MANAGEMENT OF WHITEFLIES AND SLUGS ON GREENHOUSE POINSETTIA IN KENYA

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A THESIS SUBMITTED IN PARTIAL FULFILMENT FOR THE
REQUIREMENT OF THE AWARD FOR A DEGREE OF
MASTER OF SCIENCE IN CROP PROTECTION

DEPARTMENT OF PLANT SCIENCE AND CROP PROTECTION
FACULTY OF AGRICULTURE
UNIVERSITY OF NAIROBI

October, 2023

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This thesis is my original work and has not been submitted for award of a degree in any other University.

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Supervisors(s) Declaration

We confirm that this thesis has been submitted with our approval as university supervisors.

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DEDICATION

I dedicate this work to my wife, Florence Achieng, who has been a consistent source of support and encouragement during difficult and easy times. It is also dedicated in the memory of my late mother Consolata Anyango. She taught me never to give up on any great value but always to work hard. She inspired me to further my education beyond a bachelor’s degree level but did not live to see my graduation.

ACKNOWLEDGEMENT

I wish to express my gratitude to my heavenly Father, the Living God, for granting me the grace to extend my studies, advance my knowledge and complete this work. Special appreciation goes to my supervisors; Dr. Dora Kilalo and Prof. Florence Olubayo for the guidance and support offered throughout the period of this work.

My appreciation also goes to my great friends and ex-working colleagues, Eliphus Murithi and Rose Nyakundi for the technical support they offered to me in data analysis.

Finally, I must appreciate the persistent encouragement and moral support I received from my wife, Florence Achieng Odhiambo, throughout the duration of this work.

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LIST OF ABBREVIATION

ANOVA	Analysis of variance
CRD	Clubroot Resistance Gene
DNA	Deoxyribonucleic acid
EU	European Union
FRAC	Fungicide Resistance Action Committee
GDP	Gross Domestic Product
IPM	Integrated Pest Management
IPPC	International Plant Protection Convention
IRAC	Insect Resistance Action Committee
KEPHIS	Kenya Plant Health and Inspectorate Services
KFC	Kenya Flower Council
LSD	Least significant Difference
mtCOI	Mitochondrial cytochrome C oxidase I
NPPO	National Plant Protection Organization
PCPB	Pesticides Control and Products Board
PCR	Polymerase Chain Reaction
SPSS	Statistical Package for the Social Sciences
ToMV	Tomato Mosaic Virus
USA	United States of America

GENERAL ABSTRACT

Flower industry in Kenya, is today the country's top industry after remittance, tourism, and tea. Key problems facing this sector are freight capacity, climate change and plant health regulations. Poinsettia, *Euphobia pulcherima* (Euphorbiaceae), is a flower plant vegetatively grown in Kenya for Europe, USA, Japan markets. Whitefly (Hemiptera: Aleyrodidae) particularly, *Bemicia tabaci* (Hemiptera: Aleyrodidae) is a serious challenge to Poinsettia production. Slugs (gastropoda) are another Poinsettia pest that can cause devastating effects on the crop during establishment if not controlled. Poinsettia and other flower growers depend mainly on insecticides for pest management. But their use is highly regulated especially on crops grown for European, USA and Japan Markets, due to their negative impacts on the environment and human health. Furthermore, pests quickly develop resistance to chemical pesticides. However, pest management with some chemicals is still desirable and practically unavoidable part of integrated pest management.

This study evaluated the efficacy of *Amblyseius swirskii* (Acari: Phytoseiidae) on the management of whiteflies in greenhouse production of Poinsettia. This was compared to integrating the predatory mite with compatible synthetic chemical pesticides and a control where the pesticide was applied alone. The second objective, evaluated the effectiveness of physical barriers (type of growing benches and greenhouse floors) on the management of slugs in greenhouse production of Poinsettia. The study was carried out in Kenya, Muranga County where the growing of Poinsettias plants is done in greenhouses. A Completely Randomized Design (CRD) with two factors was applied. The first factor comprised of three pesticides treatments while the second one consisted of three different Poinsettia varieties. The three pesticides treatments were: Chemical pesticides, *A. swirskii*, and combination of *A. swirskii* with compatible synthetic pesticides (buprofezin and deltamethrin). The predatory mites were applied by broadcasting on the crop while chemical pesticides were sprayed. The experiments were conducted in two seasons in 2017 and 2018.

For the slug experiments, two factors were also applied. The first factor comprised of physical barriers while the second one was the Poinsettia varieties. Treatments under physical barriers included benches with concrete legs on pumice floor, benches with steel legs on pumice floor and benches with steel legs on concrete floors. The results of the study showed that *A. swirskii* in combination with selected compatible chemical insecticides was as effective as the application of pure chemical insecticides in reducing the population of the whitefly.

The type of whitefly infesting Poensettia in the greenhouses was identified as *Bemisia tabaci* biotype while the slug was identified as *Deroceras leave*. Utilization of the predatory mite, *A. swirkii,* on its own to manage whitefly in the Poinsettia stock was not effective and had significantly (p<0.05) higher populations of whitefly compared to the combination of *A. swirskii* and the pesticides, buprofezin and deltamethrin, which in turn was not different from the average population of whiteflies recorded on pesticides alone. The tested varieties had no effect on the population of whitefly but Red with Horizontal Leaves (RHL) had slightly more whitefly populations compared to Red with Vertical Leaves (RVL) and White with horizontal leaves (WHL). Concrete pillars on pumice had significantly (p<0.05) higher populations of slugs compared with Steel pillars on concrete or on pumice. Steel pillars on pumice provided effective vertical physical barrier reducing the mobility of slugs hence the significant reduction of the slug population. The pumice floor provided slugs with conducive environment for laying eggs and hiding places and the pillars/legs that supported benches acted as vertical physical barriers. Poinsettia varieties had an effect on slug population in the greenhouses. The Red variety with horizontal leaves (RHL) was the most vulnerable and significantly (p<0.05) attracted more slugs on the benches compared to the remaining two varieties. Availability of good greenhouse structures such as concrete floor and benches supported on steel stands, may render the effect of variety as unimportant. Therefore, irrespective of variety type, Poinsettia will grow well in this kind of greenhouse conditions without the problems of slugs.

CHAPTER 1: INTRODUCTION

1.1 Background of the Study

Flower industry in Kenya, is today the country's top industry after remittance, tourism, and tea. Key problems facing this sector are freight capacity, climate change and plant health regulations (Vellekoop, 2021). The flower sector contributes more than 70% annually to about 1 billion euros per year (Kenya Flower Council, 2023)

Poinsettia, *Euphobia pulcherima* (Euphorbiaceae) is one of the top demanded bedding plants in USA and Europe (Lutken, Clarke & Muller, 2012). It is an important plant to growers because it complements other annual bedding plants in greenhouse production. This means that during part of the year when other bedding plants are getting uprooted from the greenhouses around March to April, Poinsettia plants are planted to supply both rooted and non-rooted cuttings to the markets from May to August. The other bedding plants and Pelargoniums are planted from August to October to supply cuttings from November to the following year in March. This way, the greenhouse space is optimally utilized by the grower. Poinsettia is also fast growing because of its natural beauty. According to Islam and Joyce (2015), the attractiveness of Poinsettias that boost their demand is their colourful inflorescence (*cyathia*) and bracts (transition leaves). Poinsettia in Africa are grown vegetatively to produce rooted and unrooted cuttings for mainly European market. The rooted or unrooted plants are further developed into finished product for the Christmas sales. Its economic value has been on the increase just like any other flower product in Kenya (Kenya Flower Council, 2017). A look into the flower exports in the five-year period between 2012 and 2022 reveals this as illustrated in Table 1.1. In Kenya about four farms are doing commercial production of Poinsettia with approximately 10,000 employees. This makes Poinsettia production a significant business in Kenya (Kenya Flower Council, 2017).

Table 0.1: Kenya's flower exports between 2012 and 2022

Year	Flowers Export Value (In Kshs Billions)
2012	42.87
2013	46.33
2014	54.60
2015	62.92
2016	70.80
2017	82.25
2018	113.17
2019	104.14
2020	99
2021	101
2022	106

Source: Kenya Flower Council (2023), FPEAK (2021), Mugo (2023)

Poinsettia production in Kenya is done in greenhouses to minimise phytosanitary related risks such as pest and diseases (Kuak, 1995). The greenhouses are cladded with plastics and whitefly or thrips insect nets. Plants are grown from clean starter materials that are free from pests. For import and export of plant material in Kenya, a phytosanitary certificate is issued by Kenya Plant Health and Inspectorate Services (KEPHIS) which is the National Plants Protection Organization (NPPO). This document confirms that the plant materials of interest are admissible in Kenya or country of destination and that they are free of listed pests or have been produced in an environment free of the pests (IPPC, 2014). While the demand of this crop is growing annually, it is a challenge to reliably produce good quality rooted and non-rooted plants every year. Skills in greenhouse climate management is critical especially because of climate change experienced globally. The other challenge is on the management of insect pest and diseases. The key pests of major importance on the crop production are Botrytis, Rhizoctonia, Erwinia, whiteflies, thrips. There are two common whitefly species that may affect Poinsettia plants in Kenya. These are: *Bemisia tabaci* (tobacco whitefly) and *Trialeurodes vaporariorum* (Greenhouse whitefly) (Ronald, 2013). *B. tabaci* is of quarantine importance in most European Union (EU) countries and are listed in the E2 list of quarantine pests in EU (Gwynn, 2014). The two types are also listed in United States of America (USA) and Japan as quarantine pests (Gwynn, 2014). Whiteflies are polyphagous and are known to be vectors of several viruses affecting various other crops such as tomato, Poinsettia, and various species of bedding plants among others. The *B. tabaci* is a major pest of crops of high economic values in the whole world (Nomikou *et al*, 2001). The pest causes damages to the crop directly by feeding it sap. They produce a lot of honey dew on the crops depending on the level of infestation. Too much honeydew can reduce the photosynthetic capacity of a crop further indirectly weakening the plant. The tobacco whitefly is a vector of many viruses that affect plants (Abd-Rabau *et al*, 2010).

They spread viruses such as Tomato Mosaic Virus (ToMV). Most of the viruses transmitted by whiteflies affect food crops as well hence their significance (Philip *et al.*, 2015). Another significant pest of Poinsettias are slugs. Slugs are highly voracious pests that feed on leaves of Poinsettia crops and cause a lot of mechanical damages to the foliar (Schuder *et al.*, 2003). Unlike whiteflies that are known for the transmission of viruses. They cause mechanical damage and may at times feed on the growing tips of the of plants leading to losses of plants or stunted growth. Slugs in Poinsettia greenhouses are favoured by growing environment which includes high temperature, high relative humidity, and moist surfaces especially at early stages of growth (Schuder *et al.*, 2003).

1.2 Statement of the problem

Crop protection from pest damages is a major concern that must be effectively addressed if the production of crop must commercially continue. This is because pest in addition to the physical damage of the plants, may also transmit and spread disease causing pathogens to other crops a situation that complicates the problem of diseases management on the crops (Navas-Castillo *et al.*, 2011). While chemical pesticides have been commonly used in managing most of the pests, there have been concerns about the sustainability of their usage, given that most of them often results in pollution causing harm to humans and the environment (Leonard *et al.*, 2016). Consequently, biocontrol, and physical control methods have continued to be advocated for in pest management. This raises the question, 'To what extent are these methods effective?' This was the gist of this study with an emphasis on management of whitefly and slugs in Poinsettia production in a greenhouse within Kenya.

Management of whitefly in Poinsettia production has been effectively done by the application of chemical pesticides along with the help of physical barrier such as the structure of the greenhouse (Radwan *et al,* 2012). The physical barrier in this case may include greenhouse plastic and nets that keeps insects, beetles, dusts, birds, rodents etc. from accessing the plants. Blowing fans are also used in the greenhouses entrances to screen out flying insect from entering the greenhouse when the doors are opened. Chemical pesticides have been very efficient but their use in the management of pests has raised concerns among the environmentalists and today their use is highly regulated (Leonard *et al.*, 2016). This regulation is done to ensure environmentally sustainable production of flowers. In Kenya for example pesticides to be used in flower production must be registered with Pesticides Control and Products Board (PCPB) and updated in the Kenya Flower Council (KFC) database (Gwynn, 2014).

For this reason, a sustainable means of crop production should be employed to manage these two pests. Integrated Pest Management (IPM) has proved to be the best approach of pests' management in Poinsettia (Gonzalez *et al*., 2016). Use of biocontrol agents is a critical component of IPM and its efficacy has not been fully established for various crops growing under different environment. This project sought to evaluate the effectiveness of using a predatory mite (*A. swirskii*) in the management of whitefly and the effectiveness of physical barriers in managing slugs in Poinsettia plants cultivated to produce vegetative cuttings for export.

1.3 Justification of the study

Poinsettia production in Kenya has been increasing in the recent years. However, its production has not been a smooth process. The warm, wet, and humid propagation environment make Poinsettia cuttings susceptible to several diseases such as Botrytis (*Botrytis cinerea*), bacterial soft rot (*Erwinia carotovora*) and Rhizoctonia stem rot (*Rhizoctonia solanii*), Powdery mildew (*Oidium species*). They are also prone to pests such as fungus gnats (*Bradysia spp*), slugs, shore flies and whiteflies (Lopez, 2014). These pests further complicate the problem of diseases in addition to causing other negative effects on the crops. For instance, whiteflies feed on the leaves' saps thus, they may weaken the crop; facilitate the development of mould; fruits may not ripen uniformly; and they may also transmit disease causing micro-organism (Navas-Castillo *et al.*, 2011). This, therefore, calls for the need to effective management of the pests. Although chemical pesticides have been commonly used in pests' control, there have been great concerns about their short- and long-term effects on the environment. Again, the ability of some pests to build resistance to the available pesticides has been one of the major problems that farmers have to constantly take care of. This is because many agricultural pests especially whitefly, thrips and slugs easily develop resistance to many active ingredients when under constant exposure.

Pesticides are also regulated both locally and internationally based on specific country requirements, customer demands or environmentalist lobby groups. For example, in 1987, *B. tabaci* was intercepted at nurseries in the UK on different crops including Poinsettia and the following year, there was a total of 87 interceptions and outbreaks at growing sites in the country predominately on Poinsettia from Netherlands (Bartlett, 1992). In Europe, there was a continuous interception of the *B. tabaci* on this host plant which was the main reason for most outbreaks between 1998 and 2009 (Cuthbertson *et al*., 2011)

Meeting customer requirements is usually a focus of every grower. Most customers want assurance that they will receive plants free from insect pests and diseases. This plus country specific regulatory requirements makes it very important to get a sustainable whitefly and slug management strategy. This project sought to address these issues by integrating biological control products with other available means of pest management.

1.4 General Objective

The purpose of the study was to develop a sustainable whitefly and slugs management system for greenhouse production of Poinsettia vegetative cuttings in Kenya.

1.4.1 Specific Objectives

The specific objectives to be accomplished included:

1) To assess the efficacy of *Amblyseius swirskii* alone or integrated with compatible insecticide, on the management of whiteflies in greenhouse production of Poinsettia.
2) To determine the effect of physical barriers (type of benches and greenhouse floors) on the management of slugs in a Poinsettia greenhouse.

1.4.2 Hypotheses

This research three hypotheses:

1) The use of *Amblyseius swirskii* or its integration with compatible insecticides reduces whitefly populations infesting Poinsettia in a greenhouse.
2) The type of benches and greenhouse floors do reduce the number of slugs infesting Poinsettia in a greenhouse.

LITERATURE REVIEW

1.5 Description of a Poinsettia plant

Poinsettia (*Euphorbia pulcherrima*) (Euphorbiaceae) is a flower believed to have originated from Central America Mexico (McMahon *et al*., 1996). It belongs to Eurphobiaceae family and mainly used as a flower due to its beautiful different leaf colours between its upper and lower parts. The lower part consists of dark green leaves while those in the upper part are coloured (The Flower Expert, 2017). The coloured leaves can be red, pink, creamy or white among other colours. Commercial production of this plant started in South America and later spread out to North America (Sukma & Megawati, 2016). Today Poinsettia is commercially grown throughout Europe, America, Africa and Asia as a Christmas flower.

1.6 Economic Importance of Poinsettia

Production of Poinsettia contributes greatly to the economies of both developed and developing nations (Dunn *et al*., 2011). According to Islam and Joyce (2015), Poinsettia is categorized as one of the high economically important flowering plants, whose production mostly target Christmas sales demand across various regions in the world including Australia, Europe, Asia, and North America. Its global industrial value is estimated at $154 million (Islam & Joyce, 2015). Annual production of Poinsettia in USA and EU is estimated at a range of 50-100 million plants (Lutken *et al*., 2012). Poinsettia export is prevalent especially among European countries (Islam & Joyce, 2015). In Kenya, Poinsettia production is part of the flower farming (floriculture industry) which is a major segment of the agricultural sector that Kenya’s economy largely relies on. In 2016, earning from the industry was KSh. 70.8 billion. There are more than 200,000 people earning their living from flower industry in

Kenya (Kenya Flower Council, 2017, Spotlight on floriculture in Kenya, 2022). It is apparent that effective production of flowers such as Poinsettia is critical to the national economy.

1.7 Whitefly identification, lifecycle and the damage caused

Whiteflies are tiny insect pests that have a cover of mealy white wax over their wings and bodies, hence their name ‘whiteflies.’ However, unlike the impression by their name, whiteflies are not true flies. They are in the order called *Hemiptera* which relates to mealybugs, scales, and aphids. The average length of an adult whitefly is 1 mm, but females are usually slightly longer than the males (Gangwar & Gangwar, 2018). Whitefly has six development stages: egg, three distinct nymphal stages, pupa and adults. The whole cycle takes 6-55 days depending on the prevailing temperature, the species, and the host plant among other factors (Walker, Perring & Freeman, 2010). The adult whitefly deposits eggs under the foliage. The eggs are not visible but are covered by a white powdery substance that makes them detectable and therefore countable. Development timeline is between 9-12 days depending on the temperature and species. The egg hatches into the first nymphal stage. The first, second and third instar takes 4-6, 7-10, 7-10 days respectively depending on the species and temperature. The fourth nymphal stage is also referred to us as pupal stage. It is not the pupa since it doesn’t molt into a complete non-feeding stage. The pupal stage takes 10-11 days depending on the temp and species. The adult develops from the pupa and is capable of laying eggs 1-8 after emergence and the development period is 5-40 days depending on the species and temperature (Walker *et al*., 2010).

Whiteflies are known to cause major damage to Poinsettia. The greenhouse whitefly (*Trialeurodes vaporariorum*) and silverleaf whitefly (*Bemisia tabaci*) are known to feed on Poinsettia plants. The whitefly has a piercing and sucking mouth thus in most cases the injury on the plant is hard to detect. Nevertheless, if they are in high density, a major injury is possible where crop chlorosis and mottling are the symptoms observed, although rare (Ayalew, 2016). The whiteflies suck plant sap from the leaves thereby causing adverse effects which include stunting of the plants, development of soot-like mould on the honeydew from excretion. The result is non-uniform or non-usable cuttings and transmission of disease-causing micro-organisms (Philip *et al.*, 2015). On top of lowering the quality of the produce, pest's infestation may also lead to substantial use of financial and labour resources used to remove the egg infested leaves of Poinsettia stock plants as a way of breaking the pest life cycle. This eventually increases the cost of production thereby reducing the profitability of Poinnsettia production.

1.8 Slug identification, lifecycle and damage caused

Snails and slugs are gastropods under the phylum of Mollusca (Godan, 1983). Unlike snails, slugs don't have shell or at least their shells are vestigial. They are creatures with soft bodies covered with slimy mucus with no legs, and they have four tentacles on their front – two that host the eyes and two that function like antennae (Douglas & Tooker, 2012). Adult slugs are sufficiently big hence are clearly visible from their place of rest at dusk and dawn (Schuder *et al.*, 2003). All species of slugs are hermaphrodites though their reproduction is complex and species-specific. Each slug in a mating pair lays eggs. They breed anytime there is suitable low temperature and moist conditions. Eggs are laid in moist conditions and normally hatch within 3-6 weeks relative to temperature. They usually take one year to attain sexually productive maturity. Juveniles appear like small adults (GRDC, 2013).

Most slugs' species are crop pests: the grey garden slug (*Deroceras reticulatum*), the banded slug (*Limax marginatusi*), the tawny slug (*Limax flavus*), and the greenhouse slug (*Milax* *gagates*) (Flint, 2003). The damage caused by slugs is mostly eating and causing large, ragged holes on the plant leaves and they can consume an entire young seedling. This makes them troublesome as pests (Clement, 2002). The population of slugs and their activities in an area are determined by the environmental conditions in the area, where they require cool, moist and dark conditions (Douglas & Tooker, 2012). According to Willis *et al* (2008), rainfall may promote high population of slugs because of high soil moisture that encourages their breeding. Their activity is also highest hence cause the most damage in mild and wet conditions. In humid, wet, and cloudy weather, slugs will be persistently damaging the plants causing retarded crop growth, while warm-dry conditions will promote faster plant growth relative to the slug damaging action (Douglas & Tooker, 2012).

Slugs are known to be sensitive to changing light intensity (Vernava *et al*., 2004). Given their nocturnal nature, slugs mostly appear after sunset to feed on the plants on the ground surface, then retreat to their shelters (underneath plant residues, soil layers or rocks, etc.) at sunrise (Douglas & Tooker, 2012). Since the slugs prefer dark areas, and humid conditions, they are common and causing damage in thick crop residue/cover which provides them with good shelter (Douglas & Tooker, 2012).

1.9 Management of Poinsettia pests

1.9.1 General disease management

Most common diseases that affect Poinsettia plants include rust, powdery mildew (caused by different species of fungi in the order *Erysiphales*); and Rhizoctonia (Cruces, 2009). Poinsettias are also vulnerable to diverse foliar and stem diseases. For instance, grey mould (*Botrytis cinerea*), bacterial leaf spot (*Xanthomonas campestris*), Pythium root rot and leaf and bract blight among others. Some of the common disease management methods include use of clean planting materials (culture-indexed cuttings free from bacteria), treating irrigation water with chlorine or bromine, fungicide drenches, removal of infected plants and plant materials, spraying of fungicides and bactericides among others (UMass, 2013).

1.9.2 General insects and other pest management on Poinsettia

Poinsettia is usually affected by several minor pests and a few major ones which includes Whiteflies, *Duponchelia* larvae, *Sciarid* maggots and slugs (molluscs) (Cruces, 2009). Diverse pest management options are applied in controlling pests in Poinsettia production. These include cultural methods, mechanical/physical methods, use of synthetic chemical pesticides and use of biocontrol agents (Jayashankar *et al*., 2013).

Chemical control is the main method of controlling whitefly in Poinsettia and other plants. Long term and frequent application of chemical insecticides can lead to the development of resistance to the chemical by the targeted pest (Radwan *et al.*, 2012). Farmers, environmentalist, and other concerned groups in the world recommend the use of effective pesticides that have short residual effects on the environment, low toxicity to mammals and very selective. Given the efficacy of chemical pesticides on the pest management the strategy should be to mitigate the amount and the effects of the pesticides by developing different active ingredients (Derbah *et al.*, 2012).

Cultural control is the deliberate modification of the environment to make it less fovourable to the survival of the pest. Such activities or initiatives that changes the pest environment includes intercropping, crop rotation, disinfection, insect trapping (CABI, 2015). Appropriate fertigation and irrigation can reduce susceptibility of Poinsettia plant to whitefly. According to Debie, (2003) higher whitefly eggs are found on Poinsettia fertilized with Ammonium nitrate than with calcium nitrate. The removal of bottom leaves from Poinsettia plants improves the coverage of pesticide spray and also helps in removing whitefly eggs with the leaves or any part of the foliage being removed (Debie, 2003). The whitefly host plants such as oxalis, chickweeds, lantanas, velvet leaves, dandelion should be removed from the greenhouses especially under the benches to destroy the breeding ground for whitefly (Debie, 2003). It should, however, be noted that the cultural control strategy alone is not effective since they may take long to implement and others are just not effective (EPPO, 2005).

Sticky traps or roller traps involves the use of mass trapping flying whitefly adults in long yellow sticky traps. This strategy helps in reducing the density of flying insects like whitefly, thrips, sciara fly but is not effective on its own unless supplemented with other control strategies like application of chemical pesticides (Wizgal *et al*., 2014)

1.9.3 Use of biocontrol agents in management of pests in Poinsettia

This is a strategy of using living organisms to suppress the population of pests. It can be achieved through parasitism, predation, other natural mechanism, and it also include the human management role (CABI, 2015). Biocontrol method in pest management is usually done via three strategies: classical biocontrol, augmentation or conservation (Sanda & Sunusi, 2014).

Use of biocontrol agents (primarily augmentative use of predators and parasitoids) in pest management has been integral in greenhouse crops production such as Poinsettia, but the method is yet to be widely embraced in floriculture (van Lenteren *et al*., 2017). Examples of biocontrol agents that can be used in Poinsettia production and floriculture at large include: *Neoseiulus cucumeris, Dalotia coriaria, Eretmocerus eremicus* and *Amblyseius swirskii* among others. Despite producers' efforts, unrooted Poinsettia cuttings often have pests on them especially the whitefly, *Bemisia tabaci*. If they are not regularly monitored and controlled, within the first few production weeks, whitefly population may rapidly grow. Most biocontrol strategies are founded on weekly application of parasitic wasps (*Eretmocerus mudus* and *Encarsia Formosa*). These are generally applied immediately after the Poinsettia cuttings have rooted, but some farmers apply the parasitoids to the cuttings when they are still in the misting system. Whereas predatory mites like *Amblyseius swirskii* and *Delphastus catalinae* are used periodically in the production of other bedding plants (Brownbridge & Buitenhuis, 2017). Use of biocontrol agents as a single method is rarely successful in pests' control. But rather, success is realized by combining them with other methods in an integrated system (Brownbridge & Buitenhuis, 2017).

There are different groups of arthropods that predate on some species of gastropods like rove beetles (*Staphylinidae*), some spiders (*Arachnida*), the larvae of firefly (*Lampyridae*), centipedes (*Chilopoda*), Carabids (*Carabidae*) among others (Nyffeler & Symondson, 2001). Arthropods predators are not effective in managing slugs in the field but the vertebrate predators like rats, birds, lizards are very effective. These animals are, however, not allowed into the greenhouse where Poinsettia plants are grown to produce export cuttings since they may spread plant diseases and or even other pests.

The use of chemicals to manage slugs in field of crop has faced a lot of challenges. Insecticides are not effective against slugs apart from some carbamates that have limited impact on the gastropods but are only registered for ornamental use. The molluscicides such as iron phosphate and metaldehyde are effective. However, they have challenges since they are easily rendered ineffective by rains/misting water and cannot therefore be used in wet conditions (Gall & Tooker, 2017). Wet conditions make the Molluscicide baits less suitable for the management of slugs in a greenhouse production of Poinsettia since during the crop establishment face a lot of misting is done.

1.10 Use of *A. swirskii* as a biological agent for the management of insect pest

Amblyseius swirskii is a predatory mite native to countries in Eastern Mediterranean region like Israel, Cyprus, Italy, Turkey, Greece, and Egypt. It can be found on different crops like apples, apricot, citrus, vegetables and Cotton (EPPO, 2013). It is considered as a type III generalist predator that preys on soft-bodied arthropod pest species, plant exudate and pollen (Croft *et al.*, 2004). This predatory mite belongs to the *Phytoseidae* family, and its main characteristics is long legs with the front pair pointing forward with relatively few hairs. It has been widely used as a biological control agent of mites, thrips and whiteflies in greenhouses and nursery crops (Buitenhuis *et al.*, 2010, Messelink *et al*., 2006). It is adapted to warm and humid sub-tropical climate. The predatory mites develop between 18 ^{0}C to 36 ^{0}C at 60% relative humidity (Lee and Gillespie, 2011). The fact that the predatory mites can survive and reproduce on different types of food such as pollen, plant exudate and nectar, it means that they are able to persist even during the low pest density and improve the effectiveness as a biological control agent (Ragusa and Swirski, 1975). According to Messelink *et al*. (2008) the impact of *A. swirskii* and *Euseius ovalis* (*E. Ovalis*) in controlling Western flower thrips in greenhouse production of cucumber is higher than *Neoseiulus cucumeris* (Oudemans) another phytoseiid also commonly applied in controlling thrips while assessing phytoseiids' effect on thrips. Therefore, *A. swirskii* continues to be acknowledged as potentially suitable for controlling whiteflies and thrips. In Florida, USA, for instance, Philip *et al*. (2015) reported that *A. swirskii* is used in controlling whitefly in non-tomato crops. However, the practice is still uncommon in open seedbed cropping systems.

Notwithstanding, many Poinsettia growers in the developing and even developed countries around the world have continued to experience trouble controlling whitefly (Ronald, 2013). Ronald observed that in the period of 2012, some regions had more challenges than others. Poinsettia grown in Africa like Kenya, are meant to produce unrooted or rooted cuttings to be sold in Europe and other market destinations. These cuttings are harvested from vegetative stock free from any flower bud. This means that the *A. swirskii* mite might not have enough food to subsist on in case of low whitefly or thrips population hence little or no environmental challenges. Ronald (2013) posited that the level of damage in some greenhouses was influenced by the source of the crop materials and the initiation of the pests control method. Nonetheless, scarcity of studies examining the effectiveness of the various pests' management methods used creates a dearth of knowledge or useful insights to understand this issue. Therefore, by examining the effectiveness of using *Amblyseius swirskii* in the management of whitefly in greenhouse Poinsettia production in Kenya, this study will contribute greatly to addressing and understanding the effectiveness of biocontrol.

1.11 Integrating *A. swirskii* with the compatible synthetic chemical pesticides and biopesticide

According to Pilkington *et al.* (2010), control of pests especially arthropods (for instance, whitefly) using biological predators (such as *Amblyseius swirskii*) has been successful in greenhouse crop production for quite long. This is because of the restricted environment in greenhouse production of highly valued crops like Poinsettia is suitable for effectiveness of commercially propagated biological agents. Worldwide as noted by van Lenteren (2012), most of the natural enemies available in the market for arthropod pests are applied in augmentation in greenhouse crops. However, there have been several instances where the use of these agents is to a relatively less extent due to high costs or low efficacy (Gonzalez *et al*., 2016).

It is thus no surprise that integrated pest management (IPM) practices involving biological and biopesticides control strategies are being developed to suppress pests' populations and to consequently slow the spread of pests and their associated diseases (Qureshi & Stansly, 2010). According to Chandler *et al*. (2011), biopesticides comprised of entomopathogenic micro-organisms (microbials such as bacterium, virus or fungi) are usually encouraged as an option in cases where arthropods (e.g. whitefly) biological control agents are not accessible or are inadequate for effectiveness.

Entomopathogenic baculoviruses, bacteria and fungi are often considered as examples of biopesticides alongside natural compounds and minerals. Most of commercially available microbial products are based on the species *Bacillus thuringiensis* (*B. thuringiensis* or Bt) (Vachon *et al*., 2012). *Bacillus thuringiensis* forms spores containing crystals that mostly comprise of delta-endotoxins (Cry 1 and Cry 2 proteins) which cause lysis of the target pest's gut cells (Sanahuja *et al*., 2011). Vachon *et al.* (2012) asserts that bacterial insecticides must be ingested for them to effectively control pests. After their consumption, they paralyze the target insect's gut through the toxins released by the bacteria, which then inhibits pest feeding, and eventually kills the pest.

The entomopathogenic fungi are a collection of various types whose common feature is infecting the insects and causing diseases in them. According to Humber (2012) they are mostly in two categories. That is, Hypocreales in the Entomophthoromycota and Ascomycota phylums. They infect the host through direct breach of the cuticle to get into haemocoel of the insect. This ability of invading without necessarily being consumed puts them at an advantage in fighting pests that feed on the plant's phloem like the whitefly and aphid that do not consume microorganism on the surface of the leaves (Gonzalez *et al*., 2016).

Upon spraying Entomopathogenic fungi in suspension form, effectiveness has been reported in controlling pests (such as aphids, weevils, thrips and whiteflies) in greenhouse crop production (Khan *et al*., 2012). In this regard, some of those effective include: "*Beauveria bassiana* sensu lato, *Isaria fumosorosea* (formerly *Paecilomyces fumosoroseus*), *Metarhizium anisopliae* sensu lato and *Lecanicillium* (formerly *Verticillium*)" (Bischoff *et al*., 2009). Various strains of *Beauveria bassiana* are already in use in the management of whitefly in Poinsettia greenhouses. According to Pell *et al.* (2010) those in the order Entomophthorales causes epizootics which decreases the pest population very fast. Corry and Myers (2003) argue that most commercially produced microbial products contain Baculoviridae virus alone despite that there are many viruses that infect arthropod pests. This virus is "host-specific" which means it infects a single or few related species of pests. Baculoviruses are suitable in bio-control methods since applying them will have no direct effect on other insects that are not targeted (Gonzalez *et al*., 2016).

Using entomopathogenic micro-organisms and endophytes in production of crops in greenhouses may directly and/or indirectly affect bio-control systems depending on the arthropod natural enemy, possibly resulting into good and bad outcome in the pest control (Gonzalez *et al*., 2016). Effective bio-control agents for some arthropod pests are not available or those that are available are too expensive and hence hardly used (Messelink *et al*., 2014). For others, they may be available, but their effectiveness may be limited to certain crops only. Messelink *et al.* (2014) gives an example whereby, Phytoseiid predatory mites effectively controls western flower thrips in production of sweet pepper that provides pollen and nectar, but ineffective in ornamental crops which do not have these. This can also be a problem of *A. swirskii* in the management of whitefly population in Poinsettia plant that are grown to produce vegetative cuttings. The plant grown for production of vegetative cuttings doesn’t produce pollen and nectar for the predatory mites, and this may affect its efficacy in pest management. Biopesticides are a suitable complement in bio-control programs i, provided that all the possible adverse effects of the microorganisms are considered (Gonzalez *et al.*, 2016).

According to Roy *et al* (2006), using biopesticides in crop production may have unanticipated effects on the arthropod natural enemy by altering the pest’s behaviour, or changing the pests’ population and diversity. The change in population and diversity of the pests is crucial for general predators which consume a variety of pests. Messelink *et al* (2008) indicates that variation in a single pest species because of using host-specific microbial can have an indirect effect on other pests, since food is available, or the predators do well on a mixture of pests (diets). Such effects must be considered in the application of microbials against a particular pest.

Avoidance may also lead to other adverse interactions. According to Labbe’ *et al.* (2009) there are instances where pests avoid predating crops on which microbials have been applied, hence increasing the “searching time” for the prey and minimizing the rate of predating. Mesquita and Lacey (2001) affirm that several parasitoid species are also capable of detecting entomopathogenic fungi infection on their host. Baverstock *et al.* (2005) indicates that while this may be good for the parasitoids’ young ones as it supports their survival, it may nonetheless, simultaneously diminish efficiency by prolonging the searching time. Other parasitoids are not capable of detecting the infection on their hosts which can adversely affect their efficiency, since the developing parasitoids’ offspring may not reach adulthood. Roy *et al.* (2008) posits that biocontrol agents could also consume the infected host thus diminishing the microbial’s efficiency. Cumulatively, various composite interactions as discussed can result into non-additive effect of integrated treatments. Therefore, integrating the use of

Amblyseius swirskii and biopesticides needs an in-depth investigation for a solid conclusion to be drawn.

1.12 Use of physical barriers in the management of slugs

Using physical barriers to intercept the mobility of slugs has been suggested including a band of soil around the plant; a fence of corrugated tin or wire mesh; ditches dug round the seed bench and collecting the slugs every day (Raut & Barker, 2002). Other barriers suggested are barriers made of copper (such as copper foil wrapped round the planting box or trunks to intercept slugs) which are considered effective on the basis that the reaction between copper and slime secreted by the slug/snail causes electricity to flow (Flint, 2003).

Heaping a barrier made of dry ash with a height of 1 inch and a width of 3 inches round the seedbed is further suggested (Flint, 2003). However, such barriers may become ineffective if they become damp hence making their maintenance hard rendering them irrelevant in the seedbed in most cases. While it is apparent that several physical barriers can be used to manage slugs, there is scanty information regarding the efficacy in managing slugs. Few studies have recommended the need for evaluating their effectiveness. Most of the studies focused on examining how these barriers are applied in managing the pest. Few studies have investigated the effectiveness of different types of physical barriers in controlling slugs giving mixed results. Capinera (2018) affirmed that differences exist in effectiveness of different physical barriers in the control of slugs. This study, therefore examined the effectiveness of the physical barriers in greenhouse Poinsettia production in Kakuzi Kenya. In this case, the study sought to test whether benches with steel stands and or concrete floors reduces the problem of Poinsettia infestation by slugs. The objective of this study was to investigate the impact of two different greenhouse structures on the control of slugs. The first structure consisted of benches with concrete stands in a greenhouse with pumice floor. The concrete stands are made of a block measuring 9x9x12 inches. The problem with this kind of stand is that they block both air flow and light penetration between benches. During irrigation, the concrete stands absorb fertilizer solutions. The resultant effect is the provision of dark breeding ground under the benches that are humid and poorly aerated. Benches with slender steel legs are more aerated, better lit, and less humid. Concrete floors dry up faster after irrigation or application of fertilizer as opposed to floor with pumice that absorbs water and drained fertilizer solution. The steel support pillar provides a physical barrier for the slugs to climb the benches and destroy the crop. Slugs may not move as fast on concrete floor because it is dry and may be warmer than the pumice floor. Furthermore, the slugs can easily get desiccated on the concrete floor due to less moisture. The findings of this study will contribute additional information on the effect of barriers in managing the pest.

CHAPTER 3:

EVALUATION OF *AMBLYSEIUS SWIRSKII* ON THE MANAGEMENT OF WHITEFLY IN GREENHOUSE PRODUCTION OF POINSETTIA IN MURANGA COUNT

Abstract

Whitefly (Hemiptera:Aleyrodidae) is a pest of great economic importance in Poinsettia production especially in Africa where the plant is grown vegetatively to produce cuttings for markets in Europe, USA, and Asia. Whiteflies, especially *Bemisia tabaci* have been listed as quarantine pests in the markets. The plant materials for export must be free from whitefly. The objective of this study was to compare the ability of *Amblyseius swirskii* or its combination with compatible chemical insecticides with synthetic chemical insecticide in controlling whitefly on Poinsettia stock plants grown in greenhouse condition to produce vegetative cuttings in Kenya. The experiment was conducted in 9 greenhouses that were covered with plastic and insect net (optinet 50). The experiment was conducted in two seasons, 2017 and 2018 in Kakuzi, Muranga County in Kenya. A Completely Randomized Design (CRD) with three replicates and two factors was used.

Pesticide use was the first factor which comprised of three treatments: Chemical pesticides, *A. swirskii*, and *A. swirskii* with buprofezin and deltamethrin as compatible insecticides. The second was the variety factor which consisted of three different Poinsettia varieties. The three varieties were planted in each greenhouse and there were three greenhouses per each pesticide treatment and variety. Chemical pesticides were applied by foliar spraying on a weekly basis and depending on pest pressure. The predatory mites, *A. swirskii* were applied by broadcasting them on the crop followed by several augmentations. Deoxyribonucleic acid (DNA) of whitefly sample was taken to the laboratory for identification in 2018. The quantity of whitefly adults and eggs were monitored and counted weekly. The predatory mites, *A.*

*swirsk*i were bought from Koppert Kenya. The results revealed that the mean average of adult whiteflies or eggs in greenhouses treated with chemical pesticides and *A. swirskii* with buprofezin and deltamethrin combination were comparable. Utilization of *A. swirskii* alone had significantly ($p<0.05$) higher whitefly populations than any of the remaining two treatments. The predatory mite, *A. swirskii* in combination with the compatible chemical insecticides has potential as a substitute of chemical pesticides for controlling whitefly in greenhouse Poinsettia production. Varieties did not vary in attracting and in controlling whitefly. The whitefly sample was classified as *Bemisia tabaci* biotype Q based on the sequence of the mitochondrial cytochrome C oxidase I (mtCOI) and biotype-specific polymerase chain reaction (PCR) used for anaalysis.

3.1 Introduction

The adoption of biocontrol agents as pests control method is largely being encouraged worldwide as part of modern IPM systems especially due to the growing concern on health and environmental hazards linked with chemical pesticides (Chidawanyika *et al*., 2012). Some of biocontrol agents being used in floriculture include: *Neoseiulus cucumeris, Eretmocerus eremicus* and *Amblyseius swirskii* among others (Brownbridge & Buitenhuis, 2017). Whiteflies are among the prevalent greenhouse and field crop pests globally (Ayalew, 2016). Their prevalence has been highly reported in various flower farms/Nurseries and they are known to negatively affect different flowers and herbs (Ahmed *et al*., 2009). Usually, different insecticides are used in a rotational manner to control the pests as a buffer against pesticide resistance. Nevertheless, with the high demand for production of pest-free farm products, pesticides must be frequently applied, and this has aggravated pesticide resistance, consequently undermining the efficacy of certain chemical pesticides (Gorman *et al*., 2002).

This coupled with other problems, necessitates the promotion of other pest control methods like biocontrol agents and Integrated Pest Management (IPM) program.

The success of these methods in controlling whiteflies in different crops growing under different environment need to be well understood before adopting them especially in commercial crop production.

3.2 Materials and Methods

3.2.1 Study site

The study was carried out in Kakuzi Mitubiri ward in Murang’a County. It is among the counties in central Kenya lying between latitudes 0^{o}34’ South and 107’ South and longitudes 360^{o} East and 37^{o}27’ East. Its altitude ranges from 914m to 3,533m above sea level along the slopes of the Aberdare Ranges.

Kakuzi area (Makuyu) is characterized by arid and semi-arid conditions hence the reason why crop farming is better done in greenhouses. The location falls in lower midland three (LM3) but the study itself was carried out in a controlled greenhouse environment. The study was done in two years. The first one was in 2017 and repeated in 2018. For each year, nine greenhouses were used. One greenhouse is about half a hectare covered with appropriate greenhouse plastic sheets and whitefly nets on the sides and roof vents. Each greenhouse had 75 benches. Each bench is 1m wide, 40m long, 0.7m apart and 0.75m above the ground. One bench supported 640 4-litre pots filled with pure pumice uniformly arranged in four rows along the bench at a pot density of 16 pots/m^{2}. Irrigation and fertilizer provision were done by use of drip pipes. The internal climate was controlled to provide the right growing conditions for Poinsettia stock plants. The temperature of the greenhouse was maintained at approximately between 20 and 31 ^{0}C for better yield production.

3.2.1 Crop establishment and experimental design

3.2.1.1Crop establishment

Growing of Poinsettias stock was done in greenhouses. Tip cuttings were rooted in a special facility designed for optimal rooting within the farm. Planting materials for producing young plants were assessed and only good quality materials free from visible pests were rooted. In the rooting greenhouse, chemical pesticides were used to keep off insects and diseases from negatively affecting the rooting young plants.

Transplanting was done in separate greenhouses from where the rooting had taken place. Prior to the transplanting, the greenhouses were cleaned and disinfected. They had been covered with plastic and insect net to optimize growing environment for Poinsettia. The greenhouse was meant to protect the crop from adverse weather elements and pests. The insect net and plastic were assessed and maintained to ensure no holes or gaps that could allow insects like whitefly in the greenhouses. Fans and lights were checked and maintained to ensure greenhouse temperature and humidity were well managed during the production of the plants.

The rooted young plants were transplanted into the pots with steam sterilized pumice in February 2017 and 2018. Poinsettia is an obligate short-day plant and therefor flowers when the day length is shorter than 11 hours per day (Johnson, 2020). Artificial lighting was provided every night at 100 lux for 7 hours to keep the plants vegetative. Fertigation and irrigation were done hydroponically at a volume of 200 -350ml of fertilizer solution (containing 150 to 200 ppm nitrogen and other macronutrients, plus micronutrients) per pot per day for 6 days in a week. Electric conductivity of the solution varied from 2-4 mS/cm depending on the stock age (Lopez, 2014). The main shoot pinching was done two weeks after planting. The emerging side shoots were pinched two weeks later and repeated two times after an interval of two weeks. To encourage faster growth of the shoots, misting was done once every day for six weeks. Four weeks after planting the big dark green leaves were removed once a week to open the canopy for better aeration. The bottom leaves were removed once the plant had grown bigger, about 8-9 weeks after planting. Harvesting started 12 weeks from the time of planting and continued for 11 weeks. Quality cuttings were harvested for sale to customers. The harvesting was done uniformly across all treatments in each of the greenhouses for the varieties depending on the cutting's availability. The number of cuttings per square meter for each treatment and variety was recorded.

3.2.1.2 Experimental design and treatments application

The study used a Completely Randomized Design (CRD) of two factors. The main factor consisted of three pesticide treatments which were: The Chemicals pesticides [CH], *A. swirskii* [AS] and the combination of both *A. swirskii* and chemical pesticides [AS +CH]. In this case, application of synthetic chemical pesticides was taken as the control treatment since the use of chemicals had been the conventional whitefly management method. It was not possible to have a control of greenhouses treated without application of any insecticide to control insects. All the plants were produced for export market and no quarantine pest was allowed. The second factor consisted of three Poinsettia varieties: Red Variety with Horizontal leaves [RHL], Red Variety with Vertical leaves [RVL] and White Variety with Horizontal Leaves [WHL]. The treatments were applied differently in three different sets of green houses, with each set having 3 greenhouses. See (Figure 3.1) for more information. The first set of the 3 greenhouses was treated with *A. swirskii*. The second set were treated with synthetic chemical pesticides only, while the third set was treated with a combination of *A.*

swirskii and selected compatible synthetic chemical pesticides. Each greenhouse had at least three of the varieties to be tested.

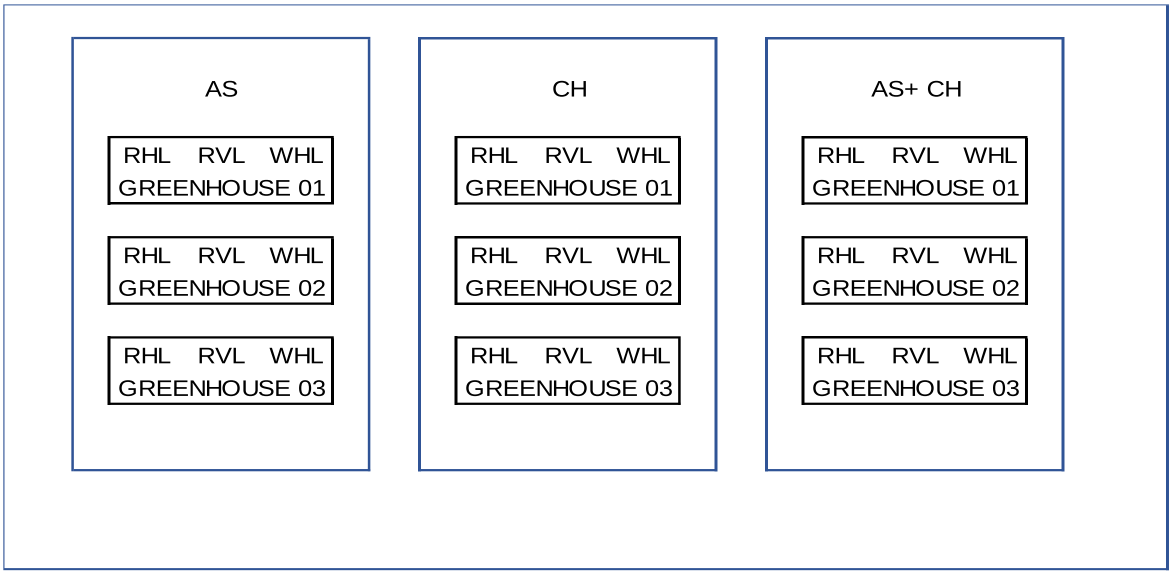


Figure 3.1: Completely Randomised Design (CRD) for pesticides and varieties.

Legend :Experiment consists of two factors of treatments. Total of 9 greenhouses used: 3 Pesticides treatments, 3 varieties treatments and 3 replications. AS, CH and AS + CH are different pesticides treatments for *A. swirskii*, Chemical pesticides, and a combination of *A. swirkii* and compatible chemical pesticides respectively. RHL, RVL and WHL are red variety with horizontal leaves, red variety with vertical leaves and white variety with horizontal leaves.

3.2.1.3 Application of *A. swirskii* on Poinsettia

This was the treatment done in the three greenhouses that received the *A. swirskii* mites as the sole insecticides against whitefly throughout the two seasons. The predatory mites were sourced from Koppert Biological Systems (K) Ltd. They were supplied in 500ml bottle container. Each bottle contained 50,000 predatory mites mixed in a bran. They were also supplied in small sachet for slow-release process.

The application of the *A. swirskii* did not start immediately after planting because of the hand watering that went on for 4 weeks. Watering or heavy misting on the foliage of the plant could wash out the predatory mites off the foliar thereby reducing their efficiency. The predatory mites were released by broadcasting approximately one third of the bottle uniformly on the plants across and long the benches. This translated into about 312 predatory mites per square meter as recommended by Koppert. The process went on for four weeks. Thereafter 24 sachets of the mites were uniformly allocated on each bench. Each sachet contained 250 *A. swirskii* mites and were designed for slow release of the predators into the plants as recommended by Koppert and in line with the findings of Buitenhuis *et al*. (2010) and Opit *et al.* (2005).

Population build-up of the biocontrol mites was monitored by scouting three days after application. This information guided the decision on when to apply another round of the mites. One monitoring station (of $1m^2$ or 16 plants) was used per 3 benches. The frequency of application of the *A. swirskii* and other pesticides is summarized in (Table 3.1). Dynomite (Pyridaben) was spot sprayed in weeks 25 and 27 on the periphery of benches in all greenhouses to manage red spider mites. Other pesticides were sprayed in the greenhouse to control the Poinsettia diseases and other insect pests like caterpillars, moths. The pesticide solution applied per hectare was 1000 litres. For detailed information of the applied pesticide in 2017 (Table 3.1). The list of compatible pesticides to *A. swirskii* mites was given by Koppert Kenya.

Table 3.1: Pesticides applied in greenhouses treated with *A. swirskii* in 2017 and 2018.

Trade name	**Active ingredient**	**Application rates**	**Category**	**Pest / Disease**	**IRAC/ FRAC**	**Week applied**
Swirski-Mite (slow release)	*Amblyseius swirskii*	100 bottles/Ha	Insecticide	Whiteflies, Thrips		25,
Swirski-Mite	*Amblyseius swirskii*	50 bottles/Ha	Insecticide	Whiteflies, Thrips		12, 15, 19, 24, 28,
Applaud 40 SC	Buprofezin 400g/L	50 ml/100 L	Insecticide	Whiteflies (eggs)	16	7, 11,
Avaunt 150 EC	Indoxacarb 150g/L	40 ml/100 L	Insecticide	Caterpillars	22A	8, 20,
Decis 2.5 EC	Deltamethrin 25g/L	50 ml/100 L	Insecticide	Whiteflies, Thrips, Moths	3	7,
Dipel DF	*Bacillus thuringiensis* subsp. Kurstaki strain ABTS-351 54%w/w	80g/100 L	Insecticide	Caterpillars, Duponchelia moth	11B$_2$	9, 14,
Dynamec 1.8 EC	Abamectin 18g/L	100 ml/100 L	insecticide	Red Spider mites, Thrips, Whiteflies	6	16,
Dynomite 150 EC	Pyridaben 150g/L	50 ml/100 L	insecticide	Whiteflies, Red spider mites, Mealy bugs	21A & 7C	8,9,10
Luna Tranquility SC 500	Fluopyram 125g/L + Pyrimethanil 375g/L	25ml/100 L	Fungicide	Powdery mildew	7&9	7,
Nimbecidine EC	Azadirachtin 0.03%	250 ml/100 L	Insecticide	Whiteflies, Thrips		11,
Ortiva Top 325 SC	Azoxystrobin 250g/L + Difenoconazole 125g/L	75 ml/100 L	Fungicide	Botrytis, Rust, Downy mildew, Powdery mildew	11/3	10, 14, 19,
Teppeki 50 WG	Flonicamid	14g/100 L	Insecticide	Whiteflies, thrips	29	9,
Thiovit Jet WP	Sulphur 80% w/w	250 gr/100 L	Fungicide	Powdery mildew	M2	14,
Tomahawk 250EC	myclobutanil 200g/L	30ml/100 L	Fungicide	Powdery mildew, rust, Ealy Blight	3	10, 18, 20, 24,
Scala 40 SC	Pyrimethanil 400 g/L	100 ml/100 L	Fungicide	Botrytis, Rhizoctonia	9	12, 15, 17,
Teldor WG 50	Fenhexamid 500g/kg	100 ml/100 L	Fungicide	Botrytis, Rhizoctonia	17	9, 25,
Pencozeb	Mancozeb 750g/kg	150 g/100 L	Fungicide	Downy mildew, Rust		8, 13, 21, 23, 27,
Ridomil Gold MZ 68 WG	Metalaxyl-M 40g/kg + Mancozeb 640g/kg	250 gr/100 L	Fungicide	Downy mildew, Botrytis, Phytophthora	8/M3	22, 26, 28,

3.2.1.4 Application of chemical pesticides

On the control plots / greenhouses, chemical control of whitefly was applied. A spray program was developed based on the available active ingredients in the market that had been registered and approved for use in flower industry by EU and GLOBAL gap. Table 3.2 shows the details of all chemicals used in the greenhouses under the chemical treatments at different time. To manage pest resistance, chemical of the same mode of action were applied for utmost three weeks followed by a chemical of different mode of action. As already mentioned in section 3.2.2.1, having a treatment with no insecticide application against whitefly, was not possible since all the plants were to produce cuttings already ordered for export.

3.2.1.5 Application of combined *A. swirskii* and the compatible synthetic pesticides for whitefly control.

In this treatment, application of *A. swirskii* began 4 weeks after planting and went on throughout the season. Compatible insecticides were applied whenever the average population of whitefly was found to be higher than 0.5 adults per yellow sticky trap in a week. During the first four weeks, synthetic chemical pesticides were used to manage the whitefly and thrips. The chemicals insecticides sprayed included: Applaud (buprofezin), Decis (deltamethrin) and Dynomite (pyridaben). *A swirkii* were applied. Refer to (Table 3.3) for detailed information of other chemicals sprayed in the treatment for the two seasons.

Table 3.2: Pesticides applied in greenhouses treated with chemical insecticides in 2017 and 2018

Trade name	Active ingredient	Application rates	Category	Pest / Disease	IRAC/FRAC	Spraying Week
Karate Zeon 5CS	lambda-cyhalothrin 50g/L	60 ml/100 L	Insecticide	Whiteflies, Thrips	3	14, 15, 16,
Applaud 40 SC	Buprofezin 400g/L	50 ml/100 L	Insecticide	Whiteflies (eggs)	16	7, 11, 16, 21, 24,
Avaunt 150 EC	Indoxacarb 150g/L	40 ml/100 L	Insecticide	Caterpillars	22A	8, 20,
Decis 2.5 EC	Deltamethrin 25g/L	50 ml/100 L	Insecticide	Whiteflies, Thrips, Moths	3	7, 11, 12, 13, 21, 22, 23,
Dipel DF	*Bacillus thuringiensis* subsp. Kurstaki strain ABTS-351 54%w/w	80g/100 L	Insecticide	Caterpillars, Duponchelia moth	11B2	9, 13,
Dynamec 1.8 EC	Abamectin 18g/L	100 ml/100 L	insecticide	Red Spider mites, Thrips, Whiteflies	6	16,
Dynomite 150 EC	Pyridaben 150g/L	50 ml/100 L	insecticide	Whiteflies, Red spider mites, Mealy bugs	21A & 7C	15, 25,
Luna Tranquility SC 500	Fluopyram 125g/L + Pyrimethanil 375g/L	25ml/100 L	Fungicide	Powdery mildew	7&9	7,
Nimbecidine EC	Azadirachtin 0.03%	250 ml/100 L	Insecticide	Whiteflies, Thrips		12,
Ortiva Top 325 SC	Azoxystrobin 250g/L + Difenoconazole 125g/L	75 ml/100 L	Fungicide	Botrytis, Rust, Downy mildew, Powdery mildew	11/3	10, 14, 19,
Teppeki 50 WG	Flonicamid	14g/100 L	Insecticide	Whiteflies, thrips	29	17, 18, 19, 29,
Thiovit Jet WP	Sulphur 80% w/w	250 gr/100 L	Fungicide	Powdery mildew	M2	14,
Tomahawk 250EC	myclobutanil 200g/L	30/100 L	Fungicide	Powdery mildew, rust, Ealy Blight	3	10, 18, 20, 24,
Scala 40 SC	Pyrimethanil 400 g/L	100 ml/100 L	Fungicide	Botrytis, Rhizoctonia	9	15, 17,
Teldor WG 50	Fenhexamid 500g/kg	100 ml/100 L	Fungicide	Botrytis, Rhizoctonia	17	9, 11, 25,
Pencozeb	Mancozeb 750g/kg	150 g/100 L	Fungicide	Downy mildew, Rust	M3	8, 13, 21, 23, 27,
Ridomil Gold MZ 68 WG	Metalaxyl-M 40g/kg + Mancozeb 640g/kg	250 gr/100 L	Fungicide	Downy mildew, Botrytis, Phytophthora	8/M3	22, 26, 28,

Table 3.3: Pesticides applied in greenhouses treated with *A. swirskii* and some compatible synthetic chemical insecticides in 2017 and 2018.

Trade name	**Active ingredient**	**Application rates**	**Category**	**Pest / Disease**	**IRAC group**	**Application week**
Swirski-Mite (slow release)	*Amblyseius swirskii*		Insecticide	Whiteflies, Thrips		25,
Swirski-Mite	*Amblyseius swirskii*	50 bottles/Ha	Insecticide	Whiteflies, Thrips		12, 15, 19,
Applaud 40 SC	Buprofezin 400g/L	50 ml/100 L	Insecticide	Whiteflies (eggs)	16	7, 11, 21, 25
Avaunt 150 EC	Indoxacarb 150g/L	40 ml/100 L	Insecticide	Caterpillars	22A	8, 20,
Decis 2.5 EC	Deltamethrin 25g/L	50 ml/100 L	Insecticide	Whiteflies, Thrips, Moths	3	7, 21, 25
Dipel DF	*Bacillus thuringiensis* subsp. Kurstaki strain ABTS-351 54%w/w	80g/100 L	Insecticide	Caterpillars, Duponchelia moth	11B$_2$	9, 13,
Dynomite 150 EC	Pyridaben 150g/L	50 ml/100 L	insecticide	Whiteflies, Red spider mites, Mealy bugs	21A & 7C	8, 9, 10,
Dynamec 1.8 EC	Abamectin 18g/L	100 ml/100 L	insecticide	Red Spider mites, Thrips, Whiteflies	6	16, 23,
Luna Tranquility SC 500	Fluopyram 125g/L + Pyrimethanil 375g/L	25ml/100 L	Fungicide	Powdery mildew	7&9	7,
Nimbecidine EC	Azadirachtin 0.03%	250 ml/100 L	Insecticide	Whiteflies, Thrips		12, 24,
Ortiva Top 325 SC	Azoxystrobin 250g/L + Difenoconazole 125g/L	75 ml/100 L	Fungicide	Botrytis, Rust, Downy mildew, Powdery mildew	11/3	10, 14, 19,
Teppeki 50 WG	Flonicamid	14g/100 L	Insecticide	Whiteflies, thrips	29	19,
Thiovit Jet WP	Sulphur 80% w/w	250 gr/100 L	Fungicide	Powdery mildew	M2	14,
Tomahawk 250EC	myclobutanil 200g/L	3oml/100 L	Fungicide	Powdery mildew, rust, Ealy Blight	3	10, 18, 20, 24,
Scala 40 SC	Pyrimethanil 400 g/L	100 ml/100 L	Fungicide	Botrytis, Rhizoctonia	9	15, 17,
Teldor WG 50	Fenhexamid 500g/kg	100 ml/100 L	Fungicide	Botrytis, Rhizoctonia	17	9, 11, 25,
Pencozeb	Mancozeb 750g/kg	150 g/100 L	Fungicide	Downy mildew, Rust	M3	8, 13, 21, 23, 27,
Ridomil Gold MZ 68 WG	Metalaxyl-M 40g/kg + Mancozeb 640g/kg	250 gr/100 L	Fungicide	Downy mildew, Botrytis, Phytophthora	8/M3	22, 26, 28,

3.2.3 Data collection and Analysis

3.2.3.1 Data collection

Four types of data were collected from each greenhouse, and variety. The data included the population of whitefly adults, whitefly eggs, yield in terms of number of usable cuttings per meter square, and climate data [temperature & humidity].

Whitefly adults and eggs populations were monitored by counting them in the six benches every Tuesday and Thursday. Six yellow traps from Koppert Kenya were used per bench, hanged about 30 cm from the top of the plants. All adult whiteflies found on the traps and plant foliar were counted and recorded. Every leaf in each plant was checked for the presence of the eggs. The population build-up of the biocontrol agents (*Ambylysius swirskii*) was monitored by counting them in every plant within one meter square three days after they were applied. 8 out of 16 plants should have at least one leaving predatory mite, otherwise another release of the biocontrol agent would be required. Crop performance yield data was collected immediately the harvesting started 12 weeks after planting. Only good export quality cuttings were harvested. An export quality cutting must have a stem of about 1 to 1.5 cm, a growing point and at least 3 leaves. It should be mature enough to withstand long distance shipment stress for over 4 days. All harvested cuttings were exported to the customers. Quantity of cuttings harvested was recorded for each treatment. Other information monitored was climate [Temperature and Relative Humidity]. Data loggers from Watchdogs company were used for monitoring and recording temperature, and relative humidity in the farm.

3.2.3.2 Data collection and analysis

The whitefly population and the cutting yield data were subjected to analysis of variance (ANOVA) to evaluate treatment effects. The Fischer’s least significant difference (LSD) test was applied to decide whether there were significant differences between means of different treatments. Data analysis was done for the purposes of hypotheses testing and it was done with the help of Microsoft Excel worksheet and the Statistical Package for Social Sciences (SPSS) version 29.0.1.0. The data collected was first entered in a Microsoft Excel worksheet to aid in data cleaning. This involved the removal of typo mistakes and outliers. The cleaned data was entered into SPSS for data analysis using Completely Randomised Design approach. In this regard, the differences in the means of whitefly adults or eggs populations were statistically compared for each treatment.

3.2.3.3 Sampling whitefly for identification

Samples of adult whiteflies were collected from the infested plants in July 2023, stored in 95% alcohol, and finally deoxyribonucleic acid (DNA) was isolated and sent for identification in Macrogen laboratory in Netherlands. The first step was to identify the species of the whitefly, followed by the establishment of the biotype and finally, do some sequencing of the DNA extract. The results of sequencing could help in tracing the origin of the whitefly. The origin was important because the starter material for the Poinsettia stock had been sourced from Europe.

3.2.4. DNA extraction

DNA was extracted from individual whitefly adults sample collected from the greenhouses by adding 20 µl Quanta Extracta extraction reagent (Quanta Biosciences) and incubating at 95°C for 30 min.

3.2.4.1PCRs and gel electrophoresis

Two polymerase chain reaction (PCR) processes were carried out, one with universal primer sets against mitochondrial cytochrome oxidize 1 (mtCOI) to identify the species and the other with biotype specific three sets of primers to identify the whitefly group or biotype. The region of mtCOI was targeted because it is one of the DNA parts commonly used for determining the genetic structure of *B. tabaci* (De Barro *et al*., 2011). The primers in the first process were ZUM3432 and ZUM3433 (Table 0.14). Polymerase chain reaction products were separated by gel electrophoresis using 0.8% w/v agarose. After completion, the gel was stained with SERVA DNA stain G and exposed to UV for analysis.

Table 3.4: Universal PCR composition - amplification of the mtCOI fragment.

Component	**Volume (µl)**
Toughmix (2x)	12.5
ZUM3432 (20 µM)	0.5
ZUM3433 (20µM)	0.5
Water	6.5
Template (DNA extract)	5

The selection of the biotype specific primer combinations for the second PCR process depended on the results of the first process. The biotype specific primers combinations used were meant to establish the biotype of *B. tabaci*. Biotype “B”, “Q” and “New word”, were targeted. The primer combinations for “B”, “Q” and “New world” were ZUP3426 & ZUP3430, ZUP3428 & ZUP3431 and ZUP3427 & ZUP3429 respectively. The PCR process was run for each combination (Table 3.5). Polymerase chain reaction products were separated by gel electrophoresis using 0.8% w/v agarose. After completion, the gel was stained with SERVA DNA stain G and exposed to UV for analysis.

Table 3.5: Biotype-specific PCR composition for each primer combination.

Component	**Volume (µl)**
Toughmix (2x)	12.5
Forward primer (20 µM)	0.5
Reverse primer (20 µM)	0.5
Water	6.5
Template (DNA extract)	5

3.2.4.2 Sequence analysis

The PCR product obtained by the mtCOI universal PCR were confirmed by gel electrophoresis and sequenced (Eurofins Genomics). The contig was assembled using Seqman Pro 15 (Lasergene) and cropped to comply with the sequences deposited in the *B. tabaci* mtCOI database (Global *Bemisia* dataset release version 15 May 2017) as described by Boykin and colleagues (Boykin, *et al*. 2014). The coverage for this fragment was 2. The reference sequences and the obtained sequence were aligned using the MUSCLE algorithm in Megalign Pro 15 (Lasergene). Based on the alignment, a phylogenetic tree was build using the Neighbor-Joining BioNJ algorithm (Gascuel, 1997). The tree was visualized with Figtree (Rambaut 2012) and routed using the provided outgroup sequences.

3.3Results

3.3.1 Effects of pesticide treatment on whitefly population in 2017

Analysis of data collected for two months, from February to March 2017 is summarized in Table 3.6. It shows that the average number of adult whiteflies in the greenhouses treated with a combination of *A. swirskii* and selected compatible chemical insecticides (AS + CH), was significantly ($p<0.05$) higher than the average in the greenhouses treated with synthetic chemical insecticides (CH). It was also significantly ($p<0.05$) higher than the mean of adult whiteflies in greenhouses treated with *A. swirskii* (AS) alone during Feb- March. However, in April- May and June to July, adult whiteflies in AS treated greenhouses were higher and significantly ($p<0.05$) different from the ones treated with chemical insecticides. April to May averages show that both CH and AS + CH treatments significantly ($p<0.05$) controlled the adult whitefly compared to AS treatment. There was no significant difference between the two treatments (CH and AS + CH).

All the applied insecticide treatments did not have any significant effect on the population of the whitefly eggs for the data collected from Feb to March. However, over time there were differences in the averages of whitefly eggs population where those in AS treatment were significantly ($p<0.05$) higher than those of CH and AS + CH treatments. Once again, AS was significantly less effective than the rest of the two insecticide treatments. In the same period, no whitefly egg was recorded in any of the greenhouses treated with AS or AS + CH. The average quantity of the eggs counted from greenhouses treated with AS alone was significantly higher than the means in the other greenhouses treated with either CH or AS + CH. Whitefly adults’ density under AS treatment was increasing with time while the AS + CH reduced (Table 3.6).

Table 3.6: Effects of different insecticide treatments on adult whiteflies and eggs population in 2017

Months	Treatment	Adult whiteflies			Whitefly eggs		
		Mean [adult whitefly/m^2]	Standard deviation	LSD $_{0.05}$	Mean [eggs/m^2]	Standard deviation	LSD $_{0.05}$
Feb - Mar	AS + CH	0.034a	0.026	0.016	0.008b	0.022	0.018
Feb - Mar	CH	0.002b	0.004	0.016	0.000b	0.000	0.018
Feb - Mar	AS	0.000b	0.000	0.016	0.000b	0.000	0.018
Apr - May	AS + CH	0.021b	0.017	0.141	0.011b	0.022	1.745
Apr - May	CH	0.001b	0.001	0.141	0.000b	0.000	1.745
Apr - May	AS	0.180a	0.257	0.141	1.722 a	3.696	1.745
Jun - Jul	AS + CH	0.009b	0.007	0.202	0.000b	0.000	1.594
Jun - Jul	CH	0.002b	0.003	0.202	0.000b	0.000	1.594
Jun - Jul	AS	0.246a	0.476	0.202	2.503a	4.401	1.594

Legend
AS = *Amblyseius swirskii.*
AS + CH = *Amblyseius swirskii* + synthetic chemical pesticides.
CH = Synthetic chemical pesticides.
Means followed by the same letter are not significantly different.

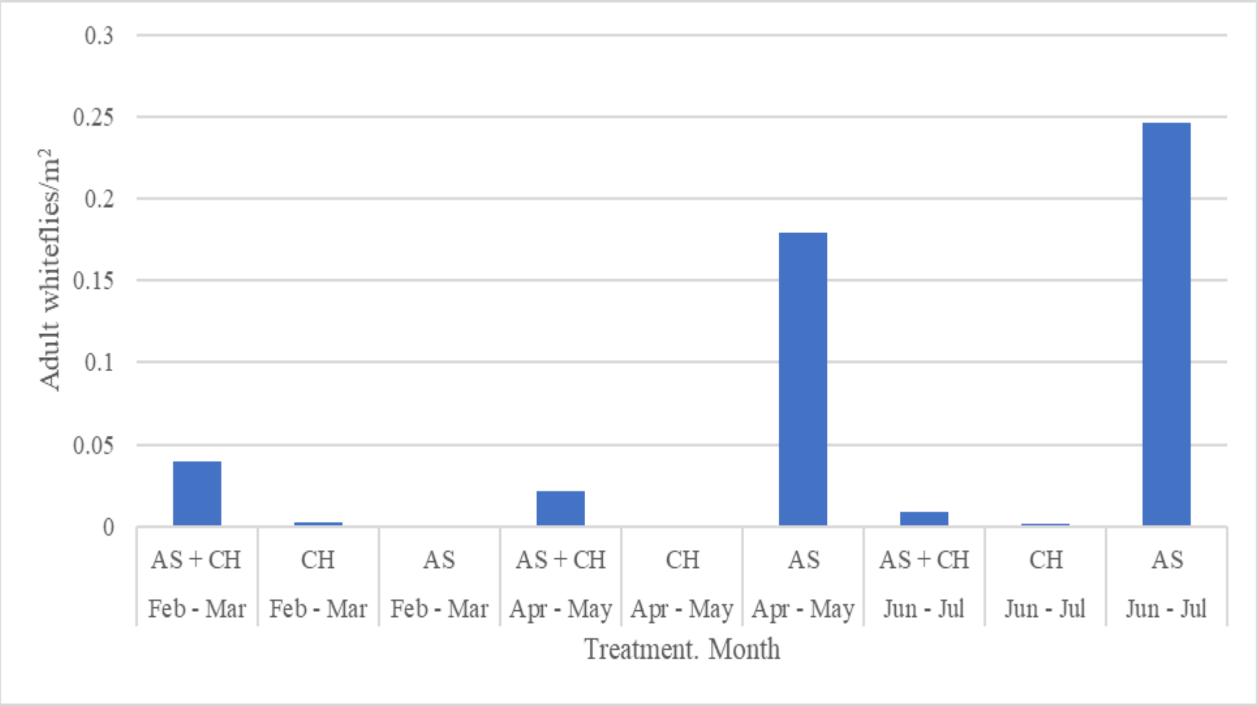


Figure 3.2: Trend of adult whiteflies population from Feb to July in 2017

3.3.2Effects of pesticide treatment on whitefly population in 2018

Unlike 2017, the analysis of data collected in the two months after planting (February and March) showed that there was no significant difference in the effects of the three insecticide treatments on both the population of the adult whitefly and the eggs (Table 3.77).

A similar trend of results in 2017 was observed in 2018 where the analysis of data collected in the third and fourth months after planting (April and May) displayed that the application of synthetic chemical insecticides ha the least population of adult whitefly but the difference between the means of adults in greenhouses treated with AS + CH and CH were not significant (Table 3.7). The application of *A. swirskii* on its own had higher population of the adult whitefly. The average number of whitefly adults in the greenhouses treated with AS was significantly ($p<0.05$) higher than in the ones treated with either of the two treatments. Synthetic chemical insecticides (CH) significantly ($p<0.05$) reduced the egg population compared to AS but was not significantly different from AS +CH. AS +CH compared to AS in reducing the eggs population (Table 3.7).

The last two months in Poinsettia season of 2018, witnessed significant differences in the effects of the insecticide treatments on the population of both adults and eggs. The synthetic insecticide treatment (CH) had significantly ($p<0.05$) lower populations of both the whitefly adults and the eggs compared to AS and AS+ CH treatments. AS and AS +CH did not differ in effect on either whitefly adults or eggs for the month of April- May (Table 3.77). Figure 3.3 summarizes the trend of whitefly population across the three treatments from February to July. The trend of the adult whiteflies population in 2018 showsgradual increase from February – to July in greenhouses treated with *A. swirskii* treatment (AS).

Table 3.7: Effects of different pesticide treatments on adult whiteflies and eggs population in 2018.

Month	Treatment	Adult whiteflies			Whitefly eggs		
		Mean [adult whitefly/m^2]	Standard deviation	LSD $_{0.05}$	Mean [eggs/m^2]	Standard deviation	LSD $_{0.05}$
Feb - Mar	AS + CH	0.011b	0.009	0.012	0.003b	0.008	0.007
Feb - Mar	CH	0.008b	0.016	0.012	0.001b	0.003	0.007
Feb - Mar	AS	0.017b	0.012	0.012	0.004b	0.009	0.007
Apr - May	AS + CH	0.021ba	0.017	0.026	0.009ba	0.016	0.614
Apr - May	CH	0.004b	0.006	0.026	0.000b	0.000	0.614
Apr - May	AS	0.040a	0.052	0.026	0.886a	1.198	0.614
Jun - Jul	AS + CH	0.007b	0.059	0.368	0.000b	0.000	3.471
Jun - Jul	CH	0.002b	0.017	0.368	0.000b	0.000	3.471
Jun - Jul	AS	0.456a	4.100	0.368	4.018a	6.965	3.471

Legend

AS = *Amblyseius swirskii*

AS + CH = *Amblyseius swirskii* + synthetic chemical pesticides

CH = Synthetic chemical pesticides

Means followed by the same letter are not significantly different within the same period of data collection.

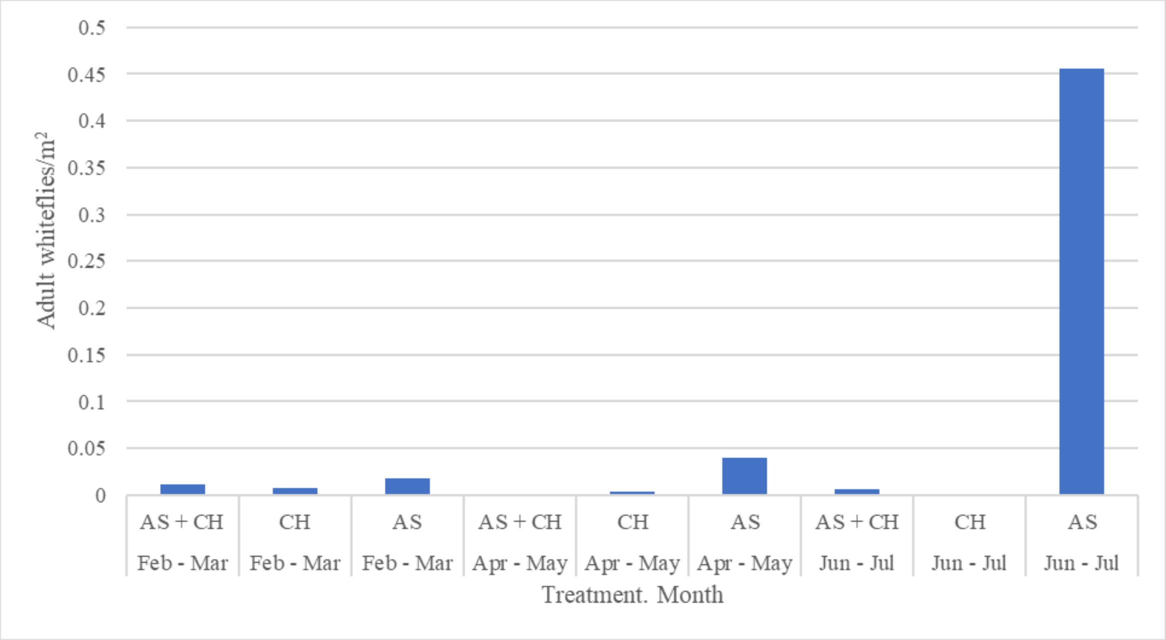


Figure 3.3: Trend of adult whiteflies population from Feb to July in 2018

3.3.3Influence of variety treatment on whitefly population - 2017

The influence of Poinsettia varieties on the population of the adult whitefly was not significantly different among the three tested varieties between February- March, and April - May. However, there was an indication that Red variety with vertical leaves (RVL) was the least affected while the Red variety with horizontal leaves (RHL) was the most affected. June and July, shows that RHL significantly ($p<0.05$) attracted more whitefly adults than RVL or WHL but the effects of RVL and WHL were not different (Table 3.8). The same observation was made on the effects of variety on whitefly eggs management. There was no significant difference in the effects of variety on reducing the population of whitefly eggs from the analysis of data collected in February &March and April &May. However, the number of eggs recorded in June and July showed that the RHL had significantly more eggs laid on them than on either of the two remaining varieties (Table 3.8).

Table 3.8: Influence of the varieties on adult whiteflies and eggs population in 2017

		Adult whiteflies			Whitefly eggs		
Month of data collection	Treatment	Mean [adult whiteflies/m^2]	Standard deviation	LSD $_{0.05}$	Mean [eggs/m^2]	Standard deviation	LSD $_{0.05}$
Feb - Mar	RVL	0.013b	0.018	0.022	0.001b	0.003	0.018
Feb - Mar	WHL	0.019b	0.034	0.022	0.007b	0.022	0.018
Feb - Mar	RHL	0.010b	0.017	0.022	0.000b	0.000	0.018
Apr - May	RVL	0.009b	0.015	0.141	0.000b	0.000	1.745
Apr - May	WHL	0.073b	0.176	0.141	0.010b	0.022	1.745
Apr - May	RHL	0.119b	0.224	0.141	1.723a	3.696	1.745
Jun - Jul	RVL	0.003b	0.003	0.202	0.000b	0.000	1.594
Jun - Jul	WHL	0.006b	0.007	0.202	0.033b	0.100	1.594
Jun - Jul	RHL	0.248a	0.475	0.202	2.470a	4.421	1.594

Legend
AS = *Amblyseius swirskii*
AS + CH = *Amblyseius swirskii* + synthetic chemical pesticides
CH = Synthetic chemical pesticides
Means followed by the same letter are not significantly different within the same period of data collection.

3.3.4 Influence of variety treatment on whitefly population - 2018

The effect of variety on the whitefly adults or eggs population was not quite different from the observation made in the previous year. The three varieties did not differ in attracting whiteflies and in the egg populations collected. However, RHL had slightly more averages of whitefly population on them while RVL and WHL had fewer populations of whitefly. Variety RVL had the least number of whitefly eggs from Feb to July. Variety RHL had the most whitefly eggs in June –July experiment period (Table 3.99).

Table 3.9: Influence of the varieties on adult whiteflies and eggs population in 2018.

		Adult whiteflies			Whitefly eggs		
Month of data collection	Treat ment	Mean [adult whiteflies/ m^2]	Standard deviation	LSD $_{0.05}$	Mean [eggs/m^2]	Standard deviation	LSD $_{0.05}$
Feb - Mar	RVL	0.016a	0.015	0.012	0.000a	0.000	0.007
Feb - Mar	WHL	0.008a	0.010	0.012	0.007a	0.011	0.007
Feb - Mar	RHL	0.012a	0.013	0.012	0.001a	0.003	0.007
Apr - May	RVL	0.013a	0.012	0.026	0.005a	0.014	0.614
Apr - May	WHL	0.018a	0.019	0.026	0.309a	0.623	0.614
Apr - May	RHL	0.033a	0.055	0.026	0.581a	1.201	0.614
Jun - Jul	RVL	0.012a	0.027	0.368	0.000a	0.000	3.471
Jun - Jul	WHL	0.137a	0.295	0.368	0.687a	1.805	3.471
Jun - Jul	RHL	0.315a	0.692	0.368	3.331a	7.099	3.471

Legend

AS = *Amblyseius swirskii*
AS + CH = *Amblyseius swirskii* + synthetic chemical pesticides
CH = Synthetic chemical pesticides
Means followed by the same letter are not significantly different.

3.3.4 Weather patterns

In both seasons, the temperature drops from May to July getting below 24^{o}C. The trend is that it got slightly cooler from May to July. 2018 was slightly cold compared to 2017 and the all time low temperature occurred at the end of 2018 in July (Fig 3.4). 2018 was more humid than 2017. Humidity got higher from the month of May to June (Fig.3.5).

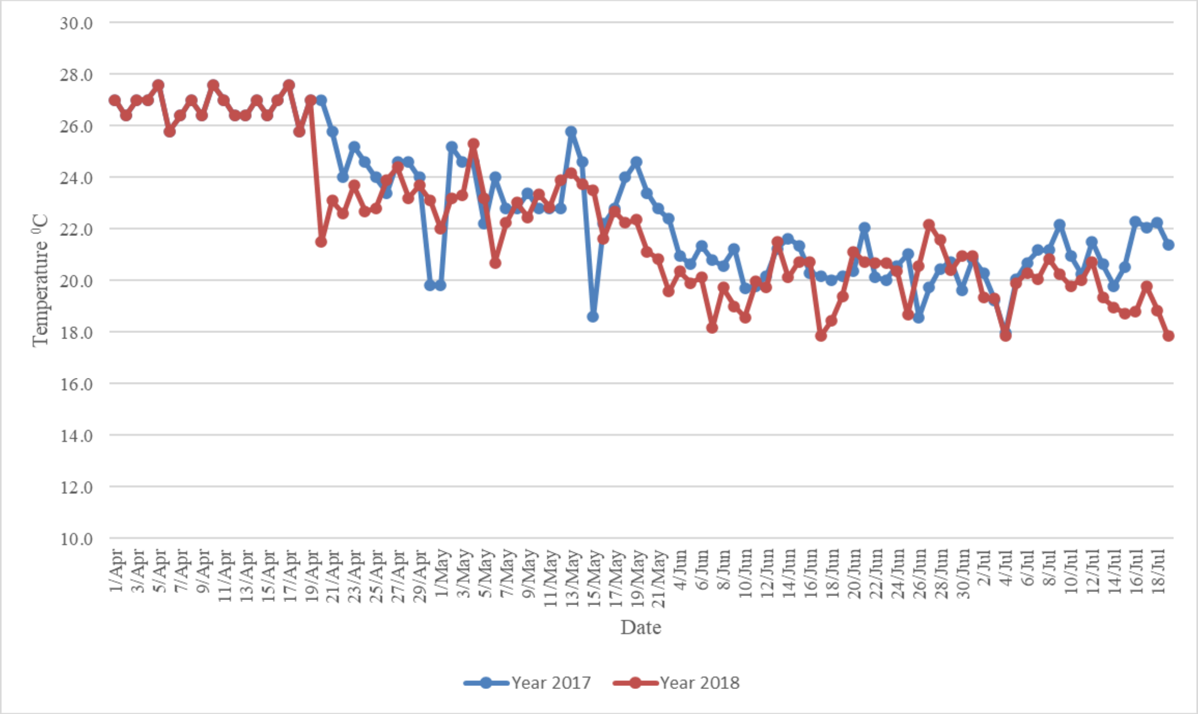


Figure 3.4: Trend of temperature in the farm in 2017 and 2018

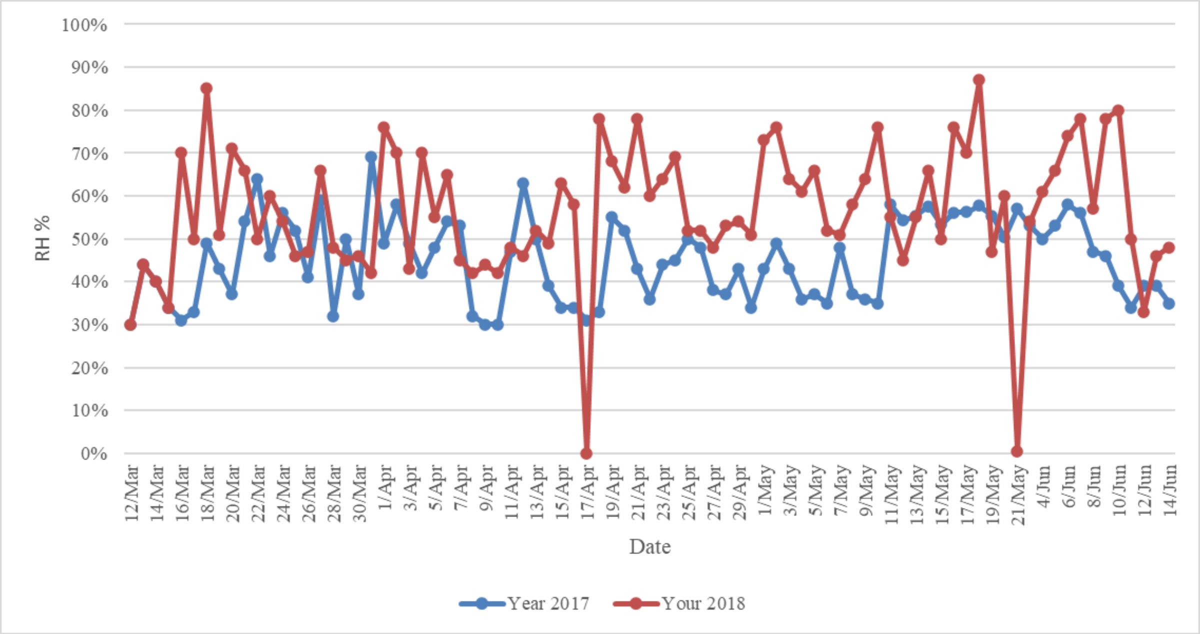


Figure 3.5: Trend of relative humidity in the farm in 2017 and 2018.

3.3.5 Poinsettia yield in the first season – 2017

The yield per square meter was recorded for each variety in every treatment in 2017. Chemical treatment (CH) had the highest yield and was significantly ($p<0.05$) higher than either of the two, AS and AS+ CH treatments (Table 3.10).

Table 3.10: Effects of insecticide treatments on the Poinsettia yield in 2017.

TREATMENT	**MEAN [Cuttings/m2]**	**STD. DEVIATION**	**LSD $_{0.05}$**
AS	200.4b	99.2	63.2
AS +CH	199.8b	84.8	63.2
CH	400.4a	72.4	63.2

Legend
AS = *Amblyseius swirskii*
AS + CH = *Amblyseius swirskii* + synthetic chemical pesticides
CH = Synthetic chemical pesticides

3.3.6 Poinsettia yield in the second season – 2018.

Once again, the areas treated with Chemical pesticide (CH) had significantly ($p<0.05$) highest yield which compared to the two, AS and AS +CH treatments. AS and AS +CH did not differ in the average yield harvested although that from AS was slightly higher (

Table 3.11)

Table 3.11: Effects of pesticide treatments on poinsettia yield in 2018

TREATMENT	**MEAN [Cuttings/m2]**	**STD. DEVIATION**	**LSD $_{0.05}$**
AS	191.6b	34.6	126.4
AS +CH	184.4b	77.1	126.4
CH	339.0a	59.1	126.4

Legend
AS = *Amblyseius swirskii*
AS + CH = *Amblyseius swirskii* + synthetic chemical pesticides
CH = Synthetic chemical pesticides

3.3.7 Identification of the whitefly sample

The results of running mtCOI PCR product through gel electrophoresis, showed that the gene was of whitefly *B. tabaci*. (Figure 3.). Polymerase chain reaction (PCR) results using biotype specific primer combinations against the extracted DNA, further revealed that the PCR product was of a whitefly in the group of *Bemisia tabaci* biotype Q (Figure 3.7: Biotype-specific PCR.)

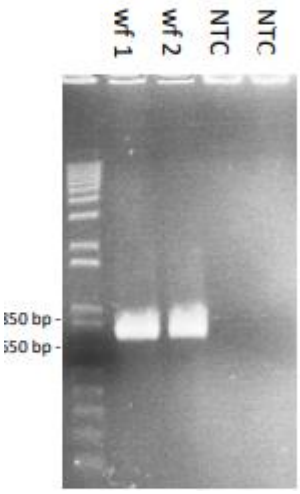


Figure 3.6: universal PCR products.Whitefly samples .

5 µl of each uni PCR product (wf1 and wf2 are duplicates of the whitefly DNA extract, NTCs are negative template controls) were separated on a 0.8 (w/v) agarose gel and stained with Serva stain. Universal PCR was run at 50 annealing temperature. Expected fragment size for *B. tabaci* should be 745 bp.

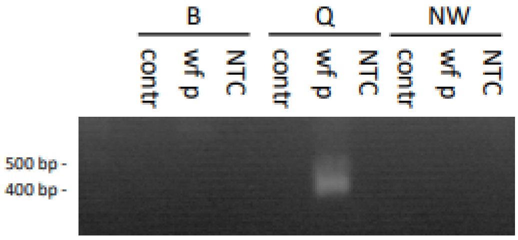


Figure 3.7: Biotype-specific PCR.

20 µl of each PCR product (control is a negative control sample, wf P is the unknown whitefly sample, NTC is a negative control) were separated on a 0.8 (w/v) agarose gel and stained with Serva stain. The group letter indicates the primer combination and thus the biotype.

Comparison of the obtained sequence with sequences from a curated collection (Boykin *et al.*, 2014) refer to section 3.2.4.2, places the unknown sample in a *B. tabaci* clade containing sequences of Mediterranean biotype Q specimen. It was also established that the whitefly sample could have originated from Portugal.

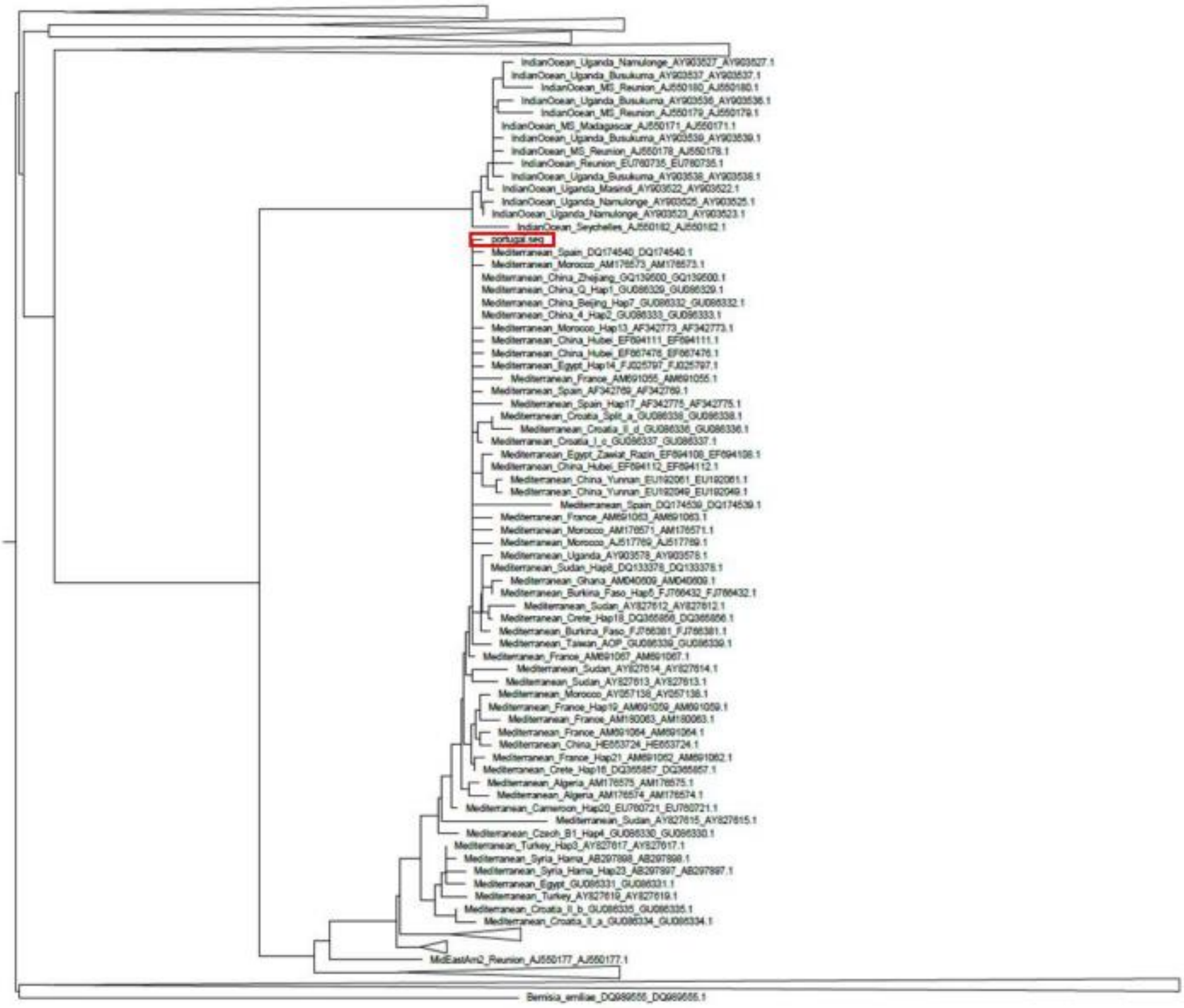


Figure 3.8: Phylogenetic tree based on mtCOI sequence.

The mtCOI sequence obtained from the unknown sample (Portugal, marked with red box) was aligned (MUSCLE) with 544 reference *B. tabaci* mtCOI sequences as well as 21 sequences derived from related species. Based on the alignment a tree was constructed (BioNJ) and visualized with Figtree. For rooting the sequences of the related sequences were used. Clades not relevant for this study were collapsed.

3.3 Discussion

The whitefly samples in the greenhouse were identified as *Bemsia tabacii* biotype Q through the process of DNA sequencing at Macrogen lab in Netherlands. This study revealed that a combination of *A. swirskii* with some selected compatible synthetic insecticides (Buprofezin and Abamectin) can provide an effective and reliable strategy for controlling whitefly (*B. tabaci*) in a greenhouse production of vegetative Poinsettia mother stock in Kenya. This is evident from the fact that the effects of chemical insecticides (CH treatment) on the population of whitefly eggs or adults was not different from that of the combination of *A. swirskii* and the compatible chemical insecticide (AS +CH) treatment, and it became better over time reducing whitefly populations while in AS alone the populations and the eggs increased. This finding confirms the reports by earlier studies that targeting treatments at certain life-stages of a pest considerably improve the control of its population (Candy, 2003, Cuthbertson *et al*., 2003). In this work, the combination of *A. swirskii* and some compatible chemical and biological insecticides constituted different products that targeted different life stages of the whitefly (*B. tabaci*). For example, *A. swirskii* targeted whitefly juvenile since it predates on eggs and crawlers of *B. tabaci (*Nomikou *et a*l., 2003) while Buprofezin (Applaud) and abamectin (Dynamec) also targeted the eggs.

The findings also showed that the use of *Amblyseius swirskii* on its own as a strategy for managing whitefly (*B. tabaci*) in a greenhouse production of vegetative Poinsettia mother stock in Kenya is not effective.   The results concur with the findings of Brownbridge and Buitenhuis (2017) that the use of biocontrol agents as a single method is rarely successful in pests' control. Rather, success is realized by combining it with other methods in an integrated approach. This can also be attributed to the fact that Poinsettia is an obligate short-day plant and therefore if grown to produce unrooted cuttings then they must be purely vegetative. (Roll, 1997). Vegetative Poinsettia stock has no flower to produce pollen or nectar for *A. swirskii* mites in cases of low supply of prey such as whitefly and thrips. This situation could have played a role in the observed reduced efficacy of the predatory mites. Different studies have suggested that phytoseiid mites like *A. swirskii* are particularly persistent on plants that provide flowers and additional floral nectaries as an alternative source of food besides the targeted prey like thrips or whitefly (Shipp and Ramakers, 2004). The study by Athurs *et al*. (2009) showed that *A. swirskii* was highly effective in maintaining low population of *S. dorsalis* in sweet pepper throughout the test just by a single mite release. The predatory mite was also reported to be effective in controlling Western Flower Thrips (*Frankliniella occidentalis*) in greenhouse cucumbers (Messelink *et al.*, 2005). This was partly due to the supply of alternative food as nectar and pollen by the flowering sweet pepper stock or cucumbers. For better whitefly population control, other studies recommend the use of more than one natural enemies as a means of enhancing the control of the pest in greenhouses as long as the biocontrol agents are compatible (Chow *et al*., 2008). For example, the study by Buitenhuis *et al*. (2008) shows that *N. cucumeris* and *A swirskii* are not compatible with both predators feeding on the juvenile stages of the other. Chow *et al.* (2008) reported that *Orius insidiosus* and *Amblyseius degenerans* were less effective in controlling population of Western Flower Thrips in the stock of greenhouses roses when they were released together because of the intra-guild predation between them.

The prevailing climatic condition in the Poinsettia greenhouses could have also reduced the effectiveness of the predatory mites. The greenhouses became cooler from June to July with the average temperature falling to about 20 ^{0}C from about 27 ^{0}C in April (Fig3.4). *A. swirskii* is native to Eastern Mediterranean region which is warm and humid sub-tropical climate (EPPO, 2013). Therefore, this predator may be less effective in cooler conditions and at a reduced relative humidity (Lee and Gillespie, 2011).

Another strategy of managing pest is by screening varieties for the selection of more pest resistant varieties as a way of optimizing production cost and sustaining crop yield in a more environment friendly condition (Junior *et al.*, 2003). Studies have demonstrated that *Bemisia tabaci* prefers plants or cultivars with leaves having thick trichomes for egg laying and the eggs are always stalked (*Amad et al.*, 2014). In some resistant varieties, few eggs hatch into nymphs (Vieira *et al.*, 2016) while in some the number of adults developing from nymphs are reduced because of antibiosis (Fekri *et al.*, 2013). This study screened three Poinsettia varieties against whitefly mainly *B. tabaci* preference. There were red variety with vertical Leaves (RVL), Red variety with Horizontal Leaves (RHL) and White variety with Horizontal Leaves (WHL). The three varieties did not show differences in mitigating whitefly infestation. However, there was an indication that RHL was more susceptible to whitefly infestation with slightly higher averages observed compared the other two varieties used. In 2017 June – July, the population of whitefly adults or eggs on RHL was significantly higher than in the rest of the two varieties. In the following season the populations were consistently higher for RHL although not different from the rest of the varieties screened. The whitefly did not lay eggs on RVL with none or few numbers of eggs laid. This is indicates that it is not preferred compared to the other two varieties screened. Is it because the leaves are vertical?.

The pesticide treatment had significant impact on the whitefly population and hence yield of the tested varieties. Greenhouses treated with chemicals had higher yields recorded than the combination of *A. swirskii* and a synthetic chemical insecticides or *A. swirskii* alone. This could be attributed to the early control of the whiteflies and hence little or no sap was drawn from the plants.

The efficacy of biocontrol agents (*A. swirskii*) in the management of whiteflies in greenhouse production of Poinsettia is relatively low. However, integrating *A. swirskii* with compatible synthetic insecticides enhanced its efficacy in controlling *B. tabaci* in greenhouse Poinsettia production. This, therefore, reduces or eliminates problems associated with use of synthetic pesticides. It is also not advisable to exclusively use the biocontrol agents in the management of whiteflies in greenhouse production of Poinsettia. Instead, if they must be used, they should always be combined with other compatible chemical pesticides and/or biopesticide.

CHAPER 4:

EFFECT OF THE TYPE OF BENCHES AND FLOOR ON MANAGEMENT OF SLUGS IN GREENHOUSE POINSETTIA PRODUCTION

Abstract

Slugs (Gastropoda) are among the pests known to cause major damages to food and horticultural crops. The management of mollusc pests on a crop involves the use of pesticides like metaldehyde, methiocarb and iron phosphate. Metaldehyde and Methiocarb apart from being expensive are not registered for use on ornamentals in Kenya. Therefore, there is a need to come up with slug management method which would also easily fit in IPM programme within the greenhouse. Such methods may include the use of natural enemies, improvement of greenhouse structure, breeding for more resistant varieties and the use of selected chemical molluscicides that are compatible with IPM programme. The current study evaluated the efficacy of using physical barriers in controlling slugs in a greenhouse production of Poinsettia in Kenya. There were two factors in this study. The physical barrier and the variety factors. Under the physical barrier were three treatments: Benches supported on steel pillars/legs in a pumice floor; benches supported on steel pillars in a concrete floor and the control which was benches supported on concrete pillars in a pumice floor. The latter is control because, it was the common greenhouse structure used by Poinsettia growers. There were three treatments on the variety factor: red variety with horizontal leaves (RHL), red variety with vertical leaves (RVL), white variety with horizontal leaves (WHL).

The experiment was conducted in two seasons, 2017 and 2018, in Muranga County. The study evaluated the effects of three different physical barriers and the three different varieties on the management of slug’s population in greenhouse production of Poinsettia.

The study revealed that the applied physical barrier was effective in reducing the slugs population. Greenhouses with concrete floors and benches supported on steel pillars had the greatest impact on controlling slugs but this effect was not significantly better than that of the greenhouses with pumice floors and benches supported on the concrete benches. Slug population was significantly higher in greenhouses with pumice floor and benches supported on the concrete blocks. The impact of variety in managing slug's population in the greenhouse also had a significant effect on slug population. The Red variety with horizontal leaves (RHL) was the most vulnerable and significantly attracted more slugs on the benches than the remaining two varieties. It is, however, worth noting that with a good greenhouse structure like a concrete floor and benches supported on steel stands, the effect of variety may not be important. Meaning that, irrespective of variety type, Poinsettia grows well in this kind of greenhouses without the problems of slugs.

4.1Introduction

Slugs are among the pests known to cause major damage to horticultural crops. According to Zala *et al.* (2018), slugs damage the crops especially by eating plant leaves and destroying seedlings at their emergence. The symptom of the damages inflicted by slugs differ from one crop variety to the other but the most common is raged holes on the plant leaves, with slime trails often close to the damaged spots (Douglas & Tooker, 2012). Effective management of slugs is thus vital in horticultural crops production. Zala *et al.* (2018) asserts that several methods are recommended for controlling slugs including physical barriers, baited traps among others. However, this is not justified by experimental evidence and therefore, the effectiveness of each of the methods needs to be tested and approved across different crop varieties.

One of the most effective means of managing slugs is by altering their habitat to make it less favourable for their survival or mobility. In the tropical and subtropical regions, Poinsettia and other high value crops are grown on benches raised from the ground mainly supported with pillars of concretes for better management of pest and diseases (Jena *et al.*, 2020). The floors are either left bare or filled up with pumice, gravels, sand etc. Some growers also use mypex to cover the floors of their greenhouses. Benches supported on block pillars on the pumice/gravel floors might create favourable condition for the survival of slugs which attack and destroy the growing Poinsettia crop. This study focused on investigating the effectiveness of physical barriers in controlling slugs in greenhouse production of Poinsettia. Capinera (2018) indicates that the materials used in physical barriers influences their effectiveness but highlighted the scarcity of data on effectiveness of different materials. This study therefore assessed the effect of the different types of benches and greenhouse floors (physical barriers) on the management of slugs in greenhouse production of Poinsettia where a lot of misting and watering is done during the establishment or the crop that renders the use chemical Molluscicide in effective.

4.2 Materials and Methods

4.2.1 Study area

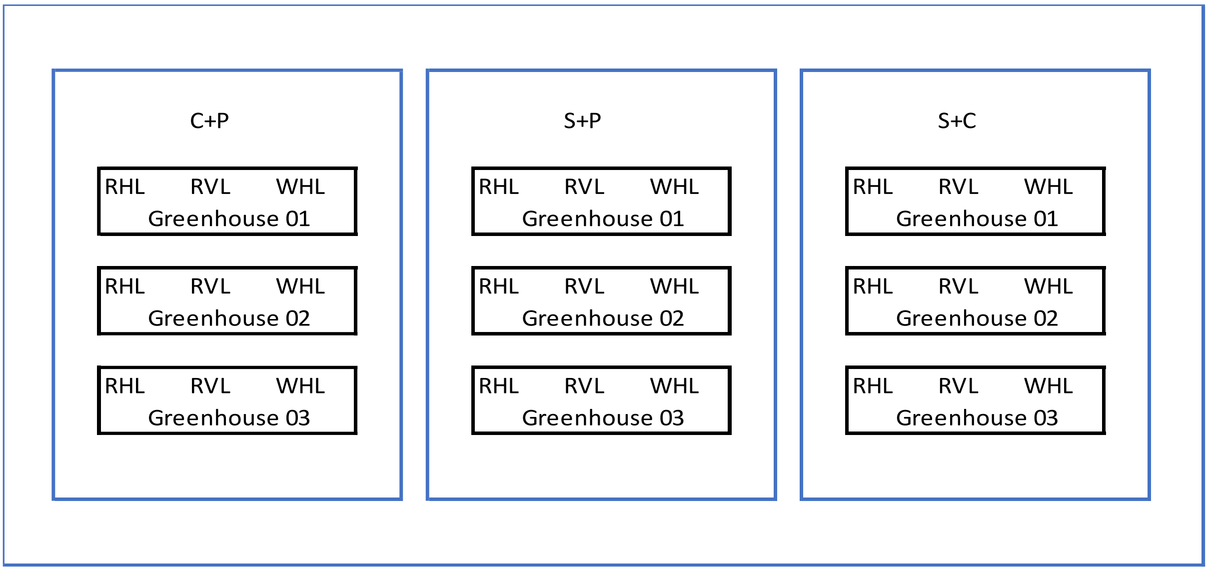
The study was also conducted in Kakuzi Mitubiri ward in Murang’a County within altitude ranges of 914m – 3,533m above sea level along the slopes of the Aberdare Ranges. The study was however conducted in a controlled greenhouse environment in two different years (2017 and 2018). For each year, nine greenhouses were used. Each greenhouse had 75 benches measuring 1m wide, 40m long, 0.7m apart and 0.75m above the ground each. One bench supported 640 pots filled with pure pumice uniformly arranged in four rows along the benches at pots density of 16 pots/m^2.

4.2.1Treatments and experimental design

4.2.1.1Greenhouse experiment and design

Completely Randomized Design of two factors was used in this study. The first factor was the physical barrier which consisted of three treatments including: benches with concrete stands on a pumice floor (C+P) in this case the control; benches with steel stands on a pumice floor (S+P); and benches with steel stands on a concrete floor (S+C).

The second factor was the variety of Poinsettia: Red Variety with Horizontal leaves (RHL), Red Variety with Vertical leaves (RVL) and White Variety with Horizontal leaves (WHL). The treatments were applied differently in three different sets of green houses, with each set having 3 greenhouses. For better understanding refer to Figure 4.1



Legend: Two factors of treatments. Total of 9 greenhouses used: 3 Physical barriers treatment, 3 varieties treatments and 3 replications. C+P, S+P and S+C were different physical treatments for benches with concrete pillars on pumice floor, benches with steel pillars on pumice floor, and benches with still pillars on concrete floor respectively. RHL, RVL and WHL are red variety with horizontal leaves, red variety with vertical leaves, and white variety with horizontal leaves.

Figure 4.1: Completely Randomized Design (CRD) for Physical barriers and varieties.

The first factor was the physical barrier which included three treatments. The treatments were benches with concrete pillars on pumice floor(C+P), benches with steel pillars on pumice floor (S+P), and benches with still pillars on concrete floor (S+C) treatments. The second factor was the variety which had three treatments too. The treatments were: red variety with horizontal leaves (RHL), red variety with vertical leaves (RVL), and white variety with horizontal leaves (WHL). For each physical barrier treatment, there were three replicates. Each replicate consisted of whole greenhouse. Therefore, each physical barrier treatment consisted of 3 greenhouses. This arrangement resulted into 9 greenhouses in total. Each greenhouse had a stock of the three varieties such that the plant for each variety were planted on 6 beds.

4.2.1.2Application of the treatments

Each treatment was replicated three times. That is, for each physical barrier treatment, there were three greenhouses. Each of the three greenhouses had the 3 varieties of Poinsettia. Experimental plots consisted of 6 benches [gross area of 360m^2]. The effect of the crop development stage on the population of slugs was studied by inspecting the slug population at the various development stages of the crop. The impact of relative humidity on the population distribution of slugs was also studied.

The first treatment was the steel benches with concrete stands on a pumice floor [Control] (C+P). The second treatment was steel benches with steel stands on a pumice floor (S+P). While the third treatment was the steel benches with steel stands on concrete floor (S+C). See Figures 4.2 – 4.4 for more information. Three greenhouses were used for each treatment. Three varieties were common across all the treatments.

Figure 4.2: Steel benches supported with steel pillars on a concrete floor.

The floor is clean with no debris suitable for slugs-egg laying and hiding.

Figure 4.3: Steel benches supported with steel pillars on a pumice floor.

The floor is filled with pumice creating a suitable hideout and conducive egg laying environment for slugs. The fertilizer drains into the soil making it moist which is an environment most suitable for the survival of slugs.

Figure 4.4: Steel benches supported with concrete blocks on a pumice floor (C+P).

4.2.2 Data Collection and Analysis

4.2.2.1 Data collection

Slugs' presence on the greenhouse floor, concrete stands, benches and in the foliar was inspected and if found collected and recorded across the three varieties in all treatments on weekly basis. The prevailing climate information out of the greenhouses was monitored using climate sensors from watchdog company. The information collected was on temperature and relative humidity.

4.2.2.1.1 Sampling slugs for identification

Slugs were selected from the nine greenhouses and placed in ziplock moist bag for pictorial identification. A total 50 slugs were collected towards the end of the first and second seasons. The identification was done by comparing the physical features outlined in Table 4.1 against pictures of different species.

Table 4.1: Template for identifying slugs physical features.

Feature	**Description**
Shell	
Body colour	
Length	
Tentacles and head colour	
Tentacle contraction	
Body shape	
Presence of keel	
Mantle	
Sole colour	
Tail	
Shape of contracted body	

4.2.2.2 Data analysis

The slug population was subjected to analysis of variance (ANOVA) to evaluate treatment effects. The Fischer's least significant difference (LSD) test was applied to decides whether there were significant differences between means of different treatments.

Data analysis was done for the purposes of hypotheses testing and it was done with the help of Microsoft Excel worksheet and the Statistical Package for Social Sciences (SPSS) version 29.0.1.0. The data collected was first entered in a Microsoft Excel worksheet to aid in data cleaning. This involved the removal of typo mistakes and outliers. Factorial analysis was done using the Completely Randomized Design approach. In this regard, the difference in the mean slug population distribution was statistically assessed in the light of the treatments in the two factors. Statistical differences in the mean slug population distribution between different treatments were used to assess the effectiveness of the different types of treatments.

4.3 Results

4.3.1 Effect of physical barriers on slug population in the first season- 2017

The results shows that slug density was significantly ($p<0.05$) higher in greenhouses with benches supported on rock/concrete pillars and pumice floor. No slug was recorded in all greenhouses with concrete floors (S+C) in the whole season of 2017. An average of 0.09 slugs/m^2 square were recorded in the greenhouses with benches supported on steel stands but on pumice floor (S+P). However, the mean population of slugs between S+C and S+P treatments were not significantly different. 1.90 slugs/m^2 were recorded under the treatment of benches supported on concrete stands and pumice floor (C+P). The mean slug population for this treatment is significantly higher than for any of the remaining two treatments (Table 4.2).

Table 4.2: Mean slug population in greenhouses with different physical barriers in 2017

Physical barrier	**Mean (slugs/m^2)**	**Standard deviation**	**LSD$_{0.05}$**
C + P	1.90a	1.28	0.52
S + C	0.00b	0.00	0.52
S + P	0.09b	0.11	0.52

Legend

C+P = Benches with Concrete stands on a Pumice floor; S+P = Benches with Steel stands on a Pumice floor; S+C = Benches Steel stands on a Concrete floor

Means followed by the same letter are not significantly different.

4.3.2 Effect of physical barriers on slug population in the first season- 2018

In the second season (2018), once again, no slug was recorded in all greenhouses with concrete floor (C+P). Some experimental plots in greenhouses with pumice floor but with benches supported on steel stands (S+P treatment) had some slugs (0.24a slugs/m2) but this effect was not significantly different from that of S+C. All plots in the greenhouses with pumice floor and benches supported on concrete stands (C+P treatment) recorded some slugs (2.10b). The treatment of C+P was significantly less effective than either S+C or S+P (Table 4.3). At night the humidity was lower in greenhouses with benches supported on steel stands and concrete floors (S+C) but highest in greenhouses with benches supported on concrete blocks on a pumice floor (C+P) as shown in Fig.4.5

Table 4.3: Mean slug population in greenhouses with different physical barriers in 2018**.**

Physical barrier	**Mean (slugs/m^2)**	**Standard deviation**	**LSD$_{0.05}$**
C + P	2.10a	1.55	0.58
S + C	0.00b	0.00	0.58
S + P	0.24b	2.15	0.58

Legend

C+P = Benches with Concrete stands on a Pumice floor; S+P = Benches with Steel stands on a Pumice floor; S+C = Benches Steel stands on a Concrete floor

Means followed by the same letter are not significantly different.

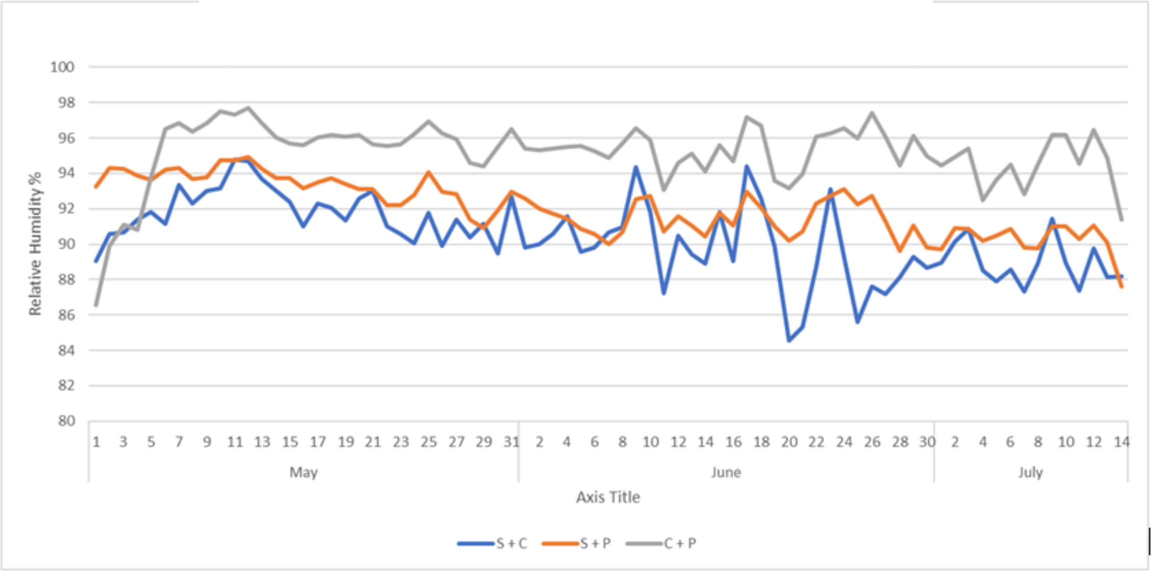


Figure 4.5: Trend of relative humidity in the greenhouses in 2018

4.3.3 Influences of Poinsettia varieties on the slug population in first season - 2017

The Red Variety with Horizontal leaves (RHL) attracted a lot of slugs (1.10 slugs/m^2) and the population was significantly ($p<0.05$) higher on this variety than on RVL and WHL. The effect of RVL and WHL on the slug population were not different as the two varieties attracted near equal populations, 0.45 and 0.43 slugs/m^2, respectively (Table 4.4). The results show that red variety with horizontal leaves attracted the highest population of slugs especially in Concrete with Pumice floor treatment.

Table 4.4: Influence of variety on the population of slugs in 2017

Variety	**Mean (slugs/m^2)**	**Standard deviation**	**LSD$_{0.05}$**
RHL	1.10a	1.70	0.52
RVL	0.45b	0.75	0.52
WHL	0.43b	0.68	0.52

Legend

RVL = Red variety with vertical leaves; WHL = White variety with horizontal leaves; RHL = Red variety with horizontal leaves.

Means followed by the same letter are not significantly different.

4.3.3 Influences of Poinsettia varieties on the slug population in the second season - 2018

Like the previous season, varieties RVL and WHL were the most resistant to slugs. Their influence on slug's population was not significantly difference as both varieties recorded a mean of 0.47 slugs/m^2 and 0.61 slugs/m^2 respectively. Variety RHL was the most vulnerable. It had an average of 1.27 slugs/m^2 which was significantly lower than either RVL or WHL. The impact of variety in the population of slugs was significantly effective. The Red variety with Horizontal leaves (RHL) was the most preferred by the slugs leading to significant low control on slug population than the rest of the two varieties. The influence of RVL and WHL on the slug population was not significantly different (

Table 4.5).

Table 4.5: Influence of variety on the population of slugs in 2018

Variety	**Mean (slugs/m^2)**	**Standard deviation**	**LSD$_{0.05}$**
RHL	1.27a	1.99	0.58
RVL	0.47b	0.68	0.58
WHL	0.61b	0.78	0.58

Legend

RVL = Red vertical leaves Poinsettia; WHL = White horizontal leaves Poinsettia; RHL = Red horizontal leaves Poinsettia. Means followed by the same letter are not significantly different.

1.12.1 Identification of the slugs

Slugs were identified by examining their physical features and comparing with features from pictures collected from different sources. The identified features are given in

Table 4. From the pictures obtained from Terrestrial Mollusc Key (Mclean, 2018), the physical characteristics of the slug were closely related to *Deroceras laeve* from the family of *Agriolimacidae.*

Table 4.6: Physical features of the slug samples collected from the greenhouse.

Feature	Description
Shell	Not present
Body colour	Dark brown, no marking spot, or blotches
Length	20-25mm
Tentacles and head colour	Black colour
Tentacle contraction	Contracted inverted
Body shape	Cylindrical
Presence of keel	Present, short.
Mantle	like a saddle, swollen posteriorly, had mantle groove.
Sole colour	Cream to white
Tail	Rounded.
Shape of contracted body	Bell-shaped

Figure 4.6: A sample of slug: average length is 20-25mm.

Figure 4.7: A sample of slugs: featuring head, tentacles, and mantle.

1.13 Discussion

Previous studies have reported that the use of physical barriers made of different materials or treated with different substances like repellents or irritants is effective control strategy for slugs and snails in field crop (Schuder *et al.*, 2003). This present study evaluated the effect of two types of physical barriers in managing slugs in a greenhouse production of Poinsettia. The first one is the type of a greenhouse floor as a physical barrier to the horizontal movement of slugs. The second barrier are the type of legs/pillars supporting benches which act as physical barrier to the vertical movement of slugs. The findings demonstrates that the treatment of greenhouses with concrete floor and with benches supported on steel stands (S+C) was the most effective in slug management since no slugs was found in these greenhouses. This can be attributed to the fact that concrete floors are drier than floor with pumice stones. In case of any water spillage the concrete floor dries up faster. The humidity in the greenhouses with concrete floors is lower especially at night (Figure 4.4). This situation makes them least preferred by slugs (Glen *et al.*, 1991). The skin of a slug consists of a single epithelial cell layer and for that reason stands a higher chance of losing water through desiccation in drier environment (South, 1992) a condition that makes it difficult for them to move. Concrete floor also makes it difficult for the replenishment of the active slugs' population with slugs underneath the floor, through vertical dispersal. This creates a situation where the vertical mobility of hidden slugs from below the ground to the surface and the horizontal mobility of the active slugs on the surface of the floor, are completely curtailed. Port *et al*. (2021) explains that the most reliable and effective way of managing slugs on crop is by ensuring that both the vertical mobility of slugs from under the soil and their horizontal movement on the surface are completely controlled.

It is important to note that greenhouses with concrete floors are very expensive to construct and therefore if cost becomes a serious limiting constraint than the use of pumice on the floor but with benches supported on steel stands (S+P treatment), would give a cheaper yet still effective option. In fact, from the study, the difference between this kind of greenhouses (S+P treatment) and the greenhouses with concrete floor (S+C) in terms of slug management was not significantly different.

Greenhouses with benches supported on concrete blocks in a pumice floor (C+P), registered the highest population of slugs in both seasons. Their effect on slug population was significantly lower than the effects of the aforementioned treatments. In addition to the disadvantages of the pumice floor which is creating a better breeding ground and hideouts for the slugs, the big concrete block also absorbs water during fertigation and thereby made the greenhouse more humid at night.

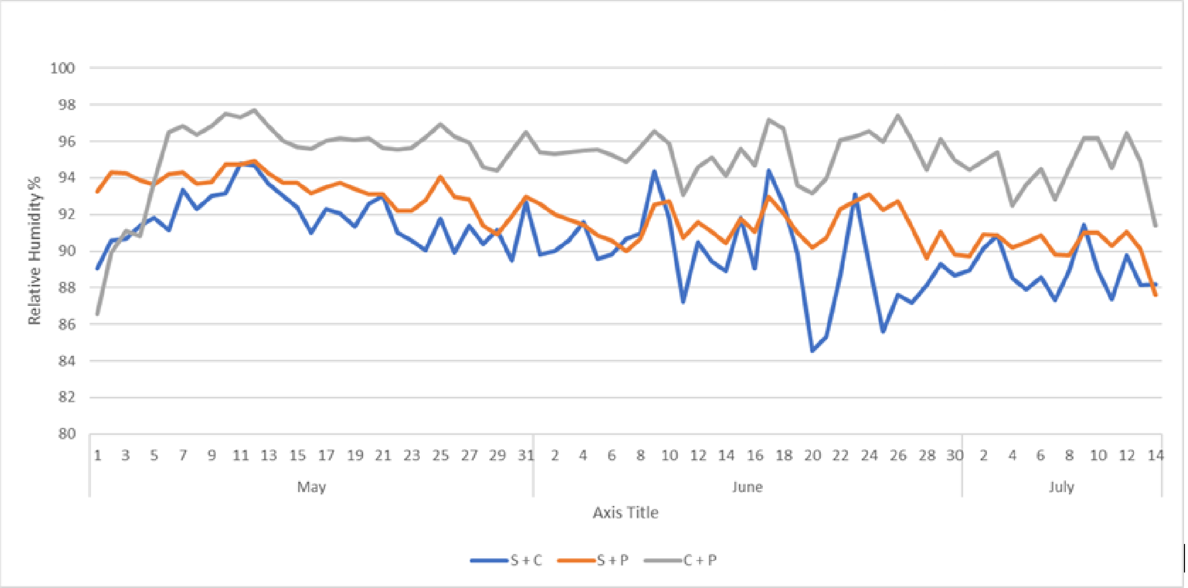


Figure 4.5). The concrete blocks also obstruct light and therefore reduce light under the benches thereby creating a suitable environment for the survival and breeding of slugs which like humid, cool, and shady environment conditions (Growth, n.d.). Concrete block, as a stand or pillars for benches, did not provide a good physical barrier to curtail the vertical movement of slugs from the floor on to the benches with plants. The steel stand barred the gastropod from climbing up into the pots with and this is why benches with steel pillars or stands, the population of slugs were effectively controlled.

The effects of variety in managing slug's population in the greenhouse also had a significant effect on slug population. The Red variety with horizontal leaves (RHL) was the most vulnerable and significantly attracted more slugs on the benches than the remaining two varieties. It is, however, worth noting that with a good greenhouse structure like a concrete floor and benches supported on steel stands, the effect of variety might not be important. Meaning that, irrespective of variety type, Poinsettia grows well in this kind of greenhouses without the problems of slugs and of cause humidity which is a very critical conditions in the management of mollusc and other pests like botrytis.

The inference from the findings is that benches set on steel stands on a concrete floor (S+C) or benches set on steel stands on a pumice floor (S+P) are effective physical barriers in the management of slugs on Poinsettia mother plants grown in Kenya. However, when benches with concrete stands and a pumice floor (C+P) are used, then the effectiveness of slug management diminishes. Of great importance is the type of stand that support benches. Steel narrow stands effectively curtails the vertical mobility of slugs leading to effective management of their population in the crop. In addition to the favourable conditions created for the slugs by the pumice floor, the rock pillars further worsen the stituation by reducing light under the benches and making it more humid due to fertilizer solution obsorbed on the surfaces of the rocks. A study carried out by Port *et al.* (2021) showed that slugs easily recolonized a grassland by moving vertically more than horizontally and therefore physical barriers working against their vertical mobility offer more effective control measures for population build up.

CHAPTER 5:

GENERAL DISCUSSSION, CONCLUSIONS AND RECOMMENDATIONS

5.1 General Discussion

Amblyseius swirskii has attracted a lot of interest as a biocontrol agent for whitifies, and thrips in greenhouse crops. It has been tested and released in many European countries as well as USA, North Africa, China (EPPO, 2013). This study intended to evaluate the effectiveness of using the biocontrol agent in managing whiteflies especially *B. tabaci* in a greenhouse production of Poinsettia in Kenya. It was established that using *A. swirkskii* as the sole insecticide against whitefly was not effective irrespective of the cultivar grown. The efficacy of the *A. swirskii*, got lesser and lesser as time went by from the month of April to July. The low efficacy could have been due to many reasons. First, the mite might have suffered from insufficient supply of food as the crop foliar got thicker and the mites had to cover a bigger area to get whitefly to feed. *A. swirskii* persists longer during low prey density by subsisting on alternative sources of food such as pollen and nectars (Ragusa and Swirski, 1975). But Poinsettia is an obligate short-day plant and therefore if grown to produce vegetative cuttings, then it must be purely vegetative. (Johnson, 2020).

Vegetative Poinsettia stock has no flower to produce pollen or nectar. Insufficient food supply to the predatory mites could have hindered their performance. The cool weather that set in from May to July (Figure 3.) could have negatively affected the activities of the predatory mites. *A. swirskii* does well in warmer and humid subtropical climate and may not perform optimally in a cooler environment and at a reduced relative humidity (Park et al., 2010). Finally, as the plant grew bigger, the foliar got thicker and the mites had a bigger area to cover during hunting this condition coupled with the aforementioned factors could have rendered the predatory mites ineffective in the management whitefly in the poinsettia stock.

The results concur with those of Brownbridge and Buitenhuis (2017) who reported that the use of biocontrol agents as a sole method is rarely successful in pest control, except if it is combined with other methods in an integrated system. This study also confirmed that when *A. swirskii* supplemented with compatible synthetic chemical insecticides was effective against whitefly. In this case, the compatible pesticides were buprofezin and deltamethrin used twice, week 21 and 25, to reduce the whitefly pressure in the greenhouses with this kind of treatment. In fact, the effect of *A. swirskii* combined with the two synthetic insecticide against whitefly was comparable to that of using synthetic chemicals only.

This study also investigated the effect of modifying the greenhouse environment in controlling slugs (gastropods) in Poinsettia crop. The modification of the environment involved creating physical barrier in a greenhouse that curtails smooth movements of slugs and minimizes suitable breeding ground for the slugs. It was established that greenhouses with concrete floors and steel benches supported on steel pillars creates an environment that is hostile for the survival of slugs. This is because the humidity is low, there is no ground for laying eggs and the greenhouse is generally dry. No slug was found in such greenhouses in both seasons. However, concrete floor could be expensive.

The study also established that using steel benches supported on steel pillars on a pumice floor achieved similar results. That is, an environment hostile for slug survival and movement. Slugs find it difficult to climb up the steel pillars which act as a physical barrier to their movement. Greenhouses with benches supported on steel pillars are less humid, drier, and better lit compared to greenhouses with benches supported on concrete blocks. This method is cheaper than concrete floors and yet effective. The findings concur with Capinera (2018) who revealed that differences exist in effectiveness of different physical barriers in controlling slugs.

5.2 Conclusions

The efficacy of biocontrol agents (*A. swirskii*) in management of whiteflies in greenhouse production of Poinsettia is relatively low. The application of *Amblyseius swirskii* as a lone insecticide for controlling whitefly and particularly *Bemisia tabaci* is not effective. However, the predator mite in combination with compatible synthetic chemical insecticides such buprofezin and deltamethrin applied twice at the 21st and 25th week effectively controls whitefly in Poinsettia stock plant growing in a greenhouse in Kenya.

Physical barriers can be used in greenhouse Poinsettia production for the management of slugs. This study concludes that the type of physical barriers used has an effect on slug survival and movement. Using steel stands on pumice floor and steel stands on concrete floor reduces slug infestation in greenhouse Poinsettia whereas using concrete stands on pumice floor does not reduce slug infestation in greenhouse Poinsettia.

5.3 Recommendations

- Given the high regulation on use of chemical pesticides, the study recommends that Poinsettia growers should embrace the use of biocontrol agents (specifically *A. swirskii*) in controlling whiteflies in greenhouse production of Poinsettia with judicious combination of insecticides.
- The study also recommends that when using physical barriers for slug management in greenhouse Poinsettia production, growers should consider using steel benches with steel stands on a pumice or consider using the very steel benches on steel stands on a concrete floor.
- Lastly, the study recommends that other studies be conducted to investigate the efficacy of *A. swirskii* in controlling other pests in Poinsettia production apart from whiteflies.

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