Crops*, 625-679. https://doi.org/10.1007/978-3-030-22304-5_22

Debener, T., & Linde, M. (2009). Exploring complex ornamental genomes: the rose as a model plant. *Critical reviews in plant sciences*, *28*(4), 267-280.

De Cleene, M., & De Ley, J. (1976). The host range of crown gall. *The botanical review*, *42*(4), 389-466.

De Curtis, F., Lima, G., Vitullo, D., & De Cicco, V. (2010). Biocontrol of *Rhizoctonia solani* and *Sclerotium rolfsii* on tomato by delivering antagonistic bacteria through a drip irrigation system. *Crop Protection*, *29*(7), 663-670. https://www.sciencedirect.com/science/article/pii/S0261219410000244

Demoz, B. T., & Korsten, L. (2006). *Bacillus subtilis* attachment, colonization and survival on avocado flowers and its mode of action on stem-end rot pathogens. *Biological Control*, *37*(1), 68-74.

Dessaux, Y., & Faure, D. (2018). Quorum sensing and quorum quenching in *Agrobacterium*: a "go/no go system"? *Genes*, *9*(4), 210.

Dinham, B. (2003). Introduction to Part 1. *Silent Invaders: Pesticides, Livelihoods and Women's Health*.

Dolan, C., Opondo, M., & Sally Smith, S. (2012). Gender, rights & participation in the Kenya cut flower industry.

Dudutech (2022). Traps and Pheromoes https://www.dudutech.com/category/traps-and-pheromones/#:~:text=For%20example%2C%20Western%20flower%20thrips,What%20other%20benefits?

Du Jardin, P. (2012). The Science of Plant Biostimulants–A bibliographic analysis, Ad hoc study report. https://orbi.uliege.be/bitstream/2268/169257/1/Plant_Biostimulants_final_report_bio

Emil, R., Svetlana, M., Todorovic, M., & Ivan, P. (2007). Possibilities of biological and chemical control of verticillium wilt in pepper. *Phytoparasitica*, *35*(5), 436-441. https://doi.org/10.1007/bf03020601

Ertter, B. (2001). *Native California roses*. An on-line Monograph published through the University of California. https://ucjeps.berkeley.edu/ina/roses/roses.html

Escobar, M. A., & Dandekar, A. M. (2003). *Agrobacterium tumefasciens* as an agent of disease. *Trends in plant science*, *8*(8), 380-386. https://www.cell.com/trends/plant-science/abstract/S1360-1385 (03)00162-6

Eastwell, K. C., Sholberg, P. L., & Sayler, R. J. (2006) Characterizing potential bacterial biocontrol agents for suppression of *Rhizobium vitis* ,causal agent of crown gall disease in grapevines. *Crop Protection.* 25, 1191‑1200.

Etminani1, F., Harighi1, B., Bahramnejad, B., & Ali, A. M (2024). Antivirulence effects of cell-free culture supernatants of endophytic bacteria against grapevine crown gall agent *Agrobacterium tumefaciens* and induction of defense responses in plantlets. *BMC Plant Biology*. https://doi.org/10.1186/s12870-024-04779-1. BioMed Central+1

Everlon C. R., Luana, A.A., Carlos, H. B. S., Edvan T. F., Luziane, R. S., Lucas A. L. C., Daniel, G. P., Daniel, N., Olubukola, O. B. Maria, Q. V., Mateus, M & Nicolas, D. (2024). Effects of *Trichoderma harzianum* and *Bacillus subtilis* on the root and soil microbiomes of the soybean plant. Frontier Plant Science 15: 1 – 14.https://www.frontiersin.org/journals/plantscience/articles/10.3389/fpls.2024.1403160/full

Farag, M. A., Zhang, H., & Ryu, C. M. (2013). Dynamic chemical communication between plants and bacteria through airborne signals: Induced resistance by bacterial volatiles. J. Chem. Ecol. **2013**, 39, 1007‑1018.

Fira, D., Dimki´C, I., Beri´, T., Lozo, J., & Stankovi, S. (2018). Biological control of plant pathogens by *Bacillus* species. *Journal of Biotechnology*, *285*, 44‑55.

Ferrini, F., & Nicese, F. P. (2002). Response of English oak (Quercus robur L.) trees to biostimulants application in the urban environment. *Journal of Arboriculture*, *28*(2), 70- 75.

Fiasson, J., Raymond, O., Piola, F., Sanlaville-Boisson, C., & Jay, M. (2003). Classification | Chemotaxonomy and molecular taxonomy. *Encyclopedia of Rose Science*, 127-135. https://doi.org/10.1016/b0-12-227620-5/00018-5

Flower Hub (2023).The History of Kenya Flower Industry part 1 https://theflowerhub.net/the-history-of-the-kenyan-flower-industry-part-1/

The Flower Hub. (2025). *The History of the Kenyan Flower Industry – Part 1*. https://theflowerhub.net/the-history-of-the-kenyan-flower-industry-part-1/

FloralDaily (2023). *Kenya Flower Council presents annual report.* FloralDaily. https://www.floraldaily.com/article/9549495/kenya-flower-council-presents-annual-report/

Floriculture Kenya. (2025). *Kenya's flower exports reached $835 million in 2024*. https://www.capitalfm.co.ke/business/2025/02/kenyas-flower-exports-hit-835mn-in-2024

Food and Agricultural Organisation - FAO. (2012). Resistance Management Guidelines. https://www.google.com/search?q=resistance+management+guidelines+in+flower+farms&sca

Fougère-Danezan, M., Joly, S., Bruneau, A., Gao, X. F., & Zhang, L. B. (2015). Phylogeny and biogeography of wild roses with specific attention to polyploids. *Annals of Botany*, *115*(2), 275–291.

Furuya, N., Shimokusuzono, F., Nakamura, Y., Nishimura, K.,Takeshita, M., Matsuyama, N., Manabe, K., & Takanami, Y. (2004). Crown gall of tobacco caused by *Agrobacterium tumefasciens* biovar 1 in tobacco fields. *Journal of General Plant Pathology*, *70*(1), 39-44. https://doi.org/10.1007/s10327-003-0088-1

Gaffney, T., Friedrich, L., Vernooij, B., Negrotto, D., Nye, G., Uknes, S., Ward, E., Kessmann, H., & Ryals, J. (1993). Requirement of salicylic acid for the induction of systemic acquired resistance. *Science*, *261*(5122), 754-756) https://doi.org/10.1126/science.261.5122.754

Ge, J., Li, D., Ding, J., Xiao, X., & Liang, Y. (2023). Microbial coexistence in the rhizosphere and the promotion of plant stress resistance: a review. *Environmental Research*, *222*, 115298.

Gelvin, S. B. (2003). *Agrobacterium*-Mediated Plant Transformation: the Biology behind the "Gene-Jockeying" Tool. *Microbiology and Molecular Biology Reviews*, *67*(1), 16-37. https://doi.org/10.1128/mmbr.67.1.16-37.2003

Gelvin, S. B. (2017). Integration of Agrobacterium T-DNA into the plant genome. *Annual review of genetics*, *51*,195-217. https://doi.org/10.1146/annurev-genet-120215-035320

Getu, M. (2009). Ethiopian floriculture and its impact on the environment. *Mizan Law Review*, *3*(2). https://doi.org/10.4314/mlr.v3i2.54011

Goodner, B., Hinkle, G., Gattung, S., Miller, N., Blanchard, M., Qurollo, B., Goldman, B. S., Cao, Y., Askenazi, M., Halling, C., Mullin, L., Houmiel, K., Gordon, J., Vaudin, M., Iartchouk, O., Epp, A., Liu, F., Wollam, C., Allinger, M. & Slater, S. (2000). Genome sequence of the plant pathogen and biotechnology agent *Agrobacterium tumefasciens* C58. *Science*, *294*(5550):2328. https://doi.org/10.1126/science.1066803

Gheysen, G., Villarroel, R. & Van Montagu, M. (1991). Illegitimate recombination in plants: a model for T-DNA integration. *Genes & development*, *5*(2), 287-297. https://genesdev.cshlp.org/content/5/2/287.short

Glazebrook, J. (2001). Genes controlling expression of defence responses in arabidopsis — 2001status. *Current Opinion in Plant Biology*, *4*(4), 301-308. https://doi.org/10.1016/s13695266(00)00177-1

Gomez, K. A., & Gomez, A. A. (1984). Statistical procedures for agricultural research. John wiley & sons.

Goyal, R. K., & Habtewold, J. Z. (2023). Evaluation of legume–rhizobial symbiotic interactions beyond nitrogen fixation that help the host survival and diversification in hostile environments. *Microorganisms*, *11*(6), 1454.

Grady, E. N., MacDonald, J., Liu, L., Richman, A., & Yuan, Z. C. (2016). Current knowledge and perspectives of *Paenibacillus*: *A review of Microbiology*, *Cell Factories*, *15*, 203.

Craigie, J. S. (2011). Seaweed extract stimuli in plant science and agriculture. *Journal of applied phycology*, *23*(3), 371-393.

Grant, M., & Lamb, C. (2006). Systemic immunity. *Current Opinion in Plant Biology*, *9*(4), 414-420. https://doi.org/10.1016/j.pbi.2006.05.013

Grimm, R. (1987). Control of crown gall in Swiss apple nurseries. *EPPO Bulletin*, *17*, 269–272

Gudin, S. (2000). Rose: Genetics and breeding. *Plant Breeding Reviews*, 17, 159–189.

Gyul'akhmedov, A. N., & Gasanova, J. (1992). Effect of microelements on apple yield and quality. *Biologicheskie Nauki*, *3*, 3–17. https://www.cabidigitallibrary.org/doi/full/10.5555/19930324349

Harman, G. E., Howell, C. R., Viterbo, A., Chet, I., & Lorito, M. (2004). Trichoderma species — opportunistic, avirulent plant symbionts. *Nature Reviews Microbiology*, *2*(1), 43-56. https://doi.org/10.1038/nrmicro797

Hawkins, N. J., Bass, C., Dixon, A., & Neve, P. (2019). The evolutionary origins of pesticide resistance. *Biological Reviews*, *94*(1), 135-155.

Hermosa, R., Viterbo, A., Chet, I. & Monte, E. (2012). Plant-beneficial effects of *Trichoderma* and of its genes. *Microbiology*, *158*(1), 17-25. https://doi.org/10.1099/mic.0.052274-0.

Hoekema, A., Hirsch, P. R., Hooykaas, P. J., & Schilperoort, R. A. (1983). A binary plant vector strategy based on separation of vir-and T-region of the Agrobacterium tumefaciens Ti-plasmid. *Nature*, *303*(5913), 179-180.

Hoorn, C. (2024, March 26). *Certifications in floriculture and what they all mean*. Floristry Trade Club. https://www.floristrytradeclub.co.uk/post/certifications-in-floriculture-and-what-they-all-mean.

Hoorn, C. (2024). *Certifications in floriculture and what they all mean*. Floristry Trade Club. https://www.floristrytradeclub.co.uk/post/certifications-in-floriculture-and-what-they-all-mean.

Hortifresh Journal (2025). Kenya's flowers farms, goes solar in commitment to sustainability goal.https://hortfreshjournal.com/kenyas-flowers-farms-goes-solar-in-commitment-tosustainabilitygoal/#:~:text=To%20grow%20flowers%20sustainably%2C%20more,for%20exports%20in%20the%20country.

Hortfresh Journal. (2024, October 12). *Embracing global certification for market access*. https://hortfreshjournal.com/embracing-global-certification-for-market-access/

Howell, C. R. (2003). Mechanisms employed by *Trichoderma* species in the biological control of plant diseases: The history and evolution of current concepts. *Plant Disease*, *87*(1), 4-10. https://doi.org/10.1094/pdis.2003.87.1.4

Howell, C. R. (1995). Mechanisms in the Biocontrol of *Rhizoctonia solani*-Induced Cotton Seedling Disease by *Gliocladium virens*: Antibiosis. *Phytopathology*, *85*(4), 469. https://doi.org/10.1094/phyto-85-469.

Howell, C. R., & Stipanovic, R. D. (1995). Mechanisms in the biocontrol of *Rhizoctonia solani*-induced cotton seedling disease by *Gliocladium virens*: antibiosis.

Inomata, Y. S., Oikawa, S., Shinokawa, T., & Suzuki, E. (1992). The effect of gibberellins treatment on flowers of Japanese pears (*Pyrus pyrifolia Nakai*) after late frost damage. *Bulletin of the Fruit Tree Research Station, 23,* 123–126.

Jat, L. K., Singh, Y. V., Meena, S. K., Meena, S. K., Parihar, M., Jatav, H. S., & Meena, V. S. (2015). Does integrated nutrient management enhance agricultural productivity *Journal of Pure Applied Microbiology*, *9*(2), 1211-1221. https://www.academia.edu/download/38153510/JPAMVO9N2P1211-1221.pdf

Jennifer, O. (2017). *Diseases of roses*. OSU Extension - Oklahoma State University. https://extension.okstate.edu/fact-sheets/diseases-of-roses.html

Jin, S. G., Prusti, R. K., Roitsch, T., Ankenbauer, R. G., & Nester, E. W. (1990). Phosphorylation of the VirG protein of *Agrobacterium tumefasciens* by the autophosphorylated VirA protein: Essential role in biological activity of VirG. *Journal of Bacteriology*, *172*(9), 4945-4950. https://doi.org/10.1128/jb.172.9.4945-4950.1990

Juliette, P., Dominiek, V., Marcus, D., Rob, M., & Felix, L. W., (2020).Predators and Parasitoids-in-First: From Inundative Releases to Preventative Biological Control in Greenhouse Crops. Frontier In Sustainaibilty Food Sytems 4: 23-38 https://www.frontiersin.org/journals/sustainablefoodsystems/articles/10.3389/fsufs.2020.595630/full

Justine, C. Maxime, D., Thi, T. A. N., Martine, D., & Rusell, J. T. (2023). Effect of biostimulants on leafy vegetables (Baby leaf lettuce and Batavia lettuce) exposed to abiotic or biotic stress under two different growing systems. *Agronomy*, *13*(3), 879. https://doi.org/10.3390/agronomy13030879.

Kassa, A. T. (2024). Participatory Decision-Making in Resource Allocation in Areas Exposed to Floriculture Industry in Ethiopia: A Socio–Legal Analysis of the Case of Sululta Area in Oromia Regional State. *Haramaya Law Review*, *13*, 25-48

Kassa, A. T., Keno, G. I., Makahamadze, T., & Bekele, D. (2025). The socio-economic impacts of the floriculture industries on the smallholders in Ethiopia: The case of Sululta district in Oromia national regional state. *Frontiers in Horticulture*, *3*. https://doi.org/10.3389/fhort.2024.1504800

Kado, C. I. (1976). The tumor-inducing substance of *Agrobacterium tumefasciens*. *Annual Reviw of Phytopathology*, *14*(1), 265-308 https://doi.org/10.1146/annurev.py.14.090176.001405

Kado, C. I. (2002). Crown gall tumours. In S. Brenner & J. H. Miller (Eds.), *Encyclopedia of Genetics* (pp. 1–3). Academic Press.

Kado, C. I. (2014). Historical account on gaining insights on the mechanism of crown gall tumorigenesis induced by *Agrobacterium tumefasciens*. *Frontiers in microbiology*, *5*, 340.

Kairu, G. M., Nyangena, C. M., Javed, Z. U. & Crosse, J. E. (1984). Iatrogenic effects of captafol on bacterial blight of coffee. *Plant Pathology*, *33*(1), 131-132. https://doi.org/10.1111/j.1365-3059.1984.tb00596.x

Kariuki, G. M. (2015). Major diseases of roses in Kenya. *HortFresh Journal*, *9*(10), 39-42.

Kavutu, A. M., Mwangi, M., Kahuthia G. R., Wanjohi, W. (2008). Application of antagonistic bacteria in management of crown gall disease on roses in Kenya. In Proceedings of the 31st International Horticultural Congress (IHC)—Horticulture for aWorld inTransition/International Symposium on Sustainable Control of Pests and Diseases, Angers, France

Kawaguchi, T., Inoue, K., & Tanina, K., (2015). Evaluation of the nonpathogenic *Agrobacterium vitis* strain ARK-1 for biological control of grapevine crown gall. *Plant Disease.* (Earlier field trials and meta-analysis supporting ARK-1 use). APS Journals+1

Kawaguchi, A., & Inoue, K. (2012). New antagonistic strains of non-pathogenic *Agrobacterium vitis* to control grapevine crown gall. *Journal of Phytopathology 160,* 509‑518.

Khan, A. R., Mustafa, A., Hyder, S., Valipour, M., Rizvi, Z. F., Gondal, A. S., Yousuf, Z., Iqbal, R., & Daraz, U. (2022). *Bacillus* spp. as Bioagents: Uses and Application for Sustainable Agriculture. *Biology 11,* 1763.4 – 20.

Kim, W. I., Cho, W. K., Kim, S. N., Chu, H.;,Ryu, K.Y., Yun, J. C., & Park, C.S. (2011). Genetic diversity of cultivable plant growth-promotingrhizobacteria in *Korea Journal Microbiology Biotechnology*, *21,* 777–790.

Kenya Flower Council. (2019, July). *Floriculture export in Kenya* [Web exclusive story]. Farmbiz Africa. http://www.farmbizafrica.com

Kenya Flower Council. (2021).The three major challenges for Kenya's flower Industry in 2021. *FloralDaily: Global Flower News.* https://www.floraldaily.com/article/9327353/the-three-major-challenges-for-kenya-s-flower-industry-in-2021/

Kenya Flower Council (2021). *Flowers & Ornamentals Sustainability Standard (FOSS).* Kenya ‘Flower Council.

Kenya’s Flower Market Update. (2023). *Kenya’s flower market report*. Thursd. https://thursd.com/articles/kenya-flower-market-update

Kenya Flower Council (2023).Kenya Flower Council Certification Scheme Quality System Regulation. https://kenyaflowercouncil.org/images/Standards.pdf

Khan, M. S., Zaidi, A., Wani, P. A., & Oves, M. (2009). Role of plant growth promoting Rhizobacteria in the remediation of metal contaminated soils. *Environmental chemistry letters*, *7*(1), 1-19.

Knuth, M., Mohamed, M., & Solliday, A. (2025). *Evolving U.S. consumer trends in sustainable floriculture: A decade-by-decade analysis from the 1990s to the 2020s*. NC State Extension Publications. https://content.ces.ncsu.edu/evolving-us-consumer-trends-in-sustainable-floriculture

Kumar, A., Bahadur, I., Maurya, B. R, Raghuvanshi, R., Meena, V. S, Singh, D. K., & Dixit, J. (2015). Does a plant growth promoting rhizobacteria enhance agricultural sustainability? https://www.cabidigitallibrary.org/doi/full/10.5555/20153197157

Kumar, A., Patel, J. S., Bahadur, I., & Meena, V. S. (2016). The molecular mechanisms of KSMs for enhancement of crop production under organic farming. *Potassium Solubilizing Microorganisms for Sustainable Agriculture*, 61-75. https://doi.org/10.1007/978-81-322-2776-2_5

Kumar, K.V. K., Yellareddygari, S. K., Reddy, M. S., Kloepper, J. W., Lawrence, K. S., Zhou, X. G, Sudini, H., Groth, D. E., Raju, S. K., & Miller, M. E. (2012). Efficacy of *Bacillus subtilis* MBI 600 against sheath blight caused by Rhizoctonia solani and on growth and yield of rice. Rice Science, 19(1), 55-63.https://doi.org/10.1016/s1672-6308 (12) 60021-3.

Kuzmanovi, C., N., Puławska, J., Hao, L., & Burr, T. J.(2018). The Ecology of *Agrobacterium vitis* and Management of Crown Gall Disease in Vineyards. Current Topical. *Microbiology Immunology*. *2018*, 418, 15–53.

Kuźniak, E., & Urbanek, H. (2000). The involvement of hydrogen peroxide in plant responses to stresses. *Acta Physiologiae Plantarum*, *22*(2), 195-203. https://doi.org/10.1007/s11738-000-0076-4

Langan, M. (2011). Uganda's flower farms and private sector development. *Development and Change*, *42*(5), 1207-1240.

Larebeke, N. V., Engler, G., Holsters, M., den Elsacker, S. V., Zaenen, I., Schilperoort, R. A., & Schell, J. (1974). Large plasmid in *Agrobacterium tumefasciens* essential for crown gall-inducing ability. *Nature*, *252*(5479), 169-170.

Lassalle, F., Campillo, T., Vial, L., Baude, J., Costechareyre, D., Chapulliot, D., & Nesme, X. (2011). Genomic species are ecological species as revealed by comparative genomics in Agrobacterium tumefaciens. *Genome Biology and Evolution*, *3*, 762-781.https://academic.oup.com/gbe/articleabstract/doi/10.1093/gbe/evr070/589154

Lastochkina, O., Seifikalhor, M., Aliniaeifard, S., Baymiev, A., Pusenkova, L., Garipova, S., Kulabuhova, D., & Maksimov, I. (2019). *Bacillus* Spp.: Efficient biotic strategy to control Postharvest diseases of fruits and vegetables. *Plants*, *8*(4), 97. https://doi.org/10.3390/plants8040097

Lee, C., Efetova, M., Engelmann, J. C., Kramell, R., Wasternack, C., Ludwig-Müller, J., Hedrich, R., & Deeken, R. (2009). *Agrobacterium tumefasciens* promotes tumour induction by modulating pathogen defence in *Arabidopsis thaliana*. *The Plant Cell*, *21*(9), 2948-2962. https://doi.org/10.1105/tpc.108.064576

Leipold, B., & Morgante, F. (2013). The impact of the flower industry on Kenya's Sustainable development. International Public Policy Review, 7(2), 1-31.

Liu, Y., Han, S., Song, L., Li, L., Wang, H., Pan, M., & Tan, J. (2024). Screening of bacterial endophytes of larch against *Neofusicoccum laricinum* and validation of their safety. *Microbiolog Spectrum, 12,* 112-23.

Li, S., Zhang, N., Zhang, Z., Luo, J., Shen, B., Zhang, R., & Shen, Q. (2012). Antagonist *Bacillus subtilis* HJ5 controls verticillium wilt of cotton by root colonization and biofilm formation. *Biology and Fertility of Soils*, *49*(3), 295-303. https://doi.org/10.1007/s00374-012-0718-x

Li, Y., Gao., Z., Kong, W., Xiao, Y., Adjei, M.O., Fan, B. (2025). Bio control of Crown Gall Disease of Cherry Trees by *Bacillus velezensis*. *Plants, 14*,475. https://doi.org/10.3390/plants14030475

Lindström, K., & Young, J. P. (2010). International committee on systematics of prokaryotes subcommittee on the taxonomy of Agrobacterium and rhizobium. *International Journal of Systematic and Evolutionary Microbiology*, *61*(12), 3089-3093. https://doi.org/10.1099/ijs.0.036913-0.

Ljubojević, M., & Božanić Tanjga, B. (2025). Rose (Rosa× hybrida L.) Breeding. An Old Flower for a New Age. In *Breeding of Ornamental Crops: Annuals and Cut Flowers* (pp. 591-638). Cham: Springer Nature Switzerland.

Loise M., Paul N., Margaret, K. (2015). Assessment of Occupational Safety and Health Practices for Organophosphate and Carbamate Pesticides in Flower Farms in Naivasha, Nakuru County Kenya *International Journal of Science and Research*. 6:391.

.Maarten, H., Ryder, M., John, E. S., Anna, S., & Stephen, K. F. (1987). Genetic analysis of agrocin 84 production and immunity in *Agrobacterium spp. Journal of*

Bacteriology, *169*(9), 4184-4189. https://doi.org/10.1128/jb.169.9.4184-4189.1987

Maina, G., Mutitu, E. W., & Ngaruiya, P. N. (2013). The Impact of *Agrobacterium tumefasciens* and other soil borne diseases on productivity of roses in East African Region.https://erepository.uonbi.ac.ke/bitstream/handle/11295/34815/2.ESA_.D02_KFC_Semina r_Issue_-_Agrobacterium.pdf?sequence=1

Make it Kenya / KEPROBA. (2024). *Horticulture sector in Kenya — statistics and trends*. Make it Kenya / KEPROBA. https://makeitkenya.go.ke/about-keproba/news-room/latest-news/horticulture-sector-in-kenya

Mala, M., Prasun, K., Mukherjee, B. A., Horwitz, C. Z., Gabriele, B., & Susanne Z. C. (2012).*Trichoderma* plant Pathogen interactions: Advances in genetics of biological control. *Indian Journal of Microbiology*, *52*(4)522-529. https://doi.org/10.1007/s12088-012-0308-5.

Martínez-Abarca, F., Herrera-Cervera, J. A., Bueno, P., Sanjuan, J., Bisseling, T., & Olivares, J. (1998). Involvement of salicylic acid in the establishment of the *Rhizobium meliloti*-alfalfa symbiosis. *Molecular Plant-Microbe Interactions®*, *11*(2), 153-155. https://doi.org/10.1094/mpmi.1998.11.2.153.

Matthysse, A. G., Thomas, D. L., & White, A. R. (1995). Mechanism of cellulose synthesis in *Agrobacterium tumefasciens*. *Journal of bacteriology*, *177*(4), 1076-1081.

McManus, P. S., & Stockwell, V. O. (2001). Antibiotic use for plant disease management in the United States. *Plant Health Progress*, *2*(1). https://doi.org/10.1094/php-2001-0327-01-rv

Mebrat, S., Degwale, A., Mekonen, T., & Mebrat, A. (2022). Flower production prospects and sustainability challenges in Ethiopia: A systematic review. *Frontiers in Environmental Science*, *10*, 1026544.

Meena, R. K, Singh, R. K, Singh, N. P, Meena S. K., & Meena V. S (2016). Isolation of low Temperature surviving plant growth – promoting rhizobacteria (PGPR) from pea (*Pisum sativum L*.) and documentation of their plant growth promoting traits. *Biocatalysis and Agricultural Biotechnology*, 4(4), 806-811. https://doi.org/10.1016/j.bcab.2015.08.006

Meena, M., Swapnil, P., Divyanshu, K., Kumar, S., Harish. T. Y.N., Zehra, A., Marwal, A., & Upadhyay, R.S. (2020). PGPR-mediated induction of systemic resistance and physiochemical alterations in plants against the pathogens: *Current perspectives Journal. Basic Microbiol, 60,* 828–861.

Mekonnen, M. M., & Hoekstra, A. Y. (2010). *Mitigating the water footprint of export cut Flowers from the Lake Naivasha Basin, Kenya* (Report No. 45). Water Footprint Network.

Mercado-Flores, Y., Cardenas-Alvarez, I. O., Rojas-Olvera, A. V., Perez-Camarillo, J. P., Leyva-Mir, S. G., & Anducho-Reyes, M. A. (2014). Application of *Bacillus subtilis* in the biological control of the phytopathogenic fungus *Sporisorium reilianum. Biological Control*, 76, 36–40. https://www.sciencedirect.com/science/article/abs/pii/S1049964414000942

Miller, W. B. (2012). Current status of growth regulator usage in flower bulb forcing in North America. *Floriculture and Ornamental Plant Biotechnology*, *6*(1), 35-44.

Mitali, M. P., Manoranjan, K. & Sahu, R. K. (2012). Bioefficacy of some plant extracts on growth parameters and control of diseases in *Lycopersicum esculentum*. *Asian Journal of Plant Science and Research, 2*(2), 129-142. https://www.researchgate.net/publication/267959930_Bioefficacy_of_some_plant_extracts_on_growth_parameters_and_control_of_diseases_in_Lycopersicum_esculentum

Mnif, I. (2015). Potential of bacterial derived biopesticides in pest management. Crop Protection, 77, 52-64. https://doi.org/10.1016/j.cropro.2015.07.017

Mondal, M. F., Asaduzzaman, M., Tanka, H., & Asao, T. (2015). Effect of amino acids on the Growth and flowering of *Eustoma grandiflorum* under autotoxicity in closed hydroponic culture. *Scientia Horticulturae, 192,* 453–459. https://www.cabidigitallibrary.org/doi/full/10.5555/20153336567

Moore, L. W., Chilton, W. S., & Canfield, M. L. (1997). Diversity of opines and opine-Catabolizing bacteria isolated from naturally occurring crown gall tumours. *Applied and Environmental Microbiology*, *63*(1), 201-207. https://doi.org/10.1128/aem.63.1.201- 207.1997

Mordor Intelligence. (2024). Kenya floriculture market. *Mordor Intelligence.* https://www.mordorintelligence.com/industry-reports/kenya-floriculture-market

Murugi, G. J. (2015). *Management of crown gall disease of roses using Agrobacterium radiobacter, corn oil and copper hydroxide and oxychloride in Kenya* (Doctoral dissertation, Kenyatta University).

Musyoki, M. S., & Kihara, A. (2025). Cold chain infrastructure management strategies and performance of flower industry in Kenya. *Journal of Business and Strategic Management*, *10*(8), 1-19. https://doi.org/10.47941/jbsm.2784

Mwase, D. E. (2015). Performance of floriculture industry in East Africa: What lessons can Tanzania learn from Kenya? *Asian Business Review*, *5*(1), 20-27. https://doi.org/10.18034/abr.v5i1.48

Nation Media Group. (2025, March 10). Cut flower export cash down 32pc in 5 years on tough times. *Daily Nation*. https://nation.africa/kenya/business/cut-flower-export-cash-down-32pc-in-5-years-on-tough-times--5184454

Nagrale, D.T., Chaurasia, A., Kumar, S., Gawande, S. P., Hiremani, N. S., Shankar, R., Gokte-Narkhedkar, N., R., & Prasad, Y. G. (2023). PGPR: The treasure of multifarious beneficial microorganisms for nutrient mobilization, pest biocontrol and plant growth promotion in field crops. *World Journal of Microbiology Biotechnology*, 39 - 100

Nahed, G. A. A., Mona, H. M., & Azza, A. M. M. (2009). Physiological effect of phenylalanine and tryptophan on growth and chemical constituents of *Antirrhinum majus* plants. *Ozean Journal of Applied Sciences, 2*(4), 399–407.

Neha, D., Priyanka, K. & Chanchai, T. (2024). Global trends in floriculture. In *Floriculture and landscaping chronicles: A collaborative insights* (pp. 190–221). Stella International Publication.https://www.researchgate.net/publication/379037750_Global_Trends_in_Floriculture

Ngutu, M., Bukachi, S., Olungah, C. O., Kiteme, B., Kaeser, F., & Haller, T. (2018). The actors, rules and regulations linked to export horticulture production and access to land and water as common pool resources in Laikipia County, northwest Mount Kenya. *Land, 7*(3), 110. https://doi.org/10.3390/land7030110

Njogu, G. M. (2022). *Economic sustainability in the floriculture value chain in Kenya* (Doctoral dissertation, Strathmore University).

Niyokuri, A., Nyalala, S., & Mwangi, M. (2017). Effects of bioslurry and plant biostimulant Hicure® on yield, flower quality and vase life of carnation (*Dianthus caryophyllus L*), *Journal of Applied Horticulture*, *19*(01), 29-34. https://doi.org/10.37855/jah.2017.v19i01.05

Odongo, S. A. (2023). *Water-related Conflicts in the Malewa River Basin in Nakuru County, Kenya–1980-2012* (Doctoral dissertation, University of Nairobi).

Ophel, K., & Kerr, A. (1990). Agrobacterium vitis Sp. Nov. for strains of *Agrobacterium* biovar 3 from grapevines. *International Journal of Systematic Bacteriology*, *40*(3), 236-241. https://doi.org/10.1099/00207713-40-3-236.

Opondo. M. (2006). Emerging Corporate Social Responsibility in Kenya's Cut Flower Industry.https://www.researchgate.net/publication/265824927_Emerging_Corporate_Social_Responsibility_in_Kenya's_Cut_Flower_Industry

Oksel, C., Liyanapathiranage, P., Parajuli, M., Avin, F. A., Jennings, C., Simmons, T., & Baysal-Gurel, F. (2024). Evaluation of Chemical and Biological Products for Control of Crown Gall on *Rose Pathogens 13*, 708. https://doi.org/10.3390/pathogens1308070

Opisa, O. O. M., Achwanya, O. S., Otaye, D. O., & Muthamia, J. M. (2020). The efficacy of sterilizing agents, copper oxychloride, vegetable oil and agrowipe (botanic neem extract) against crown gall disease of roses in Kericho, Kenya. *African Journal of Biological Sciences, 2*(4), 115-121.

Oniang'o, M. O., Khare, K. B., Achwanya, O. S., & Otieno, W. (2005). Efficacy of synthetic and non-synthetic fungicides against stem canker disease of tea. *African Crop Science Journal*, *13*(3). https://www.ajol.info/index.php/acsj/article/view/46196/32598

Opisa, O. M., Achwanya, O. S., Otaye, D. O., & Muthamia, J. M. (2021). The Efficacy of Biostimulants in the Management of *Agrobacterium tumefasciens* the Cause of Crown Gall Disease of Roses in Kericho, Kenya. *African Journal of Biological Sciences, 3*(2), 9-15.

Opisa, O. M., Anchwanya, O. S., Otaye, D. O., & Muthamia, J. M. (2021). The Efficacy of Sterilization Agents, Copper Oxychloride, Vegatable

Oil and Agrowipe (Botanic Neem Exctract) against Crown Gall Disease of Roses in Kericho, *Kenya. African Journal of Biological Sciences, 2,* 115‑121.

Osho Chemicals. (2018). Enrich BM. *OshoChem.* https://www.oshochem.com

Orlale, O. (2020, February 2). *CSR Africa: A tool to improve the flower farms' social performance and rights of workers*. Hivos. https://hivos.org/csr-africa-a-tool-to-improve-the-flower-farms-social-performance-and-rights-of-workers/

Padmini, S. M., & Kodagoda, T. D. (2017). Present status and future scope of floriculture industry in Sri Lanka and its potential in women empowerment. *Sri Lanka Journal of Social Sciences, 40*(1), 31. https://doi.org/10.4038/sljss.v40i1.7499

Pan, M., Xu, J., Han, S., Sun, Y., & Tan, J. (2023). Effects of *Bacillus cereus* NJSZ-13 on Fatty Acid Metabolism of Bursaphelenchus xylophilus.Forests **2023**, 14, 2065.

Pandey, V., Ansari, M. W., Tula, S., Yadav, S., Sahoo, R. K., & Shukla, N. (2016). Dose-

dependent response of *Trichoderma harzianum* in improving drought tolerance in rice genotypes. *Planta, 243,* 1251–1264.

Parađiković, N., Vinković, T., Vinković Vrček, I., Žuntar, I., Bojić, M. & Medić-Šarić, M. (2011). Effect of natural biostimulants on yield and nutritional quality: An example of sweet yellow pepper (*Capsicum annuumL.*) plants. *Journal of the Science of Food and Agriculture*, 91(12), 2146-2152. https://doi.org/10.1002/jsfa.4431

Parewa, H. P., Yadav, J., Rakshit, A., Meena, V. S., & Karthikeyan, N. (2014). Plant growth promoting rhizobacteria enhance growth and nutrient uptake of crops. *Agric Sustain*

Paterson, J., Jahanshah, G., Li, Y., Wang, Q., Mehnaz, S., Gross, H. (2017). The contribution of genome mining strategies to the understanding of active principles of PGPR strains. FEMS *Microbiology Ecology,* 93,-249

Percival, G. C. (2010). Effect of systemic inducing resistance and biostimulant materials on apple scab using a detached leaf bioassay. *Arboriculture & Urban Forestry*, *36*(1), 41-46. *Dev*, *2*(2), 101-116.

Patentlens.net. (2014). *Binary vectors. Available at*: http: //www. patentlens.net/daisy/Ag roTran/g1/848.html.

Petre, R., Labourdette, G., Braun, C. A., Meredith, R., Hauke, K., Van Hemelrijck, W., Schoofs, H., Deckers, T., Keulemans, W., Schoevaerts, C., Becker, R. C., & De Maeyer, L. (2015). Fosetyl-Al (Aliette), a plant defence enhancer with good effect on bacteria and on ascomycetes in apples and pears. *Acta Horticulturae, 1094,* 431–438.

Pieterse, C. M., Zamioudis, C., Berendsen, R. L., Weller, D. M., Van Wees, S. C., & Bakker, P. A. (2014). Induced systemic resistance by beneficial microbes. *Annual Review of Phytopathology, 52(*1), 347-375. https://doi.org/10.1146/annurev-phyto-082712-102340

Pieterse, C. M., & Van Loon, L. C. (1999). Salicylic acid-independent plant defence pathways. Trends in Plant Science, 4(2), 52-58. https://doi.org/10.1016/s1360-1385 (98)01364-8

Prithiviraj, B., Bais, H. P., Jha, A. K., & Vivanco, J. M. (2005). *Staphylococcus aureus* pathogenicity on Arabidopsis thaliana is mediated either by a direct effect of salicylic acid on the pathogen or by SA-dependent, NPR1-independent host responses. *The Plant Journal*, *42*(3), 417-432. https://doi.org/10.1111/j.1365-313x.2005.02385.x

Ploeg, R. (2020, March 16). *Royal FloraHolland faces the biggest crisis in its history* • AIPH. AIPH. https://aiph.org/floraculture/news/royal-floraholland-faces-the-biggest-crisis-in-its-history/

Pulawska, J. (2010). Crown gall of stone fruits and nuts, economic significance and diversity of its causal agents: tumorigenic *Agrobacterium* spp. *Journal of Plant Pathology*, S87-S98.

Reimers, F., Smolka, S. E., Werres, S., Schumacher, K. P., & Wagner, K. G. (1993). Effect of ajoene, a compound derived from *Allium sativum*, on phytopathogenic and epiphytic microorganisms. *Zeitschrift für Pflanzenkrankheiten und Pflanzenschutz*, 100, 622–633.

Resource Trade Earth. (2023). *Total Kenyan exports of cut flowers & flower buds for bouquets, etc., fresh (data from 2022).* Chatham House. https://resourcetrade.earth/?year=2022&exporter=404&category=429&units=weight&aut ozoo m=1

Rikken, M. (2012). Kenya Flower Industry Global Competitiveness Report. *Nairobi: Kenya Flower Council*.

Ritz, C. M., Schmuths, H., & Wissemann, V. (2004). Evolution by reticulation: European Dogroses originated by multiple hybridization across the genus Rosa. *Journal of Heredity*, *96*(1), 4-14. https://doi.org/10.1093/jhered/esi011

Rhouma, A., Boubaker, A. A., Nesme, X., & Dessaux, Y. (2006). Plasmid and chromosomal Diversity of a Tunisian collection of *Agrobacterium tumefasciens* strains. *Tunisian journal of plant protection*, *1*(2), 73-84.

Rhouma, A., Bouri, M., Boubaker, A., Nesme, X. (2008). Potential effect of rhizobacteria in the management of crown gall disease caused by *Agrobacterium tumefaciens* biovar 1.*Jouranal of Plant Pathology, 90*, 517–526.

Rouphael, Y. and Colla G. (2015). Biostimulants in horticulture. https://www.researchgate.net/publication/284105098_Biostimulants_in_horticulture

Rouphael, Y. & Colla, G. (2020). *Biostimulants in Agriculture*

Rosenberg, E., & Zilber-Rosenberg, I. (2016). Microbes drive evolution of animals and plants: The hologenome concept. *Biology, 7,* 01395.

Ruparao, G. (2011). Use of neem and plant-based biopesticides in floriculture: Current challenges and perspectives - A review The Journal of Horticultural Science and Biotechnology. 86 (3):203 – 209 https://www.researchgate.net/publication/286603322_Use_of_neem_and_plantbased_biopesticides_in_floriculture_Current_challenges_and_perspectives_-_A_review

Ryals, J. A., Neuenschwander, U. H., Willits, M. G., Molina, A., Steiner, H. Y., & Hunt, M. D. (1996). Systemic acquired resistance. *The plant cell*, *8*(10), 1809–1819.

Ryder, M. H., & Jones, D. A. (1990). Biological control of crown gall. In D. Hornby (Ed.), *Biological control of soil-borne plant pathogens* (pp. 45–63). CAB International.

Salgado-Salazar, C., Shiskoff, N., Daughtrey, M., Palmer, C. L., & Crouch, J. A. (2018). Downy mildew: A serious disease threat to rose health worldwide. *Plant Disease, 102*(10), 1873-1882. https://doi.org/10.1094/pdis-12-17-1968-fe

Santoyo, G., Urtis-Flores, C. A., Loeza-Lara, P. D., Orozco-Mosqueda, M. D. C., & Glick, B.R. (2021). Rhizosphere Colonization Determinantsby Plant Growth-Promoting Rhizobacteria (PGPR). *Biology, 10,* 475.

Sawada, H., Ieki, H., & Matsuda, I. (1995). PCR detection of Ti and Ri plasmids from phytopathogenic Agrobacterium strains. *Applied and environmental microbiology, 61*(2), 828-831.

Sayan, S., Bikash, G., Sukamal, S. & Prahlad, D. (2015). The effect of foliar application of biozyme on yield and physico-chemical properties of rainy season crop of guava (*Psidium guajava* L.) cv. Allahabad Safed in alluvial soil of West Bengal. *International Journal of Bio-resource, Environment, and Agricultural Sciences*, 1(4), 176–185.

Scalliet, G., Piola, F., Douady, C. J., Réty, S., Raymond, O., Baudino, S., & Hugueney, P. (2008). Scent evolution in Chinese roses. *Proceedings of the National Academy of Sciences, 105*(15), 5927-5932.

Schell, J., & van Montagu, M. (2009). The Ti-plasmid of *Agrobacterium tumefasciens*, a natural vector for the introduction of nif genes in plants. *Basic Life Sciences, 9*, 159–179. https://doi.org/10.1007/978-1-4684-0880-5_12

Siddiqua, A. (2024). Rose Petals and Plates: Exploring the Culinary and Health Wonders of Roses. https://www.researchgate.net/publication/382306217_Rose_Petals_and_Plates-Exploring_the_Culinary_and_Health_Wonders_of_Roses

Sigee, D. C. (1993). *Bacterial plant pathology: Cell and molecular aspects* (p. 102). Cambridge University Press.

Siginon, B. J., Bartirum, W. J., Samson, W. W. & Joseph M. K. (2024). The Impact of COVID-19 Pandemic on the Performance of the Floriculture Sector in Kenya: A Case Study of Karen Roses, Ravine Branch, Baringo County (2016-2021)". Asian Journal of Advanced Research and Reports 18 (2):112–132. https://doi.org/10.9734/ajarr/2024/v18i2609.

Singh, U. P., Prithiviraj, B., Wagner, K. B., & Plank-Schumacher, K. G. (1995). Effect of ajoene, a constituent of garlic (*Allium sativum*), on powdery mildew (*Erysiphe pisi*) of pea (*Pisum sativum*). *Journal of Plant Diseases and Protection, 102*(2), 399–406.

Singh, S. K., Sarma, B. K., Srivastava, J. S., Singh, U. P., & Ray, A. B. (1999). Antifungal activity of Δ3-alstovenine, a plant alkaloid isolated from *Alstonia venenata*. *Folia microbiologica*, *44*(5), 510-512. https://link.springer.com/article/10.1007/BF02816251

Sharma, N. (2024). Sustainable floriculture practices and Trendshttps://doi.org/10.59317/9788197781506

Shams, M., Campillo, T., Lavire, C., Muller, D., Nesme, X., & Vial, L. (2012). Rapid and efficient methods to isolate, type strains and determine species of Agrobacterium spp. in pure culture and complex environments. *Biochemical Testing*, *4*, 3-20.

Schiff, P. L. (2002). Opium and its alkaloids. *American Journal of Pharmaceutical Education*, *66*(2), 188-196.

Sharon, M. D. (2023). *Common diseases of roses*. The Connecticut Agricultural Experiment Station. https://portal.ct.gov/

Shah, J. (2003). The salicylic acid loop in plant defence. *Current Opinion in Plant Biology*, *6*(4), 365-371. https://doi.org/10.1016/s1369-5266 (03)00058-x

Spoel, S. H., Koornneef, A., Claessens, S. M. C., Korzelius, J. P, Van Pelt, J. A, Mueller, M. J., Buchala, A. J., Metraux, J. P., Brown, R., & Kazan, K. (2003). NPR1 modulates cross-talk between salicylate-and jasmonate-dependent defence pathways through a novel function in the cytosol. *The Plant Cell*, *15*(3), 760-770.

Stacey, G., McAlvin, C. B., Kim, S. Y., Olivares, J., & Soto, M. J. (2006). Effects of endogenous salicylic acid on nodulation in the model legumes *Lotus japonicus* and *Medicago truncatula*. *Plant Physiology*, *141*(4), 1473-1481. https://academic.oup.com/plphys/article-abstract/141/4/1473/6103521

Stachel, S. E., Messens, E., Van Montagu, M., & Zambryski, P. (1985). Identification of the signal molecules produced by wounded plant cells that activate T-DNA transfer in *Agrobacterium tumefasciens. Nature*, *318* (6047), 624-629.

Stanton, G. (2021). Plant DNA repair and *Agrobacterium* T−DNA integration. *International Journal of Molecular Sciences*, *22*(16), 8458. https://doi.org/10.3390/ijms22168458

Sunita, R., Saleena, L. M., Vasudevan, P., & Nair, S. (2003). Biological suppression of rice diseases by *Pseudomonas* spp. under saline soil conditions. *Plant Soil***,** 251, 73‑82.

Szegedi, E., Otten, L., & Czakó, M. (1992). Diverse Types of Tartrate Plasmids in Agrobacterium tumefaciens Biotype Ill Strains. *Molecular plant-microbe interactions*, *5*(5), 435-438.

Tan, S., Jiang, Y., Song, S., Huang, J., Ling, N., Xu, Y., & Shen, Q. (2013). Two *Bacillus amyloliquefaciens* strains isolated using the competitive tomato root enrichment method and their effects on suppressing *Ralstonia solanacearum* and promoting tomato plant growth. *Crop Protection*, *43*, 134-140. https://doi.org/10.1016/j.cropro.2012.08.003

Thaler, J. S., Owen, B., & Higgins, V. J. (2004). The role of the Jasmonate response in plant susceptibility to diverse pathogens with a range of lifestyles. *Plant Physiology*, *135*(1), 530-538. https://doi.org/10.1104/pp.104.041566

Thursd (2023). Key competitors and challenges in the Kenya Flower Industry. https://thursd.com/articles/bold-and-beautiful-kenyan-flowers.

Thomas, D. & David, H. B. (2014) Disease resistance breeding in rose: current status and potential of biotechnological tools. Plant Science (228):107-117. https://pubmed.ncbi.nlm.nih.gov/25438791/

Thompson, B. E. (2004). *Five years of Irish Trials on Biostimulants–The Conversion of a Skeptic. USDA Forest Service Proceedings*. RMRS-P-33.

Trend Economy. (2022). Cut flowers, flower buds imports and exports. https://trendeconomy.com/data/commodity_h2/0603

Tronsmo, A. (1991). Biological and integrated controls of *Botrytis cinerea* on Apple with *Trichoderma harzianum*. *Biological Control*, *1*(1), 59-62. https://doi.org/10.1016/1049-9644 (91)90102-6

Tyce, M. (2020). A 'private-sector success story'? Uncovering the role of politics and the state in Kenya's horticultural export sector. *The Journal of Development Studies*, *56*(10), 1877-1893. https://doi.org/10.1080/00220388.2020.1715944

Tzfira, T., Li, J., Lacroix, B., & Citovsky, V. (2004). Agrobacterium T-DNA integration: molecules and models. *TRENDS in Genetics*, *20*(8), 375-383.

Ullah, I., Yuan, W., Khalil, H. B., Khan, M. R., Lak, F., Uzair, M., Abbas, A., Mirzadi Gohari, A., & Wu, H. (2024). Understanding botrytis cinerea infection and gray mold management: A review paper on deciphering the rose's thorn. *Phytopathology Research*, *6*(1). https://doi.org/10.1186/s42483-024-00262-9

University of Adelaide Newsroom. (2024). Work on controlling crown gall disease bears fruit (report on GTI-5813 trials). University of Adelaide News. University of Adelaide+1

Utkhede, R. S., & Smith, E. M. (1993) Evaluation of Biological and Chemical Treatments for Control of Crown Gall on Young Apple Trees in the Kootenay Valley of British Columbia. *Journal of Phytopathology, 137*, 265–271.

Valerie, V., Annik, P., Scott, C., & Yves, D. (1998). The cryptic plasmid of *Agrobacterium tumefasciens* cointegrates with the Ti plasmid and cooperates for opine degradation. *Molecular Plant-Microbe Interactions, 11*(7), 583–591. https://doi.org/10.1094/MPMI.1998.11.7.583

Van der Meer, E., Hoogerwerf, F., Van Meulen, J., Kerklaan, E., Posthumus, J., Van Beek, A., Elings, A., Garcia, V. N., Rikken, M., & Humphries, G. (2012). *Handbook for greenhouse rose production Ethiopia*. DLV Plant. https://edepot.wur.nl/212019

Van der Velden, N. J. A., & Smit, P. X. (2019). *CO2-behoefte glastuinbouw 2030*. Wageningen Economic Research. research.wur.nl

Van Loon, L. C., Bakker, P. A. H. M., & Pieterse, C. M. J. (1998). Systemic resistance induced by rhizosphere bacteria. *Annual review of phytopathology*, *36*(1), 453-483.

Van Wees, S. C., De Swart, E. A., Van Pelt, J. A., Van Loon, L. C., & Pieterse, C. M. (2000). Enhancement of induced disease resistance by simultaneous activation of salicylate- and jasmonate-dependent defence pathways in Arabidopsis thaliana. *Proceedings of the National Academy of Sciences, 97*(15), 8711-8716. https://www.pnas.org/doi/abs/10.1073/pnas.130425197

Varshney, V. (2001). Effect of plant extracts on *Dreschslera graminae*, the causal agent of stripe disease of barley. *Indian Phytopathology, 54*(1), 88–90. https://epubs.icar.org.in/index.php/IPPJ/article/view/18848

Vazquez-Iglesias, I., Ochoa-Corona, F. M., Tang, J., Robinson, R., Clover, G. R., Fox, A., & Boonham, N. (2020). Facing Rose rosette virus: A risk to European rose cultivation. *Plant Pathology*, *69*(9), 1603-1617. https://doi.org/10.1111/ppa.13255

Verma, M., Brar, S. K., Tyagi, R., Surampalli, R., & Valéro, J. (2007). Antagonistic fungi, *Trichoderma* spp.: Panoply of biological control. *Biochemical Engineering Journal*, *37*(1), 1-20. https://doi.org/10.1016/j.bej.2007.05.012

Veluthambi, K., Gupta, A. K., & Sharma, A. (2003). The current status of plant transformation technologies. *Current Science, 84*(3), 368-380.

Walters, D., & Heil, M. (2007). Costs and trade-offs associated with induced resistance. *Physiological and Molecular Plant Pathology, 71*(1-3), 3-17.

Waitathu, N. (2025, May 16). *More investors enter flower industry despite global economic challenges*. People Daily - Kenya latest news, business and politics. https://peopledaily.digital/business/more-investors-enter-flower-industry-despite-global-economic-challenges

Wang, D., Weaver, N. D., Kesarwani, M., & Dong, X. (2005). Induction of protein secretory pathway is required for systemic acquired resistance. *Science, 308*(5724), 1036-1040.

Wang, T., Liang, Y., Wu, M., Chen, Z., Lin, J., & Yang, L. (2018). Natural products from *Bacillus subtilis* with antimicrobial properties. *Chinese Journal of Chemical Engineering, 23*(4), 744-754. https://www.sciencedirect.com/science/article/pii/S1004954115000051

Wani, M. A., Nazki, I. T., Din, A., Iqbal, S., Wani, S. A., Khan, F. U., & Neelofar. (2018). Floriculture sustainability initiative: The dawn of new era. In *Sustainable agriculture reviews 27* (pp. 91-127). Cham: Springer International Publishing.

Weindling, R. (1934). Studies on a lethal principle effective in the parasitic action of *Trichoderma lignorum* on *Rhizoctonia solani* and other soil fungi.

Weindling, R. (1934). Studies on lethal principle effective in the parasitization on SATSA Mukhepatra: Annual technical issue 21:2017218 *Rhizoctonia solani* and other soil fungi. *Phytopathology, 24,* 1153–1179.

White, C. E., & Winans, S. C. (2007). Cell–cell communication in the plant pathogen Agrobacterium tumefaciens. *Philosophical Transactions of the Royal Society B: Biological Sciences, 362*(1483), 1135-1148.

Wildermuth, M. C., Dewdney, J., Wu, G., & Ausubel, F. M. (2001). Isochorismate synthase is required to synthesize salicylic acid for plant defence. *Nature, 414*(6863), 562-565. https://doi.org/10.1038/35107108

Williams, E. B., & Kuc, J. (1969). Resistance in Malus to Venturia Inaequalis. *Annual Review of Phytopathology, 7*(1), 265-308. https://doi.org/10.1146/annurev.py.07.090169.0012

Wissemann, V., & Ritz, C. M. (2005). The genus Rosa (Rosoideae, Rosaceae) revisited: molecular analysis of nrITS-1 and atp B-rbc L intergenic spacer (IGS) versus conventional taxonomy. *Botanical journal of the Linnean Society, 147*(3), 275-290.

Wood, D. W. (2001). The genome of the natural genetic engineer *Agrobacterium tumefasciens* C58. *Science, 294*, 2317–2323. https://doi.org/10.1126/science.1066804

Yan, P., Zhang, X. T., Hu, L. J., Wang, Y. H., Zhu, M. L., Wu, X., Q., & Chen, F. (2020). Two novel strains, Bacillus albus JK-XZ3 and *B. velezensis* JK-XZ8, with activity against Cerasus crown gall disease in Xuzhou, China. *Australas Plant Pathology. 49*, 127‑136.

Yan, H. F., Acquash, S. J., Zhang, J. Y., Wang, G. Q, Zhang, C., & Darko, R. O.(2021). Overview of modelling techniques for green house techniques and evatranspiration. International journal of Agriculture and Biological engeneering 14 (6) 1-8.

Yakhin, O. I., Lubyanov, A. A., Yakhin, I. A., & Brown, P. H. (2017). Biostimulants in plant science: A global perspective. *Frontiers in Plant Science, 7,* 2049. https://doi.org/10.3389/fpls.2016.02049

Yánez-Mendizabal, V., Zeriouh, H., Viñas, I., Torres, R., Usall, J., Vicente, A. D., Pérez-García, A., & Teixidó, N. (2012). Biological control of peach brown rot (*Monilinia* spp.) by *Bacillus subtilis* CPA-8 is based on production of fengycin-like lipopeptides. *European Journal of Plant Pathology, 132,* 609–619. https://doi.org/10.1007/s10658-011-9874-0

Yeshiwas, T., & Workie, A. (2018). Social, economic and environmental issues of floriculture sector development in Ethiopia. *Review of Plant Studies, 5*(1), 1–10.

Young, J. M., Kuykendall, L. D., Martinez-Romero, E., Kerr, A., & Sawada, H. (2003). Classification and nomenclature of Agrobacterium and Rhizobium–a reply to Farrand. *International journal of systematic and evolutionary microbiology*, *53*(5), 1689-1695.

Yuan, Z. C., Edlind, M. P., Liu, P., Saenkham, P., Banta, L. M., Wise, A. A., Ronzone, E., Binns, A. N., Kerr, K., & Nester, E. W. (2007). The plant signal salicylic acid shuts down expression of the *vir* regulon and activates quorum-quenching genes in *Agrobacterium*. *Proceedings of the National Academy of Sciences of the USA, 104,* 11790–11795. https://doi.org/10.1073/pnas.0705376104

Zarif, H., Fan, C., Yuan, G., Zhou, R., Chang, Y., Sun, J., & Wang, C. (2025). Drought Stress in Roses: A Comprehensive Review of Morphophysiological, Biochemical, and Molecular Responses. *International Journal of Molecular Sciences*, *26*(9), 4272.

Zlesak, D. C. (2006). Rose *Rosa* × *hybrida*. *Flower Breeding and Genetics: Issues, Challenges and Opportunities for the 21st Century, 26,* 695–738. https://doi.org/10.17660/ActaHortic.2006.726.88

Zhongging, R., Qin, L., Xeni, K., Ian, S. B., Eli, G. S., Adrian, M. S., Clay, F., & Xidan, W. (2022). Conformation and dynamic interactions of multipartite genome in *Agrobacterium tumefasciens*. *Proceedings of the National Academy of Sciences USA.* https://www.ncbi.nlm.nih.gov/pmc/journals/2/

Wood D. W. (2001). The genome of the natural genetic engineer *Agrobacterium tumefaciens* C58. *Science (New York, N.Y.), 294*(5550), 2317–2323.

Yan, H. F., Acquash, S. J., Zhang, J. Y., Wang, G. Q, Zhang, C., & Darko, R. O. (2021). Overview of modelling techniques for green house techniques and evatranspiration. International journal of Agriculture and Biological engeneering 14 (6) 1-8.

Xia, Y., Deng, X., Zhou, P., Shima, K., & Teixeira da Silva, J. A. (2006). The world floriculture industry: Dynamics of production and markets. *Floriculture, ornamental and plant biotechnology*, *4*, 336-347.

Yahyaoui, H., Allaoui, N.E., Batbat, A. Aziz, A., Aoujil, F., Hafidi, M., & Khaoula Habbadi (2025).Biocontrol Potential of *Bacillus* Strains from Grapevine Rhizosphere Against *Allorhizobium vitis,* Causal Agent of Crown Gall Disease in Moroccan Vineyards. *International Journal of Plant Biolology*.16, 27. https://doi.org/10.3390/ijpb16010027

Yakhin, O. I., Lubyanov, A. A., Yakhin, I. A. & Brown, P. H. (2017). Biostimulants in Plant Science: A Global Perspective. *Frontier Plant Science* https://doi.org/10.1126/science.1066804

Yánez-Mendizábal, V., Zeriouh, H., Viñas, I., Torres, R., Usall, J., De Vicente, A., Pérez-García, A., & Teixidó, N. (2012). Biological control of peach brown rot (Monilinia spp.) by *Bacillus subtilis* CPA-8 is based on production of fengycin-like lipopeptides. *European Journal of Plant Pathology*, *132* (4), 609-619. https://doi.org/10.1007/s10658-011-9905-0

Tizazu, T. Y., & Workie, M. A. (2018). Social, economic and environmental issues of floriculture sector development in Ethiopia. Review of Plant Studies, 5(1), 1-10. https://doi.org/10.18488/journal.69.2018.51.1.10

Yao, B., Zhang, J. H., He, Y., L., Liang, X. Q., Cui, X. G., Rong, L. F., Zhou, X.. H. Wei, F. J.; He, Z. L. & Ji, G. H (2022). Study on biocontrol mechanism of *Bacillus amyloliquefaciens* JK10 to control blueberry crown gall disease. *Chinese Journal of. Biology Control, 38,* 613‑625.

Yuan, Z., Edlind, M. P., Liu, P., Saenkham, P., Banta, L. M., Wise, A. A., Ronzone, E., Binns, A. N., Kerr, K., & Nester, E. W. (2007). The plant signal salicylic acid shuts down expression of the vir regulon and activates quormone-quenching genes in *Agrobacterium*. *Natural Academic Science USA. Proceedings of the National Academy of Sciences, 104*(28), 11790-11795. https://doi.org/10.1073/pnas.0704866104

Zlesak, D. C. (2006). Rose *Rosa* × *hybrida*. *Flower Breeding and Genetics: Issues, Challenges and Opportunities for the 21st Century, 26,* 695–738. https://doi.org/10.17660/ActaHortic.2006.726.88

Zhongging, R., Qin, L., Xeni, K., Ian, S. B., Eli, G. S., Adrian, M. S., Clay, F., & Xidan, W. (2022). Conformation and dynamic interactions of multipartite genome in *Agrobacterium tumefasciens*. *Proceedings of the National Academy of Sciences USA.* https://www.ncbi.nlm.nih.gov/pmc/journals/2/

Zhu, L., Huang, J., Lu, X., & Zhou, C. (2022). Development of plant systemic resistance by beneficial rhizobacteria: Recognition, initiation, elicitation and regulation. *Frontier Plant Science 13,* 952397.

APPENDICES

Appendix 1: Raw data for yield of roses on plots treated with various biostimulants

		Time in months											
Treatments	**Replicates**	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**
1	1	10	72	37	37	53	35	60	52	46	59	62	37
1	2	9	77	30	0	54	27	59	61	28	37	72	61
1	3	10	64	12	0	56	31	51	49	17	25	71	31
1	4	6	69	22	0	56	35	53	47	28	31	50	51
2	1	5	80	24	0	40	27	62	68	48	41	63	51
2	2	20	82	18	0	39	37	57	82	24	42	78	44
2	3	14	81	29	0	66	41	59	63	28	40	96	62
2	4	8	75	14	0	40	42	58	61	22	37	54	26
3	1	18	90	24	0	51	33	72	54	52	55	64	40
3	2	5	63	19	0	49	40	63	75	40	37	54	39
3	3	5	80	10	0	54	43	68	73	45	53	96	57
3	4	11	67	20	0	46	31	39	51	21	36	57	37
4	1	15	95	49	0	60	31	62	84	51	57	79	53
4	2	5	63	29	0	58	42	62	67	28	31	102	45
4	3	8	79	23	0	68	50	71	83	46	45	86	43
4	4	18	86	17	0	64	35	45	59	24	55	72	48
5	1	15	80	19	0	54	37	57	74	48	46	77	59
5	2	11	80	25	0	58	35	55	80	48	24	75	45
5	3	14	85	21	0	45	35	58	57	34	33	49	41
5	4	10	83	17	0	43	30	50	60	20	30	49	37
6	1	11	67	15	0	43	28	55	67	34	64	82	51
6	2	13	100	34	0	78	45	63	55	32	47	82	57
6	3	17	79	12	0	71	54	73	46	48	30	79	36
6	4	14	64	14	0	38	25	43	52	24	38	73	56
7	1	15	94	29	0	57	29	54	74	38	57	63	56
7	2	14	80	23	0	70	47	64	86	40	45	62	35
7	3	5	72	13	0	46	41	55	64	24	33	55	45

7	4	6	59	18	0	47	23	44	63	41	32	47	31
8	1	13	86	31	0	69	34	58	49	29	50	63	54
8	2	14	89	26	0	62	55	66	91	45	55	75	56
8	3	10	64	9	0	44	44	55	73	43	41	78	43
8	4	10	69	17	0	43	27	53	51	25	27	50	41
9	1	10	74	32	0	65	34	56	58	48	44	69	53
9	2	14	91	34	0	56	39	63	74	40	58	84	54
9	3	11	72	16	0	48	31	61	54	25	36	81	39
9	4	12	60	21	0	46	41	39	67	41	46	61	38
10	1	16	97	43	0	64	35	55	60	41	48	86	55
10	2	13	84	27	0	46	46	71	78	43	55	85	61
10	3	7	64	19	0	50	35	71	76	45	44	66	48
10	4	9	72	19	0	48	41	40	52	34	26	46	36
11	1	21	98	25	0	67	26	65	57	46	52	70	53
11	2	33	112	18	0	56	41	65	77	34	53	95	47
11	3	7	71	11	0	59	45	75	62	26	32	76	59
11	4	7	60	16	0	50	26	43	68	33	23	58	25

Appendix 2: Raw data for galls diameter on plots treated with various biostimulants

		Time in months											
Treatments	**Replicates**	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**
1	1	0	0	0	4	8	5	0	0	1.3	3.6	7.8	1.3
1	2	0	0	0	2	4	2	2.7	0	0	2.4	3.9	0
1	3	0	0	1.9	4	5	3	0	0	0	0	5.2	0
1	4	0	0	0.8	0	0	0	1.5	2	0	0	0	0
2	1	0	0.7	2.5	3	4	0	0	2.2	4	4.3	4.3	4
2	2	0	0	0.7	4	2	2	0	0	0	0	1.8	0
2	3	0	1	4.5	10	6	2	2.3	0	0	0	6.2	0
2	4	0	0	3.4	5	3	3	0.6	2	3.2	0	2.8	3.2
3	1	0	1	2.3	0	2	0	1	3.2	4.5	5.4	2.3	4.5

3	2	0	0	0	4	5	4	5	0	0	0	4.7	0
3	3	0	1	4	4	4	0	0	0	0	0	4	0
3	4	0	0	0	7	9	8	0	4.4	0.9	6	9.4	0.9
4	1	0	0.9	2	2	1	2	1.6	3.1	4.1	4.2	1.1	4.1
4	2	0	1	1.6	5	6	3	2.9	0	0	0	5.7	0
4	3	0	0.6	2	3	3	0	3	4.9	5.6	0	2.7	5.6
4	4	0	0	1.2	7	7	0	2	3	3.9	0	6.6	3.9
5	1	0	0	1.1	3	5	3	2	0	0	1.8	4.9	0
5	2	0	0	2.1	8	7	0	0	0	0	0	6.6	0
5	3	0	0	0.7	2	3	0	0	0	0	1	2.5	0
5	4	0	0	1.1	4	3	3	4.7	3.2	0	1.5	2.5	0
6	1	0	0	1.1	3	4	2	2.4	0	0	0	3.8	0
6	2	0	0	0.5	4	6	6	1.4	2	2	2.8	6	2
6	3	0	0.3	0.8	2	3	0	0	0	0	0	3.1	0
6	4	0	0	0	0	1	4	5.4	7	6.5	0	0.7	6.5
7	1	0	0	0	2	4	6	2.5	0	0	0	3.6	0
7	2	0	0	0	5	8	5	2.3	0	1.7	0	7.8	1.7
7	3	0	0	1.5	2	0	0	0	0	0	0	0	0
7	4	0	0	1.2	7	5	3	0	0	0	0	4.8	0
8	1	0	0.9	2.7	9	12	11	1.6	1.8	0	2.1	11.5	0
8	2	0	0	1	3	4	0	1.1	1.5	1.7	0	4.3	1.7
8	3	0	0	0	2	3	2	2.9	3.5	4.2	0	3.1	4.2
8	4	0	0	2.6	10	10	11	0	0	0	0	10.4	0
9	1	0	0	1.4	2	3	3	3.9	4	1	1.8	2.7	1
9	2	0	0	1.3	2	3	0	0	0	0	0	3	0
9	3	0	0	0	2	2	0	0	0	0	0	2.4	0
9	4	0	0	0	6	7	1	1.1	0	0	0	7.4	0
10	1	0	0	0	3	5	4	0	0	2.5	3.4	4.7	2.5
10	2	0	0	1.5	2	0	0	0	1.8	2.7	1	0	2.7
10	3	0	1	2.4	5	6	2	3.5	5.9	0	4	5.9	0
10	4	0	0	0	0	2	3	0	3.7	2.8	0.9	1.5	2.8
11	1	5.5	0	0	1	5	8	6.6	4.2	2.4	3.4	5.3	2.4
11	2	0	0	1.6	7	8	8	3.5	0	0	0	8.3	0

11	3	0	0	0	0	0	2	3	0	1.7	1.2	0	1.7
11	4	0	0	1.8	4	0	0	0	0	0	0	0	0

Appendix 3: Raw data for fresh galls on plots treated with various biostimulants

	Time in months												
Treatments	**Replicates**	1	2	3	4	5	6	7	8	9	10	11	12
1	1	0	0	0	0	0	0	0	0	18	14	20	0
1	2	40	12	0	0	0	0	0	0	8	12	11	0
1	3	9.7	1.3	0	0	0	0	0	0	10	9	14	0
1	4	7.5	0	0	0	0	0	0	0	12	6	11	0
2	1	3.1	0	0	0	0	0	0	0	4	6	4	0
2	2	3.5	0	0	0	0	0	0	0	5	6	7	0
2	3	10.5	4	0	0	0	0	0	0	12	11	11	0
2	4	2.4	0	0	0	0	0	0	0	7	8	7	0
3	1	4.5	0	0	0	0	0	0	0	8	8	8	0
3	2	8.8	3.2	0	0	0	0	0	0	3	6	3	0
3	3	14.1	4.5	0	0	0	0	0	0	11	8	4	0
3	4	9.3	0	0	0	0	0	0	0	13	6	9	0
4	1	0	0	0	0	0	0	0	0	3	8	7	0
4	2	19.5	0.9	0	0	0	0	0	0	13	9	7	0
4	3	15.3	4.1	0	0	0	0	0	0	5	9	16	0
4	4	5.4	0	0	0	0	0	0	0	18	15	20	0
5	1	13.5	5.6	0	0	0	0	0	0	9	6	8	0
5	2	8.9	3.9	0	0	0	0	0	0	7	6	7	0
5	3	6.4	0	0	0	0	0	0	0	17	14	7	0
5	4	0	0	0	0	0	0	0	0	21	15	20	0
6	1	1	0	0	0	0	0	0	0	6	5	6	0
6	2	12.1	0	0	0	0	0	0	0	6	5	3	0
6	3	4.6	0	0	0	0	0	0	0	5	7	8	0
6	4	14.2	2	0	0	0	0	0	0	15	18	12	0
7	1	0	0	0	0	0	0	0	0	13	11	7	0
7	2	22.4	6.5	0	0	0	0	0	0	3	4	6	0

7	3	8.4	0	0	0	0	0	0	0	6	5	5	0
7	4	9.2	1.7	0	0	0	0	0	0	14	11	8	0
8	1	0	0	0	0	0	0	0	0	10	9	10	0
8	2	2.6	0	0	0	0	0	0	0	9	10	6	0
8	3	16.3	0	0	0	0	0	0	0	4	9	8	0
8	4	4.3	1.7	0	0	0	0	0	0	14	22	16	0
9	1	12.6	4.2	0	0	0	0	0	0	10	5	6	0
9	2	10.7	0	0	0	0	0	0	0	5	7	6	0
9	3	13.4	1	0	0	0	0	0	0	12	22	28	0
9	4	0	0	0	0	0	0	0	0	12	7	21	0
10	1	0	0	0	0	0	0	0	0	13	11	11	0
10	2	2.1	0	0	0	0	0	0	0	16	8	10	0
10	3	10.1	2.5	0	0	0	0	0	0	8	6	5	0
10	4	5.5	2.7	0	0	0	0	0	0	39	17	16	0
11	1	15.6	0	0	0	0	0	0	0	20	14	14	0
11	2	10.2	2.8	0	0	0	0	0	0	6	7	5	0
11	3	24.4	2.4	0	0	0	0	0	0	10	9	6	0
11	4	11.4	0	0	0	0	0	0	0	14	14	11	0

Appendix 4: Raw data for stem length on plots treated with various biostimulants

Treats	**Reps**	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**
1	1	54.4	55.2	56.0	55.4	51.6	57.4	54.4	55.2	56.0	55.4	51.6	57.0
1	2	57.0	57.4	58.0	54.0	55.7	56.7	57.0	57.4	58.0	54.0	55.7	60.3
1	3	60.3	56.5	54.8	58.2	51.7	57.2	60.3	56.5	54.8	58.2	51.7	60.5
1	4	60.5	54.1	55.4	55.3	51.1	58.9	60.5	54.1	55.4	55.3	51.1	53.0
2	1	53.0	56.8	55.1	54.9	54.0	57.5	53.0	56.8	55.1	54.9	54.0	61.1
2	2	61.1	61.6	54.3	53.4	55.8	59.0	61.1	61.6	54.3	53.4	55.8	57.0
2	3	57.0	56.9	55.8	57.3	52.8	57.3	57.0	56.9	55.8	57.3	52.8	57.0
2	4	57.0	59.0	58.1	62.2	56.0	61.5	57.0	59.0	58.1	62.2	56.0	60.5
3	1	60.5	59.0	54.5	56.8	54.1	61.2	60.5	59.0	54.5	56.8	54.1	63.8
3	2	63.8	58.6	56.6	56.7	54.0	66.8	63.8	58.6	56.6	56.7	54.0	55.7
3	3	55.7	56.6	57.3	56.3	55.7	54.1	55.7	56.6	57.3	56.3	55.7	57.2
3	4	57.2	58.7	57.8	57.3	56.1	61.6	57.2	58.7	57.8	57.3	56.1	61.5
4	1	61.5	55.0	54.1	53.9	55.4	58.1	61.5	55.0	54.1	53.9	55.4	57.5
4	2	57.5	57.0	53.6	56.1	54.8	57.2	57.5	57.0	53.6	56.1	54.8	57.0
4	3	57.0	56.9	56.8	55.0	55.3	58.1	57.0	56.9	56.8	55.0	55.3	56.9
4	4	56.9	59.4	56.2	57.6	54.4	62.0	56.9	59.4	56.2	57.6	54.4	56.5
5	1	56.5	55.3	55.6	56.9	55.5	59.0	56.5	55.3	55.6	56.9	55.5	58.1

5	2	58.1	55.3	56.1	60.6	57.6	61.9	58.1	55.3	56.1	60.6	57.6	56.0
5	3	56.0	61.0	56.3	54.7	54.8	56.6	56.0	61.0	56.3	54.7	54.8	53.5
5	4	53.5	57.2	55.1	58.9	57.3	53.8	53.5	57.2	55.1	58.9	57.3	58.2
6	1	58.2	62.3	59.2	55.7	58.4	59.3	58.2	62.3	59.2	55.7	58.4	56.4
6	2	56.4	57.0	57.1	56.0	57.1	57.5	56.4	57.0	57.1	56.0	57.1	59.5
6	3	59.5	55.8	55.7	58.4	54.3	54.4	59.5	55.8	55.7	58.4	54.3	65.7
6	4	65.7	56.2	54.3	56.5	57.1	57.8	65.7	56.2	54.3	56.5	57.1	56.3
7	1	56.3	59.5	51.2	56.4	55.6	56.4	56.3	59.5	51.2	56.4	55.6	60.2
7	2	60.2	56.8	54.5	57.0	56.4	56.9	60.2	56.8	54.5	57.0	56.4	54.0
7	3	54.0	55.9	54.2	58.4	56.3	59.3	54.0	55.9	54.2	58.4	56.3	59.8
7	4	59.8	55.3	58.1	52.5	55.7	57.5	59.8	55.3	58.1	52.5	55.7	57.5
8	1	57.5	55.9	55.2	54.1	53.6	56.9	57.5	55.9	55.2	54.1	53.6	61.9
8	2	61.9	58.3	57.9	57.8	53.6	56.8	61.9	58.3	57.9	57.8	53.6	57.2
8	3	57.2	56.7	56.4	54.9	56.7	56.9	57.2	56.7	56.4	54.9	56.7	56.6
8	4	56.6	58.6	54.5	58.3	50.2	56.5	56.6	58.6	54.5	58.3	50.2	57.1
9	1	57.1	59.5	57.1	57.7	54.1	57.5	57.1	59.5	57.1	57.7	54.1	55.8
9	2	55.8	57.7	54.9	57.2	52.3	57.2	55.8	57.7	54.9	57.2	52.3	55.5
9	3	55.5	53.5	55.4	56.2	55.5	55.6	55.5	53.5	55.4	56.2	55.5	63.5
9	4	63.5	58.7	57.9	59.3	53.7	55.6	63.5	58.7	57.9	59.3	53.7	61.8
10	1	61.8	59.9	57.6	57.5	55.6	62.2	61.8	59.9	57.6	57.5	55.6	54.7
10	2	54.7	57.2	55.2	59.9	55.6	58.8	54.7	57.2	55.2	59.9	55.6	55.1
10	3	55.1	58.2	59.1	56.4	57.1	55.5	55.1	58.2	59.1	56.4	57.1	58.7
10	4	58.7	53.7	58.6	58.2	56.3	59.3	58.7	53.7	58.6	58.2	56.3	62.4
11	1	62.4	59.7	57.8	56.7	55.0	55.2	62.4	59.7	57.8	56.7	55.0	60.6
11	2	60.6	59.0	56.9	58.9	55.4	58.9	60.6	59.0	56.9	58.9	55.4	58.2
11	3	58.2	56.3	58.6	54.6	52.7	57.5	58.2	56.3	58.6	54.6	52.7	58.0
11	4	58.0	57.0	56.0	55.9	55.3	57.0	58.0	57.0	56.0	55.9	55.3	58.0

Appendix 5: Raw data for stem weight on plots treated with various biostimulants

Treats	**Reps**	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**
1	1	28.4	26.1	26.6	22.6	20.4	23.0	28.4	26.1	26.6	22.6	20.4	23.0
1	2	24.2	25.6	26.3	24.1	25.3	23.0	24.2	25.6	26.3	24.1	25.3	23.0
1	3	27.9	24.1	22.9	25.0	22.4	20.3	27.9	24.1	22.9	25.0	22.4	20.3
1	4	23.9	20.9	21.3	22.7	22.1	21.7	23.9	20.9	21.3	22.7	22.1	21.7
2	1	21.8	24.2	23.5	22.1	24.3	24.2	21.8	24.2	23.5	22.1	24.3	24.2
2	2	25.5	28.9	22.5	20.7	21.3	23.2	25.5	28.9	22.5	20.7	21.3	23.2
2	3	23.6	25.1	23.8	25.4	24.8	22.9	23.6	25.1	23.8	25.4	24.8	22.9
2	4	24.0	23.5	29.3	29.9	20.0	22.7	24.0	23.5	29.3	29.9	20.0	22.7
3	1	30.6	25.8	25.7	26.0	20.4	26.8	30.6	25.8	25.7	26.0	20.4	26.8
3	2	26.2	24.9	25.9	24.3	21.3	29.6	26.2	24.9	25.9	24.3	21.3	29.6
3	3	24.9	22.9	27.5	26.2	22.0	18.8	24.9	22.9	27.5	26.2	22.0	18.8
3	4	22.5	27.7	23.1	24.1	26.9	21.4	22.5	27.7	23.1	24.1	26.9	21.4
4	1	31.5	25.0	25.3	21.7	24.0	23.6	31.5	25.0	25.3	21.7	24.0	23.6
4	2	26.5	26.5	21.4	26.8	21.5	24.3	26.5	26.5	21.4	26.8	21.5	24.3

4	3	21.7	26.2	24.0	24.8	23.5	21.1	21.7	26.2	24.0	24.8	23.5	21.1
4	4	24.8	25.9	28.4	26.4	23.5	28.7	24.8	25.9	28.4	26.4	23.5	28.7
5	1	23.2	27.4	23.4	27.6	27.3	23.2	23.2	27.4	23.4	27.6	27.3	23.2
5	2	22.4	18.5	26.8	26.9	23.4	24.7	22.4	18.5	26.8	26.9	23.4	24.7
5	3	27.5	25.0	26.2	21.3	23.9	27.4	27.5	25.0	26.2	21.3	23.9	27.4
5	4	18.4	24.3	25.4	29.5	26.9	22.4	18.4	24.3	25.4	29.5	26.9	22.4
6	1	24.6	37.2	28.9	23.4	27.5	22.8	24.6	37.2	28.9	23.4	27.5	22.8
6	2	21.8	21.8	26.5	25.3	25.3	22.2	21.8	21.8	26.5	25.3	25.3	22.2
6	3	26.3	21.6	20.7	24.6	24.2	19.4	26.3	21.6	20.7	24.6	24.2	19.4
6	4	26.9	24.9	23.8	23.5	24.0	20.8	26.9	24.9	23.8	23.5	24.0	20.8
7	1	25.9	28.5	30.9	23.8	24.3	23.0	25.9	28.5	30.9	23.8	24.3	23.0
7	2	27.6	21.3	26.7	23.7	23.7	19.8	27.6	21.3	26.7	23.7	23.7	19.8
7	3	15.7	25.5	21.4	26.8	24.6	20.7	15.7	25.5	21.4	26.8	24.6	20.7
7	4	25.9	18.4	25.0	23.4	22.9	21.6	25.9	18.4	25.0	23.4	22.9	21.6
8	1	27.6	24.2	24.5	24.8	25.7	21.8	27.6	24.2	24.5	24.8	25.7	21.8
8	2	24.7	22.5	28.8	22.9	21.1	21.1	24.7	22.5	28.8	22.9	21.1	21.1
8	3	21.4	22.9	24.7	21.9	22.1	20.0	21.4	22.9	24.7	21.9	22.1	20.0
8	4	20.8	23.7	22.9	25.3	20.0	21.1	20.8	23.7	22.9	25.3	20.0	21.1
9	1	26.1	24.5	25.5	30.8	22.5	26.5	26.1	24.5	25.5	30.8	22.5	26.5
9	2	20.7	24.3	23.0	25.7	21.3	24.6	20.7	24.3	23.0	25.7	21.3	24.6
9	3	24.8	26.1	24.3	22.0	21.0	19.4	24.8	26.1	24.3	22.0	21.0	19.4
9	4	28.7	25.7	25.1	23.9	22.8	21.9	28.7	25.7	25.1	23.9	22.8	21.9
10	1	33.7	26.6	27.3	27.4	23.4	24.9	33.7	26.6	27.3	27.4	23.4	24.9
10	2	22.5	21.8	22.2	23.8	24.6	20.4	22.5	21.8	22.2	23.8	24.6	20.4
10	3	22.8	25.2	26.2	22.1	23.1	22.1	22.8	25.2	26.2	22.1	23.1	22.1
10	4	27.1	21.6	25.3	21.7	27.6	19.9	27.1	21.6	25.3	21.7	27.6	19.9
11	1	28.4	24.2	26.3	27.1	23.1	22.3	28.4	24.2	26.3	27.1	23.1	22.3
11	2	30.3	26.6	26.3	24.9	24.0	24.9	30.3	26.6	26.3	24.9	24.0	24.9
11	3	27.4	23.1	26.3	21.1	22.0	19.7	27.4	23.1	26.3	21.1	22.0	19.7
11	4	31.3	28.1	24.7	24.4	22.8	18.6	31.3	28.1	24.7	24.4	22.8	18.6

Appendix 6: Raw data for rose head length on plots treated with various biostimulants

Treatments	Replicates	1	2	3	4	5	6	7	8	9	10	11	12
1	1	2.6	2.3	2.7	2.6	2.5	2.3	2.6	2.3	2.7	2.6	2.5	2.3
1	2	2.2	2.1	2.5	2.5	2.4	2.5	2.2	2.1	2.5	2.5	2.4	2.5
1	3	2.4	2.4	2.4	2.6	2.3	2.2	2.4	2.4	2.4	2.6	2.3	2.2
1	4	2.4	2.1	2.4	2.5	2.3	2.2	2.4	2.1	2.4	2.5	2.3	2.2
2	1	2.4	2.3	2.6	2.5	2.5	2.5	2.4	2.3	2.6	2.5	2.5	2.5
2	2	2.5	2.2	2.5	2.4	2.3	2.3	2.5	2.2	2.5	2.4	2.3	2.3
2	3	2.3	2.2	2.6	2.5	2.6	2.5	2.3	2.2	2.6	2.5	2.6	2.5
2	4	2.4	2.6	2.6	2.3	2.3	2.4	2.4	2.6	2.6	2.3	2.3	2.4
3	1	2.6	2.2	2.6	2.6	2.6	2.4	2.6	2.2	2.6	2.6	2.6	2.4
3	2	2.4	2.2	2.6	2.6	2.3	2.4	2.4	2.2	2.6	2.6	2.3	2.4
3	3	2.3	2.2	2.5	2.6	2.4	2.2	2.3	2.2	2.5	2.6	2.4	2.2

3	4	2.2	2.1	2.5	2.4	2.6	2.3	2.2	2.1	2.5	2.4	2.6	2.3
4	1	2.5	2.2	2.6	2.6	2.6	2.4	2.5	2.2	2.6	2.6	2.6	2.4
4	2	2.5	2.6	2.4	2.5	2.5	2.3	2.5	2.6	2.4	2.5	2.5	2.3
4	3	2.5	2.1	2.4	2.4	3.0	2.3	2.5	2.1	2.4	2.4	3.0	2.3
4	4	2.2	2.2	2.4	2.6	2.4	2.4	2.2	2.2	2.4	2.6	2.4	2.4
5	1	2.4	2.1	2.5	2.5	2.5	2.5	2.4	2.1	2.5	2.5	2.5	2.5
5	2	2.4	2.0	2.5	2.6	2.6	2.3	2.4	2.0	2.5	2.6	2.6	2.3
5	3	2.6	2.1	2.6	2.5	2.5	2.6	2.6	2.1	2.6	2.5	2.5	2.6
5	4	2.2	2.2	2.4	2.7	2.4	2.4	2.2	2.2	2.4	2.7	2.4	2.4
6	1	2.5	2.4	2.7	2.5	2.7	2.5	2.5	2.4	2.7	2.5	2.7	2.5
6	2	2.2	2.2	2.5	2.5	2.6	2.3	2.2	2.2	2.5	2.5	2.6	2.3
6	3	2.3	2.2	2.4	2.5	2.5	2.2	2.3	2.2	2.4	2.5	2.5	2.2
6	4	2.6	2.3	2.5	2.5	2.4	2.4	2.6	2.3	2.5	2.5	2.4	2.4
7	1	2.4	2.4	2.5	2.5	2.5	2.3	2.4	2.4	2.5	2.5	2.5	2.3
7	2	2.5	2.2	2.7	2.5	2.5	2.5	2.5	2.2	2.7	2.5	2.5	2.5
7	3	2.0	2.2	2.4	2.6	2.3	2.4	2.0	2.2	2.4	2.6	2.3	2.4
7	4	2.3	2.1	2.4	2.6	2.4	2.3	2.3	2.1	2.4	2.6	2.4	2.3
8	1	2.5	2.2	2.6	2.6	2.5	2.3	2.5	2.2	2.6	2.6	2.5	2.3
8	2	2.3	2.1	2.5	2.4	2.4	2.4	2.3	2.1	2.5	2.4	2.4	2.4
8	3	2.4	2.0	2.4	2.5	2.6	2.3	2.4	2.0	2.4	2.5	2.6	2.3
8	4	2.4	2.1	2.4	2.8	2.3	2.3	2.4	2.1	2.4	2.8	2.3	2.3
9	1	2.5	2.1	2.6	2.6	2.5	2.6	2.5	2.1	2.6	2.6	2.5	2.6
9	2	2.1	2.1	2.5	2.5	2.5	2.4	2.1	2.1	2.5	2.5	2.5	2.4
9	3	2.5	2.2	2.5	2.4	2.6	2.2	2.5	2.2	2.5	2.4	2.6	2.2
9	4	2.4	2.3	2.4	2.5	2.5	2.4	2.4	2.3	2.4	2.5	2.5	2.4
10	1	2.5	2.2	2.6	2.6	2.4	2.5	2.5	2.2	2.6	2.6	2.4	2.5
10	2	2.3	2.2	2.5	2.4	2.6	2.3	2.3	2.2	2.5	2.4	2.6	2.3
10	3	2.3	2.2	2.5	2.5	2.5	2.3	2.3	2.2	2.5	2.5	2.5	2.3
10	4	2.1	2.3	2.4	2.3	2.5	2.3	2.1	2.3	2.4	2.3	2.5	2.3
11	1	2.5	2.3	2.6	2.6	2.5	2.4	2.5	2.3	2.6	2.6	2.5	2.4
11	2	2.4	1.9	2.5	2.4	2.4	2.4	2.4	1.9	2.5	2.4	2.4	2.4
11	3	2.5	2.3	2.6	2.4	2.4	2.3	2.5	2.3	2.6	2.4	2.4	2.3
11	4	2.4	2.4	2.4	2.2	2.4	2.3	2.4	2.4	2.4	2.2	2.4	2.3

Appendix7: Raw data for fresh galls on plots treated with sterilizing agents.

	Time in months											
Treatments	**Reps**	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**
1	1	21	41	56	34	55	52	70	68	84	70	64	47
1	2	16	25	24	18	20	23	19	20	19	24	10	18
1	3	17	43	24	28	26	31	18	33	35	30	27	32
1	4	7	18	33	12	15	17	19	44	25	33	11	24
2	1	10	21	35	10	15	23	28	40	29	34	19	25
2	2	12	15	25	16	18	13	14	23	16	17	11	17
2	3	8	32	19	22	20	28	12	39	21	29	28	19
2	4	3	21	24	13	9	12	15	25	27	22	16	17
3	1	6	22	28	21	26	27	30	43	40	35	24	32
3	2	9	22	40	19	20	28	25	36	30	29	20	25
3	3	11	49	44	25	28	19	34	32	46	42	25	23
3	4	8	20	24	3	7	16	12	13	16	22	18	17
4	1	13	24	25	26	20	28	34	41	28	32	27	24
4	2	20	31	31	39	41	39	40	47	30	28	15	21
4	3	15	30	32	28	23	17	12	31	24	27	18	29
4	4	11	41	32	31	29	29	33	35	26	27	20	23
5	1	12	19	24	26	18	22	29	29	22	24	21	20
5	2	34	73	86	58	61	69	49	91	95	48	39	51
5	3	14	46	33	31	29	35	32	39	31	26	20	20
5	4	12	28	36	21	24	34	25	30	28	20	21	19
6	1	11	30	30	21	24	23	28	36	34	27	26	16
6	2	12	31	32	33	20	23	36	40	28	36	20	35
6	3	13	35	41	34	22	25	21	38	32	22	24	20
6	4	5	20	23	17	19	21	13	29	15	21	11	24
7	1	15	44	31	37	41	31	30	43	39	31	24	34
7	2	14	22	24	25	25	32	29	28	30	19	15	19
7	3	29	57	60	46	46	35	54	69	71	48	34	29
7	4	19	26	22	20	30	29	23	40	26	26	21	26

Appendix 8: Raw data for yield of roses on plots treated with various sterilizing agents.

		Time in months											
Treatments	**replicates**	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**
1	1	52	25	52	25	59	29	65	24	31	29	38	8
1	2	55	29	40	31	50	38	37	19	16	42	26	9
1	3	77	29	43	32	45	50	53	26	10	61	17	5
1	4	69	42	42	37	43	42	46	38	15	56	13	10
2	1	93	12	50	17	37	29	54	20	20	34	25	4
2	2	82	39	39	52	55	69	49	48	15	57	29	10
2	3	83	39	40	33	43	62	58	31	29	48	33	6
2	4	82	32	41	37	47	56	60	21	17	53	22	9
3	1	70	20	45	33	42	47	44	26	10	33	24	9
3	2	71	42	47	51	46	63	47	29	17	54	29	10
3	3	59	27	39	47	36	57	39	28	12	45	23	10
3	4	55	44	33	40	34	55	42	39	11	62	22	14
4	1	77	32	36	31	39	46	33	26	7	52	22	8
4	2	59	42	32	42	48	46	66	29	12	38	26	9
4	3	83	49	35	46	46	60	48	62	21	61	32	14
4	4	53	39	22	41	46	52	44	31	8	49	27	13
5	1	57	25	48	39	58	46	46	19	17	48	26	6
5	2	55	28	43	35	55	64	35	33	12	46	28	4
5	3	46	44	37	37	41	54	42	24	13	47	32	3
5	4	65	27	51	31	54	54	54	34	16	41	26	12
6	1	71	25	48	35	40	46	40	30	14	49	22	14
6	2	66	51	30	48	51	51	47	28	15	52	29	8
6	3	71	47	27	40	48	39	46	42	17	46	25	14
6	4	80	40	47	38	59	51	58	29	13	59	34	17
7	1	61	38	59	39	54	51	48	37	16	47	25	10
7	2	67	37	28	38	34	52	52	42	20	44	21	11
7	3	44	58	24	48	46	51	52	23	15	47	32	16

7	4	71	43	40	46	43	53	42	30	20	57	23	11

Appendix 9: Raw data for gall diameter on plots treated with various sterilizing agents.

		Time in months											
Treatments	**Replicates**	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**
1	1	4	7.4	6.4	0	0	0	0	3.2	6	9	6.4	11
1	2	2.8	19	27	10	5	9	0	0	0	0	1.2	4
1	3	4.6	9.8	12	0	0	2	5.8	0	0	3	2.2	3
1	4	9	20	11	4	7	7	3	3.6	2	0	0	0
2	1	0	0	0	5	0	0	0	0	0	0	0	4
2	2	0	0	0	0	0	0	0	0	0	0	0	0
2	3	0	0	1	0	0	0	0	0	0	1.2	2.2	4
2	4	0	1.4	4.2	5	0	0	0	0	0	0	0	0
3	1	0	0	0	0	0	6	6.6	8.6	4	9	10.8	6
3	2	0	5.6	10	7	3	4	0	0	0	2	11	16
3	3	0	0	2.6	4	3	6	0	0	0	0	0	0
3	4	0	0	0	0	0	2	0	0	0	0	0	0
4	1	9.6	23	21	9	5	13	26	15	3.2	4.8	5.2	0
4	2	7.4	18	14	9	6	8	9.6	7.8	3.6	0	0	0
4	3	8	21	11	14	10	15	19	0	3	5.4	6.6	0
4	4	8	23	29	10	5	10	7.6	2	4.8	6.2	0	0
5	1	4.6	16	27	30	2	8	17	13	4	5	5.2	3
5	2	0	8.6	21	9	11	13	7.8	13	2	5.4	0	0
5	3	4.6	20	24	15	13	23	17	19	4.4	0	2.8	6
5	4	5	23	35	23	9	15	12	7.2	0	0	0	0
6	1	8.6	15	22	14	6	13	3.2	4.4	0	0	0	0
6	2	0	3.6	8	12	5	9	10	0	0	0	3	7
6	3	3	15	24	29	19	25	13	6	2.6	2	6.2	0
6	4	0.4	14	25	21	9	8	0	0	0	0	0	0
7	1	11.4	21	23	24	28	37	28	7	6.4	9.6	9.2	0
7	2	4.2	23	25	20	21	20	2.2	3.2	2.2	5	5.2	5

7	3	6.2	17	27	19	23	29	4.2	11	0	0	0	0
7	4	9.6	23	29	16	14	22	13	1.4	3.8	5.4	10	0

Appendix 10: Raw data for gall weight on plots treated with various sterilizing agents

Gall weight at 12 months		
Treatments	**Replicates**	**Gall weight**
1	1	57.5
1	2	34.7
1	3	4
1	4	0
2	1	5.5
2	2	0
2	3	0
2	4	0.3
3	1	0
3	2	0
3	3	0
3	4	0
4	1	9.85
4	2	26.1
4	3	0.5
4	4	28.8
5	1	16.6
5	2	0
5	3	0
5	4	0
6	1	0
6	2	8.5
6	3	2.3
6	4	0

7	1	2.5
7	2	0
7	3	18.5
7	4	0

Appendix 11: Raw data on yield of roses on plots treated with various fungicides.

		Time in months											
Treatments	**Replicates**	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**
1	1	4	23	22	12	17	13	21	14	13	21	28	23
1	2	5	33	37	42	28	37	15	25	18	32	18	19
1	3	4	32	24	23	29	19	19	36	30	25	32	34
1	4	6	44	35	34	26	42	16	37	19	30	27	29
2	1	2	31	29	23	27	33	18	24	20	28	34	29
2	2	2	32	35	49	22	35	10	26	8	27	19	27
2	3	4	20	17	17	30	13	17	22	16	20	29	14
2	4	8	28	30	23	31	19	21	42	14	31	23	36
3	1	9	38	27	36	20	35	16	26	16	30	27	33
3	2	3	11	14	15	13	17	8	19	15	27	21	16
3	3	1	26	30	24	28	30	11	31	21	31	20	28
3	4	8	30	41	36	31	29	17	37	20	37	29	26
4	1	5	40	29	35	28	34	20	25	17	28	39	23
4	2	11	24	34	15	35	28	29	40	14	32	27	27
4	3	4	38	46	45	34	33	25	43	18	40	32	39
4	4	1	22	25	24	19	30	20	27	10	27	22	23

Appendix 12: Raw data on gall diameter on plots treated with various fungicides.

		Time in months									
Treatments	**Replicates**	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**
1	1	6.98	8.3	11	11	8.8	3.9	0.7	0.1	0.02	0
1	2	5.77	8	12	10	6.8	2.2	0.4	0.1	0.01	0
1	3	4.35	4.3	8.3	9.6	7.2	2	0.3	0.1	0.01	0
1	4	6.25	11	16	13	7.8	4.4	3.4	0.9	0.15	0

2	2	5.72	9.5	12	9.8	6.5	2.6	0.4	0.1	0.01	0
2	3	8.57	13	12	8.6	2.7	0.4	0.1	0	0	0
2	4	7.16	9	13	12	5.4	2.1	0.3	0.1	0.01	0
2	1	2.62	3.1	5.1	5.6	1.9	0.3	0.1	0	0	0
3	2	7.6	9.7	9.2	2.5	0.4	0.1	0	0	0	0
3	3	1.62	0.3	0	0	0	0	0	0	0	0
3	4	4.67	7.4	11	4.7	0.8	0.1	0	0	0	0
4	1	7.62	11	12	6.7	4	1.1	0.2	0	0.01	0
4	2	5.04	6.6	14	8.4	2.2	0.4	0.1	0	0	0
4	3	4.87	5.2	5.1	4.5	4.9	3.9	0.7	0.1	0.02	0
4	4	2.73	2.8	4.2	2.6	0.4	0.1	0	0	0	0
5	1	5.4	6.3	8.3	5.1	0.9	0.1	0	0	0	0
5	2	3.4	4.8	3.7	0.6	0.1	0	0	0	0	0
5	3	5.47	7.8	8.9	5.6	0.9	0.2	0	0	0	0
5	4	6.75	7.7	7.1	2.8	0.5	0.1	0	0	0	0

Appendix 13: Raw data on fresh galls on plots treated with various fungicides.

	Time in months											
Treatments	**Replicates**	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**
1	1	22	15	15	18	14	7	6	7	7	7	14	7
1	2	4	8	4	3	3	3	5	6	4	4	8	3
1	3	5	5	10	7	6	5	5	10	6	6	9	3
1	4	10	5	8	8	8	10	8	6	4	4	9	6
2	1	10	11	10	5	11	5	6	3	8	8	9	7
2	2	4	4	3	4	4	5	3	1	6	6	9	2
2	3	9	5	16	11	6	9	5	3	3	3	12	5
2	4	7	11	16	6	5	6	8	7	4	4	8	3
3	1	8	12	5	4	7	7	7	2	5	5	6	6
3	2	10	7	5	6	9	10	8	3	2	2	4	1
3	3	3	5	4	4	3	4	4	2	1	1	2	3
3	4	17	7	9	5	10	5	8	5	6	6	11	4
4	1	8	8	8	8	9	5	7	2	3	3	4	4

4	2	6	5	8	5	7	6	7	5	4	4	6	3
4	3	5	4	11	3	2	4	7	2	4	4	8	2
4	4	12	15	12	10	13	10	9	4	10	10	14	4

Appendix 14: Raw data on stem length of roses on plots treated with various fungicides.

		Time in months											
Treats	**Reps**	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**
1	1	65.2	57.2	56.0	59.4	56.3	58.5	59.5	57.0	56.3	52.5	58.2	57.9
1	2	55.1	55.3	56.8	55.5	59.1	57.0	59.9	54.0	53.2	66.7	55.3	57.0
1	3	58.8	49.1	62.8	55.8	60.2	65.0	56.2	54.6	57.2	56.8	55.9	57.6
1	4	59.0	58.4	58.0	52.5	67.0	63.4	53.5	61.7	63.8	61.4	60.5	54.2
2	1	51.9	52.8	63.1	53.9	61.1	56.0	58.0	61.0	55.2	50.8	51.8	62.3
2	2	53.8	56.0	58.8	52.3	65.6	56.0	60.8	61.0	50.8	50.2	57.0	54.2
2	3	53.3	50.4	58.7	54.8	62.9	60.4	60.8	54.9	59.0	67.5	58.5	59.1
2	4	65.5	51.7	65.4	58.3	54.0	61.1	54.0	57.1	56.2	59.5	56.5	58.3
3	1	59.2	56.6	58.0	51.4	61.5	60.8	50.5	58.1	54.1	46.0	53.1	58.7
3	2	55.5	67.0	51.1	57.0	73.5	64.3	55.6	66.0	53.1	50.7	64.5	60.4
3	3	54.2	49.5	52.0	49.7	59.0	60.3	61.0	57.0	56.8	52.0	58.2	57.2
3	4	55.9	55.8	57.6	52.8	57.6	56.6	57.7	53.0	54.1	53.1	63.5	62.4
4	1	55.8	50.5	56.2	54.7	64.3	59.0	57.9	49.4	59.0	50.4	55.7	52.9
4	2	60.7	53.6	53.4	52.2	53.5	58.9	61.5	50.0	55.3	56.6	53.5	52.6
4	3	58.7	63.5	56.3	57.6	63.2	62.6	51.8	53.4	56.3	53.1	57.4	59.7
4	4	57.3	56.3	55.5	72.0	59.2	51.0	54.7	52.0	53.5	60.0	59.0	59.6

Appendix 15: Raw data on roses head length on plots treated with various fungicides.

		Time in months											
Treats	**Reps**	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**
1	1	2.5	2.3	2.2	2.2	1.9	2.4	2.1	1.9	2.1	2.1	2.1	2.1
1	2	2.4	2.1	2.1	2.2	2.2	2.6	2.1	2.2	2.1	2.1	2.0	2.2
1	3	2.3	2.3	1.8	2.3	2.4	3.0	2.1	2.4	1.9	2.3	2.2	2.2
1	4	2.0	2.4	2.2	2.2	2.4	2.7	2.1	2.0	2.3	2.5	2.0	2.0
2	1	2.1	2.4	2.1	2.3	2.3	2.5	2.0	2.0	2.0	1.8	2.3	2.3
2	2	2.3	2.3	2.1	2.2	2.2	2.5	2.0	2.0	2.0	2.0	2.2	2.4
2	3	1.9	2.5	2.1	2.3	2.3	2.3	2.2	2.0	2.2	3.5	2.0	2.2

2	4	2.4	2.2	2.3	2.0	2.1	2.7	1.8	2.3	1.9	1.7	2.1	2.1
3	1	2.5	2.4	2.3	2.1	2.3	2.5	2.0	2.4	1.9	2.5	2.0	2.3
3	2	2.1	2.8	2.2	1.9	2.4	2.8	2.1	2.2	2.1	2.3	2.0	2.2
3	3	2.0	2.1	2.2	2.3	2.3	2.8	2.0	2.0	2.2	1.7	2.2	2.3
3	4	2.2	2.2	2.2	2.3	2.0	2.6	2.1	2.2	2.0	2.2	2.2	2.3
4	1	2.3	2.3	2.0	2.2	2.1	2.7	2.0	2.2	2.2	2.0	1.9	2.3
4	2	2.3	2.3	2.0	2.3	2.2	2.9	2.3	2.0	2.0	2.0	2.4	2.4
4	3	2.3	2.3	2.2	2.3	2.4	2.7	2.1	2.1	2.0	1.9	2.3	2.3
4	4	2.1	2.2	2.2	2.6	2.3	2.4	2.0	2.1	1.8	2.3	2.3	2.3

Appendix 16: Raw data of roses head width on plots treated with various fungicides.

		Time in months											
Treats	**Reps**	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**
1	1	2.5	2.3	2.1	2.2	1.9	2.4	2.1	1.9	2.1	2.1	2.1	2.1
1	2	2.4	2.1	2.1	2.2	2.2	2.6	2.1	2.2	2.1	2.1	2.0	2.2
1	3	2.3	2.3	1.8	2.3	2.4	3.0	2.1	2.4	1.9	2.3	2.2	2.2
1	4	2.0	2.4	2.2	2.2	2.4	2.7	2.1	2.0	2.3	2.5	2.0	2.0
2	1	2.1	2.4	2.1	2.3	2.3	2.5	2.0	2.0	2.0	1.8	2.3	2.3
2	2	2.3	2.3	2.1	2.2	2.2	2.5	2.0	2.0	2.0	2.0	2.2	2.4
2	3	1.9	2.5	2.1	2.3	2.3	2.3	2.2	2.0	2.2	3.5	2.0	2.2
2	4	2.4	2.2	2.3	2.0	2.1	2.7	1.8	2.3	1.9	1.7	2.1	2.1
3	1	2.5	2.4	2.3	2.1	2.3	2.5	2.0	2.4	1.9	2.5	2.0	2.3
3	2	2.1	2.8	2.2	1.9	2.4	2.8	2.1	2.2	2.1	2.3	2.0	2.2
3	3	2.0	2.1	2.2	2.3	2.3	2.8	2.0	2.0	2.2	1.7	2.2	2.3
3	4	2.2	2.2	2.2	2.3	2.0	2.6	2.1	2.2	2.0	2.2	2.2	2.3
4	1	2.3	2.3	2.0	2.2	2.1	2.7	2.0	2.2	2.2	2.0	1.9	2.3
4	2	2.3	2.3	2.0	2.3	2.2	2.9	2.3	2.0	2.0	2.0	2.4	2.4
4	3	2.3	2.3	2.2	2.3	2.4	2.7	2.1	2.1	2.0	1.9	2.3	2.3
4	4	2.1	2.2	2.2	2.6	2.3	2.4	2.0	2.1	1.8	2.3	2.3	2.3

Appendix 17: Raw data on stem weight of roses on plots treated with various fungicides.

		Time in months											
Treats	**Reps**	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**
1	1	24	24	29	27	25	29	29	32	26	25	29	32
1	2	29	24	29	22	29	29	26	26	22	40	25	26
1	3	27	26	28	30	35	38	28	25	27	31	27	25
1	4	28	28	27	24	30	29	27	30	31	36	35	27
2	1	22	28	41	29	29	26	24	35	28	20	21	32
2	2	25	24	29	27	36	26	29	35	32	25	28	29
2	3	21	26	29	26	34	28	28	24	29	40	30	35
2	4	41	23	32	29	20	27	21	28	29	47	33	28
3	1	24	26	29	26	29	32	19	23	22	19	22	37
3	2	27	44	22	23	36	32	24	29	26	21	35	28
3	3	28	21	20	24	29	30	26	22	28	22	29	30
3	4	25	28	30	26	26	26	25	25	27	31	35	30
4	1	25	25	24	30	35	33	27	30	35	24	24	24
4	2	30	23	21	22	27	29	28	28	29	28	27	28
4	3	30	27	30	28	28	31	21	22	31	23	31	32
4	4	28	25	26	34	28	21	28	22	26	38	31	32

Appendix 18: Raw data on yield of roses on plots treated with various biologicals.

		Time in months											
Treatments	**Replicates**	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**
1	1	5	29	27	0	0	29	16	24	19	24	18	0
1	2	6	43	58	0	0	32	47	36	38	28	19	0
1	3	16	39	27	0	0	42	56	28	35	27	36	0
1	4	9	49	42	0	0	50	44	36	37	32	40	0
2	1	6	37	54	0	0	30	24	20	24	19	30	0
2	2	10	53	46	0	0	36	39	28	39	21	28	0
2	3	17	49	31	0	0	51	36	37	21	28	30	0

2	4	13	54	28	0	0	46	34	35	34	24	50	0
3	1	5	43	54	0	0	25	32	28	24	33	19	0
3	2	10	48	43	0	0	31	44	32	42	26	31	0
3	3	17	59	31	0	0	53	43	37	23	27	25	0
3	4	7	66	47	0	0	46	43	34	38	29	41	0
4	1	7	59	42	0	0	33	26	29	35	23	41	0
4	2	10	43	47	0	0	30	27	23	33	25	16	0
4	3	7	51	40	0	0	56	46	53	40	33	33	0
4	4	12	50	33	0	0	28	42	27	28	30	30	0
5	1	14	34	39	0	0	38	29	34	26	35	33	0
5	2	13	41	44	0	0	46	41	32	43	24	32	0
5	3	10	48	34	0	0	39	48	38	18	30	22	0
5	4	10	31	28	0	0	29	34	28	18	18	20	0

Appendix 19: Raw data for fresh galls on plots treated with various biologicals.

		Time in months											
Treatments	**Replicates**	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**
1	1	2	4	4	7	5	13	10	14	14	5	9	8
1	2	6	9	7	7	7	7	15	9	4	9	2	11
1	3	2	8	2	4	5	5	10	8	6	6	6	7
1	4	4	7	2	1	9	7	6	12	9	7	10	10
2	1	2	5	2	10	2	6	9	7	7	7	8	4
2	2	4	2	5	5	5	7	11	10	6	6	7	7
2	3	3	7	2	3	5	6	9	10	7	5	10	9
2	4	3	10	4	4	13	4	14	10	9	11	8	3
3	1	4	7	5	3	7	5	11	9	9	7	4	2
3	2	5	7	3	12	6	5	13	13	10	9	11	9
3	3	5	5	4	1	9	6	10	10	8	2	8	3
3	4	4	4	1	4	6	7	12	7	8	5	8	6
4	1	5	2	5	5	3	7	3	11	5	4	4	3
4	2	4	5	5	6	5	12	4	7	8	3	8	4

4	3	3	11	4	8	7	11	11	8	10	12	9	5
4	4	4	8	3	9	3	9	6	14	7	6	5	6
5	1	1	5	2	3	8	13	13	14	11	7	5	6
5	2	5	6	2	5	8	14	8	12	9	8	7	6
5	3	3	1	4	4	7	9	8	13	7	6	7	3
5	4	11	13	10	25	7	17	11	15	13	19	21	15

Appendix 20: Raw data for fresh galls on plots treated with various biologicals (pot trial).

	Time in months	
Treatments	Replicates	Fresh galls
1	1	0
1	2	0
1	3	0
2	1	0
2	2	1
2	3	0
3	1	0
3	2	4
3	3	1
4	1	1
4	2	0
4	3	0
6	1	1
6	2	0
6	3	4

Appendix 21: Raw data on rose stem length on plots treated with various biologicals.

		Time in months											
Treats	**Reps**	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**
1	1	45.0	60.0	51.8	57.83	58.2	58	52.0	56.3	58.0	54.7	55.2	69.0
1	2	45.0	58.5	54.0	58.43	57.4	58.9	60.6	55.2	59.9	54.6	54.1	55.6
1	3	52.5	53.0	51.0	52.81	59.7	58.5	55.2	57.3	52.4	57.5	53.5	55.6
1	4	58.0	51.8	55.8	57.61	58.6	66.1	64.7	57.4	52.3	55.9	54.3	58.8
2	1	58.0	52.8	54.5	55.96	55.3	53.1	55.2	53.0	49.1	55.0	54.0	59.9
2	2	57.5	52.7	52.6	58.31	62.2	59.0	57.2	59.6	63.1	55.8	54.6	55.5
2	3	55.0	55.0	57.5	60.50	62.2	67.8	54.2	55.1	54.6	60.1	49.9	57.3
2	4	55.5	52.4	56.7	60.25	56.3	59.1	54.9	56.0	57.4	57.7	56.0	63.2
3	1	52.5	60.4	46.1	56.50	56.9	53.3	58.8	57.2	61.3	62.5	57.1	63.0
3	2	53.5	55.0	52.8	54.20	61.5	64.4	57.3	54.3	53.4	61.4	54.5	55.9
3	3	44.8	53.0	57.2	57.50	61.8	58.0	57.0	53.4	56.0	57.3	51.7	58.4
3	4	54.0	57.6	53.7	57.42	54.6	58.6	54.6	54.3	54.1	54.6	54.2	53.4
4	1	54.5	53.7	52.1	55.50	58.7	56.5	55.3	52.6	54.0	55.0	52.6	55.1
4	2	55.0	58.8	52.5	58.72	57.5	58.3	59.8	53.3	49.6	50.3	56.6	63.9
4	3	55.0	56.1	55.2	59.61	56.3	61.3	58.5	57.2	58.2	62.4	54.7	58.0
4	4	53.2	56.4	54.5	56.08	60.1	60.5	57.5	55.5	54.7	59.9	57.4	62.4
5	1	53.5	53.6	57.3	57.25	58.0	61.3	58.1	57.5	53.9	60.5	53.4	56.6
5	2	54.5	55.1	50.5	58.19	59.4	58.8	58.1	58.6	54.2	56.1	55.5	55.7
5	3	58.8	51.1	59.8	52.92	54.8	57.6	53.7	55.8	54.4	53.1	50.5	52.0
5	4	41.0	47.8	50.8	58.63	57.2	58.3	50.0	55.2	53.9	54.8	57.0	55.5

Appendix 22: Raw data on roses stem weight on plots treated with various biologicals.

		Time in months											
Treats	**Reps**	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**
1	1	25.0	32.8	23.2	26.9	26.2	26.6	25.9	23.0	24.8	27.5	22.0	27.5
1	2	17.0	34.0	25.7	29.1	26.9	28.4	29.3	23.5	28.9	24.2	26.3	24.2
1	3	27.6	27.9	22.7	26.2	31.8	28.9	24.0	28.8	26.8	30.3	27.9	30.3
1	4	29.8	27.1	32.9	20.2	32.9	37.4	29.6	25.6	23.8	26.9	23.7	26.9
2	1	28.0	20.8	25.9	27.8	26.6	23.5	26.5	22.0	21.2	26.5	23.5	28
2	2	35.0	25.0	22.0	30.3	29.0	30.0	28.0	27.0	26.2	28.9	26.2	28.9
2	3	29.1	26.9	29.6	35.6	32.2	37.6	28.3	27.1	29.9	35.5	27.4	35.5
2	4	25.2	22.4	30.3	35.8	27.5	30.0	25.1	28.5	27.6	28.8	28.8	28.8
3	1	23.8	33.2	24.7	28.5	34.5	26.5	27.9	31.4	29.8	28.7	29.3	28.7
3	2	28.1	24.4	27.1	26.7	32.9	33.8	34.5	25.4	26.1	30.9	25.8	30.9
3	3	23.5	24.6	33.5	20.6	32.6	30.5	29.3	23.2	24.2	26.8	28.1	26.8
3	4	24.5	33.9	24.6	25.8	31.7	28.6	29.7	25.8	22.8	26.3	28.1	26.3

4	1	18.0	27.2	26.7	22.4	26.0	26.2	26.2	20.7	22.0	28.9	25.0	28.9
4	2	30.3	34.4	29.5	27.7	29.5	29.9	30.9	24.8	20.8	29.7	29.6	29.7
4	3	32.3	26.0	30.1	30.7	25.7	25.3	25.5	23.4	26.9	33.1	27.9	33.1
4	4	28.0	28.1	23.3	32.3	29.4	30.9	29.5	24.6	24.8	31.2	31.5	31.2
5	1	32.0	22.8	30.3	29.8	27.7	27.3	26.6	25.4	24.1	29.3	27.8	29.3
5	2	33.5	27.5	29.0	28.0	29.6	31.1	27.2	27.3	25.6	26.1	26.7	26.1
5	3	33.8	26.7	23.0	23.5	26.2	26.6	26.9	25.1	27.8	27.9	27.5	27.9
5	4	19.2	20.7	24.7	27.3	27.8	26.5	23.2	27.2	21.4	26.5	27.0	26.5

Appendix 23: Raw data on roses head width on plots treated with various biologicals.

		Time in months											
Treats	**Reps**	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**
1	1	4.5	4.1	4.4	4.4	3.7	4.0	4.2	3.8	3.9	3.7	3.9	3.9
1	2	4.7	4.5	4.3	4.0	3.7	3.8	4.0	4.0	4.3	3.7	4.0	4.3
1	3	5.0	4.2	4.3	3.8	3.9	3.9	4.0	4.2	4.3	3.9	4.1	4.2
1	4	5.0	4.3	4.5	4.1	4.1	4.2	4.3	4.1	4.1	3.9	4.1	4.1
2	1	4.7	4.6	4.3	4.0	4.0	4.0	4.0	4.0	4.0	3.8	4.1	4.0
2	2	4.7	4.3	4.0	4.1	4.2	4.3	4.1	4.0	4.0	4.2	4.0	4.1
2	3	4.6	4.4	4.6	4.2	3.9	4.1	4.0	4.0	4.2	4.1	4.0	3.8
2	4	4.5	4.1	3.8	4.0	4.1	4.3	4.1	3.9	4.1	4.2	4.2	4.3
3	1	4.5	4.4	4.2	4.1	3.8	3.6	3.8	4.0	4.0	4.3	4.2	4.5
3	2	4.5	4.1	4.3	4.0	4.1	4.2	4.2	3.9	4.0	4.2	4.1	4.2
3	3	4.4	4.1	3.9	3.9	4.1	4.2	4.2	4.1	4.2	4.1	4.0	4.4
3	4	4.4	4.5	3.8	3.9	4.0	4.1	4.2	4.1	4.0	3.9	4.1	3.7
4	1	4.8	4.3	4.1	4.0	3.8	4.0	4.2	3.7	3.6	3.9	3.8	4.1
4	2	4.5	4.3	4.3	4.1	4.0	4.2	4.3	3.8	3.8	4.1	4.2	4.6
4	3	4.0	4.5	4.4	4.1	4.0	3.9	4.2	4.1	4.0	3.8	4.0	4.3
4	4	4.4	4.1	4.3	4.4	4.8	4.5	4.1	3.9	3.9	4.1	4.2	4.3
5	1	5.0	4.3	4.1	4.4	4.0	4.3	4.2	3.7	3.9	4.1	4.1	4.4
5	2	4.7	4.2	4.0	3.9	3.9	4.2	4.0	4.0	4.0	4.3	4.2	4.3
5	3	4.9	4.4	4.5	3.8	3.9	4.0	4.2	4.0	4.1	3.9	3.9	3.8
5	4	4.0	4.3	3.8	4.3	4.0	4.2	4.2	4.0	4.0	4.0	4.2	3.9

Appendix 24: Raw data on roses head length on plots treated with various biologicals.

		Time in months											
Treats	**Reps**	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**
1	1	4.5	4.1	4.4	4.4	3.7	4.0	4.2	3.8	3.9	3.7	3.9	3.9
1	2	4.7	4.5	4.3	4.0	3.7	3.8	4.0	4.0	4.3	3.7	4.0	4.3
1	3	5.0	4.2	4.3	3.8	3.9	3.9	4.0	4.2	4.3	3.9	4.1	4.2
1	4	5.0	4.3	4.5	4.1	4.1	4.2	4.3	4.1	4.1	3.9	4.1	4.1
2	1	4.7	4.6	4.3	4.0	4.0	4.0	4.0	4.0	4.0	3.8	4.1	4.0
2	2	4.7	4.3	4.0	4.1	4.2	4.3	4.1	4.0	4.0	4.2	4.0	4.1
2	3	4.6	4.4	4.6	4.2	3.9	4.1	4.0	4.0	4.2	4.1	4.0	3.8
2	4	4.5	4.1	3.8	4.0	4.1	4.3	4.1	3.9	4.1	4.2	4.2	4.3
3	1	4.5	4.4	4.2	4.1	3.8	3.6	3.8	4.0	4.0	4.3	4.2	4.5
3	2	4.5	4.1	4.3	4.0	4.1	4.2	4.2	3.9	4.0	4.2	4.1	4.2
3	3	4.4	4.1	3.9	3.9	4.1	4.2	4.2	4.1	4.2	4.1	4.0	4.4
3	4	4.4	4.5	3.8	3.9	4.0	4.1	4.2	4.1	4.0	3.9	4.1	3.7
4	1	4.8	4.3	4.1	4.0	3.8	4.0	4.2	3.7	3.6	3.9	3.8	4.1
4	2	4.5	4.3	4.3	4.1	4.0	4.2	4.3	3.8	3.8	4.1	4.2	4.6
4	3	4.0	4.5	4.4	4.1	4.0	3.9	4.2	4.1	4.0	3.8	4.0	4.3
4	4	4.4	4.1	4.3	4.4	4.8	4.5	4.1	3.9	3.9	4.1	4.2	4.3
5	1	5.0	4.3	4.1	4.4	4.0	4.3	4.2	3.7	3.9	4.1	4.1	4.4
5	2	4.7	4.2	4.0	3.9	3.9	4.2	4.0	4.0	4.0	4.3	4.2	4.3
5	3	4.9	4.4	4.5	3.8	3.9	4.0	4.2	4.0	4.1	3.9	3.9	3.8
5	4	4.0	4.3	3.8	4.3	4.0	4.2	4.2	4.0	4.0	4.0	4.2	3.9

Appendix 25: Analysis of variance for various biostimulants

Analysis of variance of gall diameter of various biostimulants.

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Replication	3	99.61	33.20	0.52	
Treatments	10	774.27	77.43	1.21	0.323
Residual	30	1915.89	63.86		
Total	**43**	**2789.77**			

Analysis of variance of yield of various biostimulants

	Source of variation	d.f	s.s	m.s	F. value	Prob
1	Replication	3	84684.795	28228.265	18.5290	0.0000
2	Treatments	10	24175.500	2417.550	1.5869	0.1586
3	Error	30	45703.955	1523.465		
	Total	43	154564.250			

Coefficient of Variation: 8.25%

s_ for means group 1: 11.7685 Number of Observations: 11
y

s_ for means group 2: 19.5158 Number of Observations: 4
y

Analysis of variance of fresh galls of various biostimulants – pot trials

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
1. Replication	3	48.18	16.06	1.23	
2. Treatment	10	199.00	19.90	1.52	0.181
3. Error	30	392.82	13.09		
Total	**43**	**640.00**			

Stratum standard errors and coefficients of variation

Stratum	d.f.	s.e.	cv%
Replicates	3	1.208	15.1
Replicate.*Units*	30	3.619	45.2

Analysis of variance for gall diameter of various biostimulants - pots

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Replication	3	10.037	3.346	0.79	
Treatments	10	50.615	5.061	1.20	0.333
Error	30	127.033	4.234		
Total	**43**	**187.684**			

Stratum standard errors and coefficients of variation

Stratum	d.f.	s.e.	cv%
Replicates	3	0.55	15.4

Replicates*Units*	30	2.06	57.6

Analysis of variance for stem length of various biostimulants

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Replication	3	39.171	13.057	2.60	
Treatments	10	154.392	15.439	3.07	<.001
Months	11	686.194	62.381	12.40	<.001
Treatments. Months	110	352.395	3.204	0.64	0.997
Error	393	1977.341	5.031		
Total	**527**	**3209.492**			

Stratum standard errors and coefficients of variation

Stratum	d.f.	s.e.	cv%
Replicates	3	0.31	0.6
Replicates. *Units*	393	2.24	3.9

Analysis of variance for head length of various biostimulants

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Replicates stratum	3	0.96382	0.32127	9.92	
Treatments	10	0.84259	0.08426	2.60	0.005
Months	11	6.70664	0.60969	18.82	<.001
Treatments. Months	110	1.90409	0.01731	0.53	1.000
Error	393	12.73097	0.03239		

Total	**527**	**23.14812**

Stratum standard errors and coefficients of variation

Stratum	**d.f.**	**s.e.**	**cv%**
Replicates	3	0.05	1.2
Replicates. *Units*	393	0.18	4.3

Analysis of variance for head width of various biostimulants

Source of variation	**d.f.**	**s.s.**	**m.s.**	**v.r.**	**F pr.**
Replicates stratum	3	0.75264	0.25088	18.89	
Treatments	10	0.17391	0.01739	1.31	0.223
Months	11	5.79873	0.52716	39.70	<.001
Treatments. Months	110	1.43897	0.01308	0.99	0.528
Residual	393	5.21871	0.01328		
Total	**527**	**13.38296**			

Stratum standard errors and coefficients of variation

Stratum	**d.f.**	**s.e.**	**cv%**
Replicates	3	0.04	1.8
Replicates. *Units*	393	0.12	4.8

Analysis of variance for stem weight of various biostimulants

Source of variation	**d.f.**	**s.s.**	**m.s.**	**v.r.**	**F pr.**
Replicates stratum	3	314.834	104.945	15.36	
Treatments	10	141.845	14.185	2.08	0.025

Months	11	521.166	47.379	6.94	<.001
Treatments. Months	110	575.552	5.232	0.77	0.953
Error	393	2684.629	6.831		
Total	**527**	**4238.025**			

Stratum standard errors and coefficients of variation

Stratum	**d.f.**	**s.e.**	**cv%**
Replicates	3	0.89	3.7
Replicates. *Units*	393	2.61	10.8

Appendix 26: Analysis of variance for various sterilizing agents

Analysis of variance for gall weight of various sterilising - pots

Source of variation	**d.f**	**s.s**	**m.s.**	**Value**	**F. Prob**
1 Replication	3	251.496	83.832	0.3332	
2 Factor A	6	4742.070	592.759	2.3562	0.0499
3 Error	18	6037.721	251.572		
Total	27	11031.288			

Coefficient of Variation: 140.88%

s_ for means group 1: 5.2870 Number of Observations: 7
y

s_ for means group 2: 7.9305 Number of Observations: 4
y

Analysis of variance for yield of various sterilising - Field

Source of variation		d.f	s.s	m.s.	F. Prob
1	Replication	3	15526.80	5175.361	2.9025
2	Factor A	6	17116.389	2139.549	1.1999
3	Error	18	42793	2139.549	
	Total	27	11031.288		

Coefficient of Variation: 8.85 %

s_ for means group 1: 14.0754 Number of Observations: 7
y

s_ for means group 2: 21.1131 Number of Observations: 4
y

Analysis of variance for head length of various sterilising - Field

Source of variation		d.f	s.s	m.s.	F. Prob
1	Replication	3	0.047	0.016	0.9500
2	Factor A	6	0.229	0.029	1.7167
3	Error	18	0.400	0.017	
	Total	27	0.676		

Coefficient of Variation: 3.02 %

s_ for means group 1: 0.0430 Number of Observations: 7
y

s_ for means group 2: 0.0645 Number of Observations: 4
y

Analysis of variance for stem length of various sterilising - Field

Source of variation		d.f	s.s	m.s.	F. Prob
1	Replication	3	16.528	5.509	3.0949
2	Factor A	6	9.722	1.215	0.6827
3	Error	18	42.722	1.780	
	Total	27	68.972		

Coefficient of Variation: 2.28 %

s_ for means group 1: 0.4447 Number of Observations: 7
y

s_ for means group 2: 0.6671 Number of Observations: 4
y

Analysis of variance for stem weight of various sterilising - Field

Source of variation		d.f	s.s	m.s.	F. Prob
1	Replication	3	7.417	2.1472	0.6605
2	Factor A	6	13.500	1.688	0.4508
3	Error	18	89.833	3.743	
	Total	27	68.972		

Coefficient of Variation: 6.89 %

s_ for means group 1: 0.6449 Number of Observations: 7
y

s_ for means group 2: 0.9673 Number of Observations: 4
y

Analysis of variance for fresh galls of various sterilising - Field

	Source of variation	d.f	s.s	m.s.	F. Prob
1	Replication	3	35449.889	11816.630	0.604
2	Factor A	6	120128.500	15016.063	0.7675
3	Error	18	469566.611	19565.25	
	Total	27	625145.00		

Coefficient of Variation: 37.32 %

s_ for means group 1: 46.6253 Number of Observations: 7
y

s_ for means group 2: 69.9380 Number of Observations: 4
y

Analysis of variance for gall weight of various sterilising - pots

	Source of variation	d.f	s.s	m.s.	F. Prob
1	Replication	3	35449.889	11816.63	0.604
2	Factor A	6	120128.500	15016.063	0.765
3	Error	18	6037.721	251.572	
	Total	27	11031.288		

Coefficient of Variation: 37.2 %

s_ for means group 1: 46.6253 Number of Observations: 7
y

s_ for means group 2: 69.9380 Number of Observations: 4

Appendix 27: Analysis of variance for various fungicides.

Analysis of variance of stem length on various Fungicides

Source of variation	**d.f.**	**s.s.**	**m.s.**	**v.r.**	**F** pr.
Reps stratum	3	60.24	20.08	1.13	
Treats	3	57.04	19.01	1.07	0.362
Months	11	528.43	48.04	2.71	0.003
Treats. Months	33	683.47	20.71	1.17	0.262
Error	141	2496.52	17.71		
Total	**191**	**3825.71**			

Stratum standard errors and coefficients of variation

Stratum	**d.f.**	**s.e.**	**cv%**
Reps	3	0.65	1.1
Reps.*Units*	141	4.21	7.4

Analysis of variance of rose head length on various Fungicides.

Source of variation	**d.f.**	**s.s.**	**m.s.**	**v.r.**	**F pr.**
Reps stratum	3	0.09113	0.03038	0.75	
Treats	3	0.01364	0.00455	0.11	0.953
Months	11	3.97142	0.36104	8.89	<.001
Treats. Months	33	0.61557	0.01865	0.46	0.995
Error	141	5.72728	0.04062		
Total	**191**	**10.41903**			

Stratum standard errors and coefficients of variation

Stratum	d.f.	s.e.	cv%
Reps	3	0.03	1.1
Reps.*Units*	141	0.20	9.1

Analysis of variance of rose head width on various Fungicides.

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Reps stratum	3	0.09528	0.03176	0.78	
Treats	3	0.01423	0.00474	0.12	0.950
Months	11	3.98644	0.36240	8.95	<.001
Treats. Months	33	0.62576	0.01896	0.47	0.994
Error	141	5.70857	0.04049		
Total	**191**	**10.43028**			

Stratum standard errors and coefficients of variation

Stratum	d.f.	s.e.	cv%
Reps	3	0.03	1.2
Reps.*Units*	141	0.20	9.1

Analysis of variance of roses stem weight on various Fungicides

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Reps stratum	3	61.77	20.59	0.96	
Treats	3	92.69	30.90	1.43	0.235
Months	11	329.01	29.91	1.39	0.184
Treats. Months	33	706.29	21.40	0.99	0.486
Error	141	3036.21	21.53		
Total	**191**	**4225.97**			

Stratum standard errors and coefficients of variation

Stratum	d.f.	s.e.	cv%
Reps	3	0.65	2.3
Reps.*Units*	141	4.64	16.6

Analysis of variance of galls diameter on various Fungicides –

Source of variation	**d.f.**	**s.s.**	**m.s.**	**v.r.**	**F pr.**
Replicates	3	23.810	7.937	2.36	
Treatment	3	27.003	4.500	1.34	0.292
Error	9	60.607	3.367		
Total	**15**	**111.419**			

Stratum standard errors and coefficients of variation

Stratum	**d.f.**	**s.e.**	**cv%**
Replicates	3	1.06	25.2
Replicate.*Treatments	9	1.83	43.4

Analysis of variance of fresh galls on various Fungicides – pots

Source of variation	**d.f.**	**s.s.**	**m.s.**	**v.r.**	**F pr.**
Replicates	3	13.86	4.62	0.27	
Treatments	3	146.71	24.45	1.42	0.261
Error	9	310.14	17.23		
Total	**15**	**470.71**			

Stratum standard errors and coefficients of variation

Stratum	d.f.	s.e.	cv%
Replicates	3	0.812	9.9
Replicates.*Treatments	9	4.151	50.5

Appendix 28: Analysis of variance for various biologicals

Analysis of variance of yield on various biologicals - field

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Replicates	3	14048.	4683.	0.65	
Treatments	5	6354.	1059.	0.15	0.987
Error	15	128945.	7164.		
Total	**23**	**149347.**			

Stratum standard errors and coefficients of variation

Stratum	d.f.	s.e.	cv %
Replicates	3	25.9	5.1
Replicates.*Units*	15	84.6	16.8

Analysis of variance of gall diameter on various biologicals - field

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Replicates	3	9.6445	3.2148	4.99	
Treatments	5	5.9526	0.9921	1.54	0.222
Error	15	11.5949	0.6442		
Total	**23**	**27.1920**			

Stratum standard errors and coefficients of variation

Stratum	d.f.	s.e.	cv%
Replicates	3	0.68	17.4
Replicates * Treatments	15	0.80	20.6

Analysis of variance of fresh galls on various biologicals – field

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Replicates	3	2145.	715.	0.67	
Treatments	5	7049.	1175.	1.11	0.396
Error	15	19075.	1060.		
Total	23	28269.			

Stratum standard errors and coefficients of variation

Stratum	d.f.	s.e.	cv%
Replicates	3	10.11	8.8
Replicates * Treatments	15	32.55	28.2

Analysis of variance of stem length on various biologicals

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Reps stratum	3	10.47	3.49	0.31	
Treats	4	59.36	14.84	1.30	0.271
Months	11	883.94	80.36	7.06	<.001
Treats. Months	44	328.75	7.47	0.66	0.950
Error	177	2015.49	11.39		
Total	**239**	**3298.02**			

Stratum standard errors and coefficients of variation

Stratum	d.f.	s.e.	cv%
Reps	3	0.24	0.4
Reps.*Units*	177	3.37	6.0

Analysis of variance of stem weight on various biologicals

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Reps stratum	3	114.56	38.19	3.71	
Treats	4	60.19	15.05	1.46	0.216
Months	11	387.13	35.19	3.42	<.001
Treats. Months	44	597.28	13.57	1.32	0.108
Error	177	1822.47	10.30		
Total	**239**	**2981.63**			

Stratum standard errors and coefficients of variation

Stratum	d.f.	s.e.	cv%
Reps	3	0.80	2.9
Reps.*Units*	177	3.21	11.6

Analysis of variance of head width on various biologicals

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Reps stratum	3	0.18384	0.06128	1.56	
Treats	4	0.07597	0.01899	0.48	0.749
Months	11	14.38106	1.30737	33.19	<.001
Treats. Months	44	1.81548	0.04126	1.05	0.404
Error	177	6.97297	0.03940		
Total	**239**	**23.42932**			

Stratum standard errors and coefficients of variation

Stratum	d.f.	s.e.	cv%
Reps	3	0.03	1.3
Reps.*Units*	177	0.20	8.0

Analysis of variance of head length on various biologicals

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Replicates	3	0.07856	0.02619	0.71	
Treatments	4	0.02326	0.00582	0.16	0.960
Months	11	6.37731	0.57976	15.62	<.001
Treats. Months	44	1.89758	0.04313	1.16	0.246
Error	177	6.56949	0.03712		
Total	**239**	**14.94620**			

Stratum standard errors and coefficients of variation

Stratum	d.f.	s.e.	cv%
Reps	3	0.02	0.5
Reps.*Units*	177	0.19	4.7

Analysis of variance of fresh galls on various biologicals – pots

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Replicates	2	1.444	0.722	0.40	
Treatments	4	8.278	1.656	0.93	0.503
Error	8	17.889	1.789		
Total	**14**	**27.611**			

Stratum standard errors and coefficients of variation

Stratum	d.f.	s.e.	cv%
Replicates	2	0.347	48.0
Replicates.* Treatments	8	1.337	185.2

Analysis of variance of galls diameter on various biologicals at 12 months – pots

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatments	4	1805.2	361.0	2.78	0.033
Replicates	2	30.3	15.1	0.12	0.890
Treatments * Replicates	8	1172.0	117.2	0.90	0.542
Error	14	4418.6	130.0		
Total	**28**	**7476.8**			

https://doi.org/10.33472/AFJBS.3.2.2021.9-15 ISSN: 2663-2187

African Journal of Biological Sciences

Journal homepage: http://www.afjbs.com

Research Paper Open Access

The efficacy of biostimulants in the management of *Agrobacterium tumefasciens* the cause of crown gall disease of roses in Kericho, Kenya

Oniang'o Mary Opisa[1*], Oliver Stafford Achwanya[2], Daniel Otieno Otaye[3] and Japheth Mburugu Muthamia[4]

[1]James Finlay Kenya LTD – Flowers Division, Tarakwet Farm Kericho, Kenya. E-mail: mary.opisa@gmail.com
[2]Department of Biological Sciences, Egerton University, Box 536, Egerton, Kenya. E-mail: achwanyao@gmail.com
[3]Department of Biological Sciences, Egerton University, Box 536, Egerton, Kenya. E-mail: otayedan@yahoo.com
[4]Department of Biological Sciences, Egerton University, Box 536, Egerton, Kenya. E-mail: jmuthamia@gmail.com

Article Info

Volume 3, Issue 2, April 2021
Received : 17 July 2020
Accepted : 02 December 2020
Published : 05 April 2021
doi: 10.33472/AFJBS.3.2.2021.9-15

Abstract

Experiments were conducted at James Finlay Kenya, Tarakwet farm in Kericho county to test the efficacy of various biostimulants, i.e., biozyme 2.5 ml/L, hicure 2.5 ml/L, foltron 2.0 ml/L, codamine radicular 2.0 ml/L, alexin 2.5 ml/L and control (water sprayed only) in controlling *Agrobacterium tumefasciens* the cause of crown gall disease in roses. Treatments were either sprayed or drenched. Both greenhouse and pot trials were conducted on a susceptible rose variety '*Tropical amazone*'. Plots treated with biozyme 2.5 ml/L, hicure 2.5 ml/L, foltron 2.0 ml codamine radicular 2.0 ml/L, alexin 2.5 ml/L were longer, with a bigger head size and had better yield of marketable rose stems compared to control plots treated with water only. It was therefore concluded the application of various biostimulants on roses affected by *A. tumefasciens* by drenching or spraying improved the yield and quality of marketable rose stems. This was attributed to the fact that biostimulants boosted the immune response of roses to *A. tumefasciens* through improving the nutrient use efficiency of the plant and enhanced tolerance to biotic and abiotic stresses. However, more research is needed to elucidate this.

Keywords: *Agrobacterium tumefasciens, Biostimulants, Roses, Stress, Tumor*

1. Introduction

Kenya's horticultural industry is one of the largest in the world and ranked second in foreign exchange earnings after tea. Floriculture is the most developed sector and accounts for about 38% of all horticultural exports in the European Union. With a global market of 7% share, Kenya is the third largest exporter of cut flowers in the world (Kenya Flower Council, 2019). Floriculture provides employment to 100,000 people directly and two million people indirectly. Roses make up 74% of Kenya's flower export followed by carnations (Kenya Flower Council, 2019). Despite the tremendous contribution to the Kenyan economy, profitable production of roses is constrained by a number of plant diseases such as crown gall caused by *Agrobacterium tumefasciens* (Horst, 1983). Rose plants infected by *A. tumefasciens* are stressed and manifest the following symptoms; slower growth, stunting, yellowing, chlorotic leaves and fail to produce healthy flowers (Kado, 2002).

* *Corresponding author: Oniang'o Mary Opisa, James Finlay Kenya LTD – Flowers Division, Tarakwet Farm Kericho, Kenya. E-mail: mary.opisa@gmail.com*

https://doi.org/10.33472/AFJBS.3.2.2021.9-15 ISSN: 2663-2187

African Journal of Biological Sciences

Journal homepage: http://www.afjbs.com

Research Paper Open Access

The efficacy of biostimulants in the management of *Agrobacterium tumefasciens* the cause of crown gall disease of roses in Kericho, Kenya

Oniang'o Mary Opisa[1*], Oliver Stafford Achwanya[2], Daniel Otieno Otaye[3] and Japheth Mburugu Muthamia[4]

[1]James Finlay Kenya LTD – Flowers Division, Tarakwet Farm Kericho, Kenya. E-mail: mary.opisa@gmail.com
[2]Department of Biological Sciences, Egerton University, Box 536, Egerton, Kenya. E-mail: achwanyao@gmail.com
[3]Department of Biological Sciences, Egerton University, Box 536, Egerton, Kenya. E-mail: otayedan@yahoo.com
[4]Department of Biological Sciences, Egerton University, Box 536, Egerton, Kenya. E-mail: jmuthamia@gmail.com

Article Info

Volume 3, Issue 2, April 2021
Received : 17 July 2020
Accepted : 02 December 2020
Published : 05 April 2021
doi: 10.33472/AFJBS.3.2.2021.9-15

Abstract

Experiments were conducted at James Finlay Kenya, Tarakwet farm in Kericho county to test the efficacy of various biostimulants, i.e., biozyme 2.5 ml/L, hicure 2.5 ml/L, foltron 2.0 ml/L, codamine radicular 2.0 ml/L, alexin 2.5 ml/L and control (water sprayed only) in controlling *Agrobacterium tumefasciens* the cause of crown gall disease in roses. Treatments were either sprayed or drenched. Both greenhouse and pot trials were conducted on a susceptible rose variety '*Tropical amazone*'. Plots treated with biozyme 2.5 ml/L, hicure 2.5 ml/L, foltron 2.0 ml codamine radicular 2.0 ml/L, alexin 2.5 ml/L were longer, with a bigger head size and had better yield of marketable rose stems compared to control plots treated with water only. It was therefore concluded the application of various biostimulants on roses affected by *A. tumefasciens* by drenching or spraying improved the yield and quality of marketable rose stems. This was attributed to the fact that biostimulants boosted the immune response of roses to *A. tumefasciens* through improving the nutrient use efficiency of the plant and enhanced tolerance to biotic and abiotic stresses. However, more research is needed to elucidate this.

Keywords: *Agrobacterium tumefasciens, Biostimulants, Roses, Stress, Tumor*

1. Introduction

Kenya's horticultural industry is one of the largest in the world and ranked second in foreign exchange earnings after tea. Floriculture is the most developed sector and accounts for about 38% of all horticultural exports in the European Union. With a global market of 7% share, Kenya is the third largest exporter of cut flowers in the world (Kenya Flower Council, 2019). Floriculture provides employment to 100,000 people directly and two million people indirectly. Roses make up 74% of Kenya's flower export followed by carnations (Kenya Flower Council, 2019). Despite the tremendous contribution to the Kenyan economy, profitable production of roses is constrained by a number of plant diseases such as crown gall caused by *Agrobacterium tumefasciens* (Horst, 1983). Rose plants infected by *A. tumefasciens* are stressed and manifest the following symptoms; slower growth, stunting, yellowing, chlorotic leaves and fail to produce healthy flowers (Kado, 2002).

* *Corresponding author: Oniang'o Mary Opisa, James Finlay Kenya LTD – Flowers Division, Tarakwet Farm Kericho, Kenya. E-mail: mary.opisa@gmail.com*

REPUBLIC OF KENYA

NATIONAL COMMISSION FOR SCIENCE,TECHNOLOGY & INNOVATION

Ref No: **698384**

Date of Issue: **01/September/2025**

RESEARCH LICENSE

This is to Certify that Ms.. MARY oniango OPISA of Egerton University, has been licensed to conduct research as per the provision of the Science, Technology and Innovation Act, 2013 (Rev.2014) in Kericho on the topic: INTERGRATED MANAGEMENT OF Agrobacteium tumefassciens Cavara THE CAUSAL AGENT OF CROWN GALL DISEASE IN ROSES - Rosa hybrida for the period ending : 01/September/2026.

License No: **NACOSTI/P/25/4179011**

698384

Applicant Identification Number

Ag. 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