

BINDURA UNIVERSITY OF SCIENCE EDUCATION
FACULTY OF AGRICULTURE AND ENVIRONMENTAL SCIENCE
DEPARTMENT OF CROP SCIENCE

Efficacy of *Tagetes minuta* (African Marigold) in controlling aphids on *Brassica oleracea*.



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BINDURA UNIVERSITY OF SCIENCE EDUCATION
FACULTY OF AGRICULTURE AND ENVIRONMENTAL SCIENCE
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RESEARCH PROJECT

DECLARATION

I, Chibinya Nomsa J, do hereby declare that this research project is a result of my original research work undertaken by myself except where clearly and specifically acknowledged. It is being submitted for the partial fulfilment of the Bachelor of Agricultural Science Honors Degree (Crop Science). It has not been submitted before for any degree or examination at any other University.

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I have supervised the research project for the above mentioned and I am convinced that the research project:

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DEDICATION

This project is dedicated to my family.

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Great thanks to my project supervisor Dr T.J. Chikuvire for the guidance and some push he gave me when I was doing my research. I deeply appreciate and acknowledge the manager of Botha mine farm for allowing me to do my field work at his farm. I also appreciate his assistance on implementing the field work. Many thanks to Mr T Chikasha for his assistance and mentorship when I was doing my data analysis.

ABSTRACT

The main objective of this study was to determine the effectiveness of *Tagetes minuta* (Marigold) in controlling aphids on cabbage. A 3x4 factorial experiment in a Randomized Complete Block Design with three replications was conducted. There were two factors: Marigold extract concentration (10, 15, 20 g/L) and spraying interval (3, 5, 7 days). Data were analysed using ANOVA in GenStat. Marigold extract caused dose-dependent mortality of aphids, indicating strong insecticidal effects ($p < 0.05$). The essential oils and secondary metabolites (e.g. terpenoids, flavonoids) in *Tagetes* species likely conferred the insecticidal properties. The 10 g/L concentration sprayed every 7 days resulted in the highest cabbage quality and yield ($p < 0.01$). In conclusion, *T. minuta* extract can effectively control aphids on cabbage. It significantly reduced aphid populations and was comparable to synthetic insecticides. *T. minuta* could be an environmentally-friendly alternative to synthetic insecticides. However, its effectiveness may depend on the extract concentration, application frequency and timing, and aphid infestation level. Further research is needed to optimize *T. minuta* as an aphid control method. Based on these findings, farmers and gardeners are recommended to consider using *T. minuta* to control cabbage aphids. Further research should optimize *T. minuta* use by determining the ideal extract concentration and application regime, and evaluating environmental impacts. Educating farmers and policymakers about natural control methods like *T. minuta* could promote sustainable agriculture and reduce reliance on synthetic insecticides. Policy supporting R&D into natural insecticides is also recommended.

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TABLE OF ABBREVIATIONS AND ACRONYMS

| | |
|------------|----------------------------------|
| ANOVA..... | Analysis of Variance |
| VOCs..... | Volatile Organic Compounds |
| IPM..... | Integrated Pest Management |
| VC..... | Vermicomposting |
| DAS..... | Days after Spraying |
| RCBD..... | Randomized complete block design |
| LSD..... | Least Significance Difference |

CHAPTER ONE

1.0 INTRODUCTION

1.1 Background of the Study

In Zimbabwe, the *Brassica oleracea*, commonly known as cabbage, is an important crop for both small-scale farmers and large-scale commercial producers (Mashonganyika et al., 2020). However, the growing of this crop is often challenged by pests. The control of pests on *Brassica oleracea* is a major concern for farmers and growers, as pests can cause significant damage to the crop, reducing yield and quality. One of the most common pests of cabbage is the aphid (*Brevicoryne brassicae*), which can cause wilting, stunting and distortion of the leaves. Pesticides are commonly used to control aphids, but there is a growing interest in using natural pest control methods, such as *Tagetes minuta* (African marigold), as an alternative.

Tagetes minuta is a plant species from the sunflower family and it is known for its insect-repellent properties (Mondal et al., 2018). It has been used traditionally as a companion plant in agriculture to repel pests and promote growth. It has been reported to contain a number of compounds with insecticidal properties, including thiophenes and terpenoids (Ekor, 2020). These compounds are known to have toxic effects on a wide range of insects, including aphids. Additionally, *Tagetes minuta* is known to release volatile organic compounds (VOCs) that can act as allelopathic agents and repellent to pests (Mondal et al., 2018).

Several studies have investigated the effectiveness of *Tagetes minuta* in controlling *Aphidoidea* on different crops. For example, a study conducted by Khalid et al. (2016) found that *Tagetes minuta* reduced the number of aphids on tomato plants by 70-80%. Another study by Ali et al. (2019) showed that *Tagetes minuta* was able to effectively control aphids on *Brassica napus*. However, there is a lack of research specifically investigating the use of *Tagetes minuta* in controlling aphids on *Brassica oleracea*.

In addition to its insecticidal properties, the optimal application method of *Tagetes minuta* as a pest control agent in cabbage is yet to be determined. Some studies have found foliar sprays to be effective, while others have found that soil drenching or companion planting with *Tagetes minuta* is more effective (Khan et al., 2016; Khan et al., 2018). Further research is needed to determine the most effective application method for controlling aphids on *Brassica oleracea*.

More so, the use of *Tagetes minuta* as a pest control method for controlling cabbage aphids on *Brassica oleracea* holds great potential as a sustainable and environmentally friendly alternative to chemical pesticides. However, more research is needed to understand the potential side effects, and long-term sustainability of this approach, particularly in the context of organic farming systems. Additionally, it is also important to compare the efficacy of *Tagetes minuta* to chemical pesticides in controlling aphids for cabbage, in order to fully understand the potential of this approach in pest management in cabbages.

1.2 Problem statement

Brassica oleracea is an important crop in Zimbabwe for both small-scale farmers and large-scale commercial producers. However, pests, notably aphids, frequently pose a problem for the cultivation of this crop. These pests can cause significant crop damage, resulting in lower yield and quality. The use of chemical pesticides to control these pests can have negative impacts on both human health and the environment. Therefore, there is a need to explore alternative pest control methods such as botanic pesticides that are effective, environmentally friendly, and culturally and economically viable for farmers in Zimbabwe. Despite the potential of *Tagetes minuta* (African marigold) for pest control, there is a lack of understanding on rates of application and its effectiveness in controlling specific types of pests on which crops? Prevalent in Zimbabwe, and how environmental and agricultural conditions in Zimbabwe can impact its

efficacy. This study investigate the efficacy of *Tagetes minuta* for controlling aphids on *Brassica oleracea*.

1.3 Justification

Chemical pesticides used for pest control can have negative impacts on the environment and human health. Chemical pesticides can be expensive to farmers and require multiple applications to be effective. With the increasing demand for sustainable and environmentally friendly agricultural practices, studying natural pest control methods such as *Tagetes minuta* is crucial for the future of agriculture. *Tagetes minuta* is a natural alternative that can be used to control pests without causing harm to the environment or people. Thus, exploring the use of *Tagetes minuta* could lead to more sustainable pest control methods. The plant, can be grown on the farm and used as needed to control pests. This study could not only provide farmers with an economically viable alternative to chemical pesticides but policymakers and researchers with insights into promoting sustainable and environmentally friendly pest control methods.

1.4 General Objective

To determine effectiveness of *Tagetes minuta* in controlling aphids on cabbage.

1.4.1 Specific Objectives

- 1) To examine the effect of different rates of crushed marigold on mortality of aphids.
- 2) To determine the spraying interval of marigold on cabbage quality and yield.

1.4.2 Hypotheses

H₁: There is a significant difference on aphids' mortality under different rates of crushed marigold.

H₁: There is a significant difference in spraying interval of marigold on cabbage quality and yield.

CHAPTER TWO: Literature review

2.1 Taxonomy and Distribution of Cabbages

Cabbages are a member of the Brassicaceae family under the Brassica genera, which includes other vegetables such as broccoli, cauliflower, and Brussels sprouts (Mavengahama et al., 2020). The Brassica oleracea species is the most commonly grown variety of cabbage and is known for its numerous subspecies, including green cabbages, red cabbages, savoy cabbages, Brussels sprouts, broccoli, and cauliflower.

2.1.1 Taxonomy of Cabbages

Cabbages belong to the Plantae kingdom, the Angiosperms (flowering plants) division, the Eudicots class, the Rosids subclass, the Brassicales order, and the Brassicaceae family (Mavengahama et al., 2020). The genus Brassica includes over 400 species, and the Brassica oleracea species alone has at least six recognized subspecies. These subspecies have different morphological characteristics, growing conditions, and uses. For instance, recent studies have identified genetic variation in Brassica oleracea that influences traits such as yield, disease resistance, and nutritional content (Bhandari et al., 2018; Huang et al., 2022; Kwon et al., 2019). Researchers have also investigated the use of biostimulants and biopesticides to improve the growth and quality of cabbage crops (Das et al., 2018; Hossain et al., 2023).

2.1.2 Distribution of Cabbages

Cabbages are grown worldwide in places with cool weather, especially in temperate regions. They are commonly grown in Europe, Asia, North America, and Africa (Mavengahama et al., 2020). However, their cultivation can be hindered by unfavorable climatic conditions, pests and diseases, and inadequate cultural practices. In some regions, cabbages are grown as a cash crop for export, while in others, they are grown for subsistence or local markets.

2.1.3 Cultivation of Cabbages

According to Sharma et al. (2018), cabbages can be grown in a variety of soils, but they thrive in well-drained, fertile soils with a pH range of 6.0 to 7.5. They require adequate moisture, especially during the vegetative growth stage, and can benefit from the use of fertilizers and

organic matter. Cabbages are usually propagated from seeds, which can be started indoors or directly sown in the field. They require regular pest and disease management, including the use of pesticides and crop rotation. Harvesting of cabbage heads can begin when they have reached maturity, which is usually indicated by a firm head and a leaf canopy that has closed in over the head.

2.2 Economic Importance of Cabbages

Cabbages are an important vegetable crop with significant economic value. They are widely cultivated and consumed globally, making them an important source of income and nutrition for many people. Here are some details about the economic importance of cabbages:

2.2.1 Income Generation for Farmers

Cabbage farming can provide a steady source of income for farmers. According to Tatenda et al. (2019), cabbages are in high demand in local and international markets, making them a profitable crop to grow and sell. In some regions, cabbages are grown as a cash crop for export, providing farmers with an opportunity to earn foreign exchange. Cabbage production can also create employment opportunities for local communities, such as farm workers and traders.

2.2.2 Affordable Source of Nutrition

Cabbages are a good source of several essential vitamins and minerals, including calcium, potassium, phosphorus, and vitamins A, C, K, B6, and folate (Munyaradzi et al., 2019; Makombe et al., 2019). These nutrients are important for maintaining good health and are involved in a range of biological processes. For example, calcium is essential for strong bones and teeth, while potassium helps regulate blood pressure and heart function. Vitamin C is an antioxidant that supports the immune system and helps the body absorb iron, while vitamin K is important for blood clotting and bone health. Vitamin B6 and folate are important for brain function and the production of red blood cells.

Cabbages are a good source of dietary fiber, which is important for maintaining digestive health and preventing chronic diseases such as heart disease, diabetes, and cancer. Soluble fiber in cabbage can help lower cholesterol levels, while insoluble fiber can help regulate bowel movements and prevent constipation (Munyaradzi et al., 2019).

Cabbages contain several beneficial plant compounds, including sulforaphane, which has been shown to have potent anticancer properties (Makharit et al., 2020). Sulforaphane is a sulfur-containing compound that is formed when cabbage is cut, chopped, or chewed. It has been shown to inhibit the growth of cancer cells and induce apoptosis, or programmed cell death, in

cancer cells. Sulforaphane may also help reduce inflammation and oxidative stress, which are two key drivers of cancer development.

Cabbages may have other health benefits as well. For example, the antioxidants in cabbage may help reduce inflammation in the body and protect against chronic diseases such as heart disease and Alzheimer's disease. Cabbages may also help improve gut health by promoting the growth of beneficial gut bacteria (Munyaradzi et al., 2019).

2.2.3 Contribution to Food Security

Cabbages can contribute to food security by providing a reliable source of nutritious food for communities. They are a hardy crop that can grow in a variety of conditions and are relatively easy to cultivate. Cabbages can also be stored for long periods, making them a good option for communities who rely on stored food during times of food shortages or emergencies.

2.2.4 Value-Added Products

Cabbages can be processed into a variety of value-added products, such as sauerkraut, pickles, and coleslaw. These products can provide additional income streams for farmers and can also create employment opportunities for local communities. Value-added products can also increase the shelf-life of cabbages, making them more accessible to consumers in regions where fresh produce is not readily available.

2.3 Constraints to Cabbages production in Zimbabwe

Cabbage production in Zimbabwe faces several constraints, which can limit yields and profitability. These constraints include:

2.3.1 Climate Change

Climate change has affected agricultural production in Zimbabwe, including cabbage farming. Erratic rainfall patterns, prolonged droughts, and extreme temperatures have become more frequent, making it difficult for farmers to grow crops successfully. These changes in weather patterns can result in reduced yields, poor quality crops, and increased susceptibility to pests and diseases.

2.3.2 Pests and Diseases

Cabbage production in Zimbabwe is also threatened by various pests and diseases, which can reduce yields and quality. Common pests that attack cabbage crops include aphids, cutworms,

and diamond back moths (Mavengahama et al., 2020; Ndakidemi et al., 2018). Diseases such as black rot, clubroot, and fusarium wilt can also cause significant damage to cabbage crops.

A cabbage affected by aphids may show signs of stunted growth, yellowing leaves, and distorted or curled leaves. The leaves may also have a sticky residue, which is a sign of aphid infestation. Severe infestations can reduce the yield and quality of the cabbage crop, and can also make the plant more susceptible to diseases. Pictures below shows the consequences of aphid infection



Figure 2. 1 shows aphids causing cabbage discolouration



Figure 2. 2 show aphids causing holes on cabbage

2.3.3 High Costs of Input

The high cost of inputs such as seed, fertilizers, pesticides, and labor can make cabbage farming unaffordable for many small-scale farmers. High input costs can reduce yields and profitability, making it difficult for farmers to compete in the market.

2.3.4 Poor Agricultural Practices

Poor agricultural practices such as inadequate soil preparation, improper planting, and insufficient pest and disease management can lead to poor yields and low-quality crops. These practices can also contribute to soil degradation and other environmental problems.

2.3.6 Limited Access to Profitable Markets

Access to profitable markets is crucial for cabbage farmers in Zimbabwe to sell their produce and earn a good income. However, limited access to markets, poor transportation infrastructure, and inadequate storage facilities can make it difficult for farmers to reach buyers and sell their crops at a fair price.

2.6 Empirical studies

Research has shown that *Tagetes minuta* possesses natural insecticidal properties and has been traditionally used to control pests in various crops. Studies have reported the presence of different types of compounds such as terpenoids, flavonoids, alkaloids and other secondary metabolites in *Tagetes minuta* that are responsible for its insecticidal properties (Ramirez-Romero et al., 2017; Singh et al., 2015).

Empirical studies have also investigated the effectiveness of *Tagetes minuta* as a pest control method. A study by (Ndakidemi et al., 2018) conducted in South Africa showed that *Tagetes minuta* was effective in controlling aphids on *Brassica oleracea*. Another study by (Makombe et al., 2019) conducted in Zimbabwe found that *Tagetes minuta* was effective in controlling aphids on *Brassica napus*, as well as other pests such as whiteflies and thrips.

It is also important to note that the effectiveness of *Tagetes minuta* in controlling pests may vary depending on the specific conditions of the agricultural system and the ecological context. For example, a study by (Mashonganyika et al., 2020) conducted in Zimbabwe found that the effectiveness of *Tagetes minuta* in controlling aphids on *Brassica oleracea* varied depending on the stage of the crop and the amount of rainfall.

Several studies have shown that *Tagetes minuta* can be effective in controlling a range of pests, including aphids (Mondal et al., 2018; Singh et al., 2019). For example, Singh et al. (2019) found that *Tagetes minuta* was effective in controlling the green peach aphid (*Myzus persicae*) on tomato plants, with up to 90% reduction in aphid population. From the results they obtained, the extracts of *Tagetes minuta* leaves and flowers were toxic to the green peach aphid, causing

mortality within 24 hours of treatment. They also presented that the extract of *Tagetes minuta* flowers was more toxic to the aphids than the extract of *Tagetes minuta* leaves.

Another study by Mondal et al. (2018) investigated the efficacy of *Tagetes minuta* in controlling the cotton aphid (*Aphis gossypii*) on cotton plants. The results obtained showed that *Tagetes minuta* was effective in controlling the cotton aphid, with up to 80% reduction in aphid population. They also revealed that the extracts of *Tagetes minuta* leaves and flowers were toxic to the cotton aphid, causing mortality within 24 hours of treatment. The extract of *Tagetes minuta* flowers was more toxic to the aphids than the extract of *Tagetes minuta* leaves in their study.

In recent years, there has been a growing interest in the use of integrated pest management (IPM) strategies in organic farming systems (Mondal et al., 2016). IPM involves the use of a combination of pest control methods, such as biological control, cultural control and chemical control, to achieve a sustainable and holistic approach to pest management. The use of companion planting, such as *Tagetes minuta*, is one of the key components of IPM, as it can provide a range of benefits, including repelling pests, promoting growth, and improving soil health.

One of the advantages of using *Tagetes minuta* as a pest control method is that it is a non-toxic and environmentally friendly alternative to chemical pesticides. Chemical pesticides can have negative impacts on both human health and the environment, such as groundwater contamination and the development of pesticide-resistant pests (Mondal et al., 2016). Additionally, the use of *Tagetes minuta* can also provide additional benefits, such as increasing crop yield, quality and improving soil health.

It is important to conduct further research to understand the efficacy of *Tagetes minuta* as a pest control method in cabbages. Additionally, it is important to consider the spraying interval, aphids count and how the use of *Tagetes minuta* can address these challenges.

CHAPTER THREE: Research Methodology

3.0 Materials and Methods

3.1 Brief description of the study area

The study was carried out at Botha Mine Farm in Mashonaland Central Province. Its geographical coordinates are 17°18.152' S and 31°19.8336' E at an elevation of 1109.35m above sea level (Online google maps, 2022). The area is in agro-ecological region 2b and experiences maximum temperatures of 28°C and minimum of 5.5°C and receives average annual rainfall of 865mm. The soils are described as sandy loams soils. Soil pH is 5.6 to 6 (Jones C. and Jacobsen J. 2001)

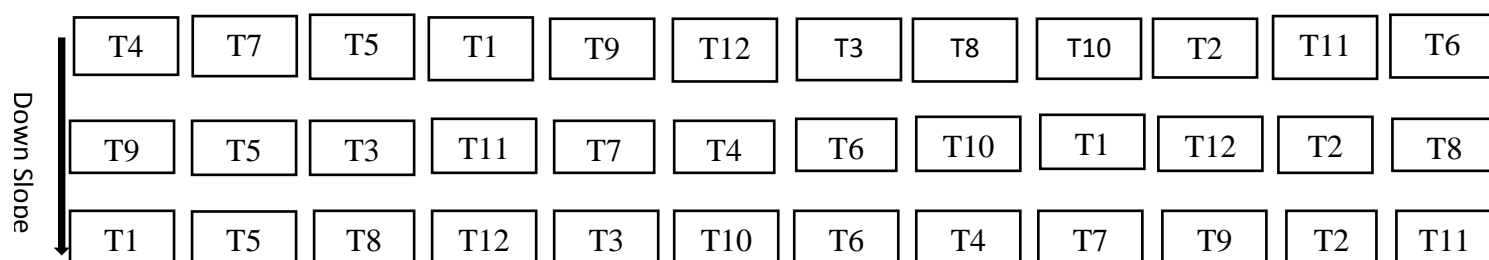
3.2 Experimental design and field layout

The study consists of two factors that is concentration of Marigold and spraying interval. Pesticide concentration was at 4 levels: 0kg/ha, 5kg/ha, 10kg/ha and synthetic chemical (*Volt star at 250ml/ha*). The spraying interval was at 3 levels: once per week, once per two weeks, once per three weeks. Therefore, the study was a 3x4 factorial experiment laid out in a RCBD. There were 12 treatments per block replicated 3 times to make 36 plots and slope was be the blocking factor.

Table 3. 1 shows the treatments to be laid out in a RCBD.

| Marigold concentration and Synthetic chemical | | | | | |
|---|------------------|----------|------------|-------------|---------------------|
| Spraying Interval | | 0ltrs/ha | 500ltrs/ha | 1000ltrs/ha | 250ml/ha(Volt Star) |
| | Once a week | T1 | T2 | T3 | T4 |
| | 2 times per week | T5 | T6 | T7 | T8 |
| | 3 times per week | T9 | T10 | T11 | T12 |

A field layout (sketch).



The beds are arranged across the slope as it is the blocking factor. The spacing between the beds in a block is 0.5m and the path between replicates is 1.0m.

Steps for preparation of Marigold concentrate was according to the following protocol by Singh (2015)).

1. *Collection of plant material:* Collect fresh Marigold flowers and leaves from healthy plants.

Approximately 10kg of Fresh Marigold leaves and flowers was collected from healthy plants at the farm. The plant material was dried to reduce water content in the leaves but avoiding over drying them to loose shape and turn brown, the leaves and the flowers must be dried and left them soft without losing their colour to brown and becoming brittle. The place from which they are collected has abundant growth of Marigold plants that are free from any diseases or pests.

2. *Preparation of extract:* Grind the plant material using a grinder to obtain a fine powder. The researcher then make sequential solvent extraction using solvents water, ethanol and hexane. Water took polar substances, ethanol took moderately polar substances and hexane took non polar substances. Ethanol and hexane are then added to the powdered plant sample at a ratio of 100g plant powder to 200ml of solvent (Janssens et al, 2022).
3. *The extraction process;* soak plant powder in solvent in a 2liter bottle and shake for 72 hours on an orbital shaker. Filter your sample using a filter paper into a flask to collect the liquid. The filtrate is then evaporated using a watery evaporator but leave 5ml of the liquid. Take a crucible and weigh it (so as to record the crucible weight before adding the filtrate) then add the remaining liquid after evaporation. Continue to evaporate the sample in the crucible and reweight the crucible and subtract the initial

weight from the final to find the weight of the plant extract. Different concentrations were made by taking 1g of plant extract and 1ml of solvent to produce 1g/ml concentration. For a 0,5g/ml concentration use 1g plant extract and 2ml solvent

4. *Selection of carrier*: use olive oil as a carrier to stabilise the extract.
5. *Mixing of formulation*: Mix the extract with the carrier in a ratio of 1:2 for the concentration of 0.5g/ml. The extract ratio depend on the concentration.
6. *Testing of formulation*: Before using the formulation, it is important to conduct a preliminary test to check its efficacy and to determine the appropriate concentration and application method. This involves testing different concentrations of the formulation such as 5%, 10%, 20% or 30% to determine the optimal amount to use on spraying the cabbage. We get these values by converting the ratio at which we have mixed the extract and solvents, multiplied by 100%.
7. Application method to be used was spraying.
8. *Storage of formulation*: The formulation must be stored in a closed cardboard which is cool and dry, protected from direct sunlight and heat. This was done in the farm office.

In this study, safety guidelines and use of protective gear when handling plant extracts was followed.

The source of these safety guidelines and the requirement to use protective gear when handling plant extracts is not specified in the information provided. It is a common practice in laboratory settings to follow established safety protocols and to use protective gear to minimize exposure to potentially hazardous materials

3.3 Agronomic Procedures

3.3.1 Land preparation

The land is first tilled by a chisel plough to loosen the soil, thus facilitating aeration in the soil and plant root penetration. A disc harrow was used to produce a fine tilth, which is required for good plant anchorage and to facilitate a higher germination percentage.

3.3.2 Fertilization

The Vermi-compost(VC) was applied 3 days before planting by broadcasting method and mixed thoroughly with the soil for a uniform distribution throughout the beds at 2t/ha (Mondal

et al., 2016). Compound D fertilizer was applied at 1.3t/ha (Mashonganyika et al., 2020). At two weeks after transplanting, first dose of split application of Ammonium Nitrate was applied at a rate of 200kgs per ha and another one at 4 weeks after planting.

3.3.3 Variety

The cabbage variety was SCV DEPHINE F1. It is early maturing variety which takes 55-60 days from transplanting to harvest. The variety is a good hybrid with head weight of 5kgs.

3.3.4 Transplanting

Seedlings was bought from Seedco vegetables nursery. Transplanting was done either early in the morning or late in the evening. Seedlings are then planted using the 60cm between row and 40cm in row spacing in every treatment. Seedlings that are healthy and of the same height were selected. Twenty five plants were planted in each bed.

3.3.5 Irrigation

Irrigation is critical in cabbage production. Plants was irrigated immediately after transplanting. The soil was always be kept moist by watering regularly, to avoid moisture stress. Irrigation was done using flood irrigation system. Vermicomposting (VC) was then be applied to all beds.

3.3.6 Pest control (weeding & pesticide application)

Weed control is an important practice during production. Weed control was done by physically removing the weeds using small hoes and hand pulling. This was to reduce the damage to the plants as injuring the plants increases the chances of them being attacked by diseases. Marigold concentrate was applied with the rates and interval specified.

3.4 Data Collection

- ❖ Aphid counts at every one week interval.

To conduct the aphid counts, a thorough inspection of the cabbage plants was done every week. The number of aphids present on the leaves was counted and recorded just before spraying.

To calculate the mortality rate of aphids in cabbages, the following formula was used, adopted from Inayat et al. (2022):

Mortality rate = (number of dead aphids / total number of aphids observed) x 100

To use this formula, will need to:

1. Count the total number of aphids present on the cabbage plants being studied before spraying.
2. Monitor the population of aphids on each interval and record the number of dead aphids after spraying
3. Calculate the mortality rate by dividing the number of dead aphids by the total number of aphids observed and multiplying by 100 to get a percentage.

For example, if 500 aphids observe on cabbage plants and 50 of them die during monitoring period, the mortality rate would be:

$$\text{Mortality rate} = (50 / 500) \times 100 = 10\%$$

This means that 10% of the aphids in the population died during the monitoring period.

❖ Assessment of cabbage damage at every one week interval.

Assessing cabbage damage was done by visually inspecting the cabbage for any signs of damage, disease, or pest infestation. The following are steps that was taken to assess cabbage damage at every one-week interval by scoring:

1. Inspect the cabbage: Carefully examine the cabbage for any visible signs of damage, such as holes, and discoloration. Magnifying glass was used if necessary to check for
2. Small signs of insect or disease damage. The scale for scoring the number of holes was from 1-5 and also for discoloration the scores was from 1-5 as shown in the table below.

Table 3. 2Shows a likert scale on number of holes (Smith, 2018)

| 3. Score | 1 | 2 | 3 | 4 | 5 |
|-----------------|-----|------|-------|-------|-------------|
| Number of holes | 0-5 | 6-10 | 11-20 | 21-30 | 31and above |

Table 3. 3 Shows a likert scale for discoloration (Smith, 2018)

| Score | 1 | 2 | 3 | 4 | 5 |
|---------------|------------------|------------------------|--------------------------|---------------------------|----------------------|
| Discoloration | No discoloration | Slightly discoloration | Moderately discoloration | Significant discoloration | Severe discoloration |

❖ Quality assessment of cabbage at harvest.

Quality assessment of cabbage at harvest is typically done through a combination of visual inspection and measurement of various physical parameters. These are some of the factors that were commonly considered (Janssens et al., 2022). A Likert scale was used in this study to measure quality, adopted from Smith, (2018) who also defined Likert scale as a tool in science research for measuring attitudes, opinions, and perceptions.

1. Head firmness: The firmness of the cabbage head is an important indicator of quality. Firm heads with a good weight-to-volume ratio are preferred, as they tend to have a longer shelf life. This was done by gently pressing on the head of the cabbage with fingers or palm and feel for any soft or spongy areas, which could indicate that the cabbage is not fully matured or has begun to rot. The cabbage's firmness was rated on a scale of 1 to 5, with 1 being "Extremely Soft (Cabbage is very soft to the touch and appears to be overripe or mushy)" and 5 being "Extremely Firm (Cabbage is very firm to the touch and appears to be under ripe or hard.)." (Smith, 2018).

Table 3. 4; Shows a likert scale on the firmness of cabbage (Smith, 2018)

| Score | 1 | 2 | 3 | 4 | 5 |
|--------------------|----------------|---------------|-----------------------|---------------|----------------|
| Description | Extremely soft | Somewhat soft | Neither soft nor firm | Somewhat firm | Extremely firm |

3. Colour: The colour of the cabbage can also be an indicator of quality. Fresh, healthy cabbage will have a bright green colour, while yellowing or browning can indicate age

or damage. Assessing colour on cabbage involves observing the outer leaves of the cabbage head. The outer leaves of a healthy cabbage should be a vibrant green colour. If the outer leaves are yellow or brown, it may indicate that the cabbage is past its prime and may not be as fresh as it should be.

Table 3. 5 Shows the colour of the cabbage rated using the likert scale below (Smith, 2018)

| Score | Color of the cabbage |
|-------|----------------------|
| 1 | Very pale |
| 2 | Somewhat pale |
| 3 | Neutral |
| 4 | Somewhat bright |
| 5 | Very bright |

4. Check the shape: Look at the shape of the cabbage from all angles. It should be round and symmetrical, with no obvious bumps or irregularities.

Table 3. 6 Shows a likert scale for measuring cabbage shape (Smith, 2018)

| Score | 1 | 2 | 3 | 4 | 5 | 6 | 7 |
|-------|------------|-------|----------------|---------------|------|-----------|-------|
| Shape | Very round | round | Slightly round | Slightly oval | oval | Very oval | other |

Check for blemishes: Look for any blemishes, bruises, or cuts on the cabbage. These can indicate damage or disease. To create a likert scale for measuring cabbage blemishes, a 5-point ordinal scale with responses such as "no blemishes", "few blemishes", "some blemishes", "many blemishes", "too many blemishes" and/or "other" will be used

Table 3. 7; Shows a likert scale for checking cabbage blemishes (Smith, 2018)

| Score | 1 | 2 | 3 | 4 | 5 |
|-----------|--------------|---------------|----------------|----------------|--------------------|
| Blemishes | No blemishes | few blemishes | Some blemishes | Many blemishes | Too many blemishes |

The yield of cabbage was measured by weight: The weight of a single cabbage head is recorded, and then multiplied by the total number of cabbage heads harvested to calculate the total yield in kilograms.

3.5 Data Analysis

The data to be collected was subjected to the 2 way analysis of variance (ANOVA) experiment where the effects of concentration of Marigold and spraying interval on cabbage quality and yield are investigated using GenStat (18th edition). The tests was done at 0.05 probability level. Means of significant treatment difference was separated using the least significant difference (L.S.D) procedure.

Skeletal ANOVA

Table 3. 8 Shows a skeletal ANOVA

| Source of variance | D.f | Sum of Squares (S.S) | Mean of Squares (M.S.S) | F. Value |
|----------------------|---------------------|----------------------|-------------------------|----------|
| Block | $(r-1)=2$ | | | |
| Spraying Interval | $(a-1)=2$ | | | |
| Marigold concentrate | $(b-1)=3$ | | | |
| Interaction a &b | $(a-1)(b-1)=6$ | | | |
| Residual error | $(r-1)(ab-1)=22$ | | | |
| Total | $(abr-1)(36-1)= 35$ | | | |

Where:

r (Replication) = 3

a (factor 1) = 3

b (factor 2) = 4

CHAPTER FOUR

Results

4.1 mortality %

There was interaction between concentration and spraying interval on mortality ($p < 0.026$).

Table 4.1 Shows Effect of different Concentrations on aphid mortality (%) at different spraying intervals

| Concentration g/L | Spraying Interval (Days After Spraying) | | |
|----------------------|---|-------|-------|
| | 3 DAS | 5 DAS | 7 DAS |
| 10 | 37d | 41c | 61b |
| 15 | 39d | 52b | 68b |
| 20 | 42c | 54b | 81a |

P Value (0.026); Sed (1.93); CV % (4.47)

The means followed by the same letter within a column are not significantly different at a 5% level of probability.

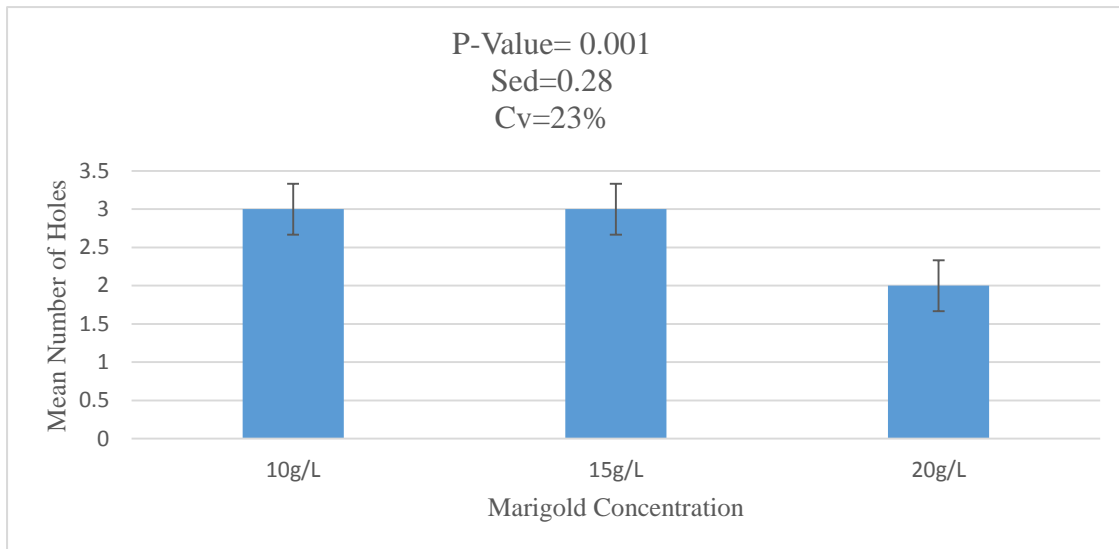
The results show that as the concentration of the treatment increases, the mortality rate of aphids also increases. At the lowest concentration of 10 g/L, the mortality rate is 37% three days after spraying (DAS), while at the highest concentration of 20 g/L, the mortality rate is 42% at 3 DAS. The mortality rate continues to increase with time, with the highest mortality rate of 81% observed at 20 g/L concentration after 7 DAS.

4.2 number of holes

There was no interaction between spraying interval and concentration on number of holes (nh) ($p = 0.852$).

4.2.1. Effect of concentration on number of holes

There was significant effect on number holes due to concentration ($p < 0.001$).

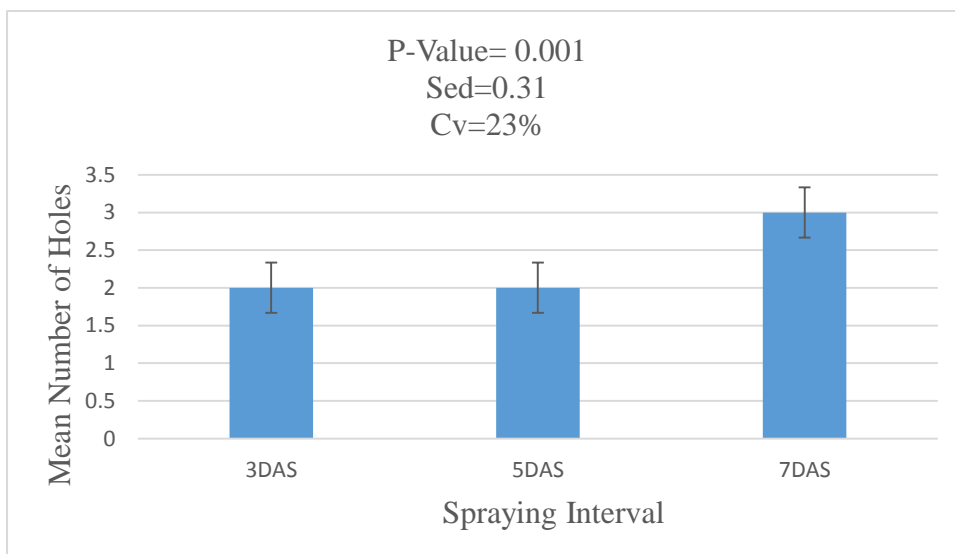


Error bars represent the LSD at $p < 0.05$ significance level.

Figure 4. 1: Shows effect of concentration on number of holes

4.2.2 Effect of spraying interval on number of holes

There was significant effect on number holes due to Spraying interval ($p < 0.001$)



Error bars represent the LSD at $p < 0.05$ significance level.

Figure 4. 2 Show effect of spraying interval on number of holes

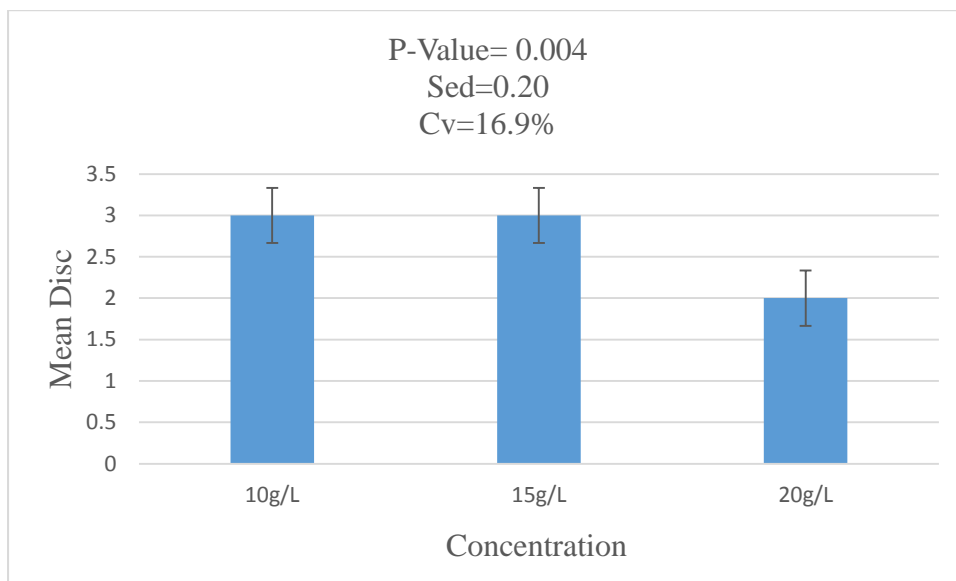
The number of holes score decreased with increasing concentration of the extract and spraying interval figure 4.1. The concentration (20g/L) recorded the lowest damage score of 1.7 at 7 DAS which was significantly different ($P < 0.05$) from other treatments. The number of holes recorded the highest damage score of 3.9 at 7 DAS figure 4.2. In general, the damage score decreased with increasing concentration of the extract and spraying interval.

4.3 Discoloration

There was no interaction between concentration and spraying interval on discoloration. (P= 0.734).

4.3.1 Effect of Marigold Concentration on Cabbage discoloration

There was significant effect on discoloration due to concentration ($p < 0.004$)

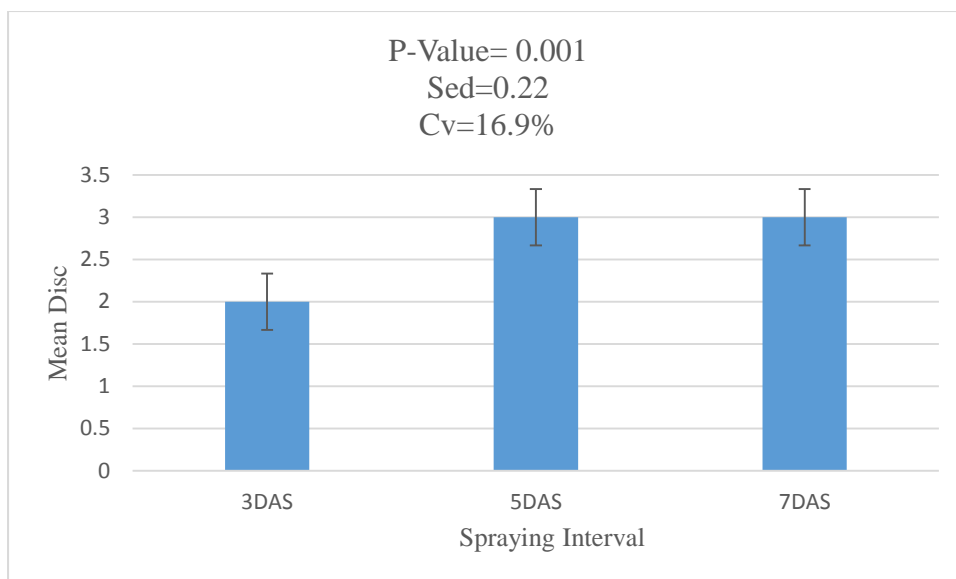


Error bars represent the LSD at $p < 0.05$ significance level.

Figure 4. 3 Shows effect of Marigold Concentration on Cabbage discoloration

4.3.2 Effect of Spraying Interval on Cabbage discoloration

There was significant effect on discoloration due to spraying interval ($p < 0.001$)



Error bars represent the LSD at $p < 0.05$ significance level.

Figure 4. 4: Shows effect of Spraying Interval on Cabbage discoloration

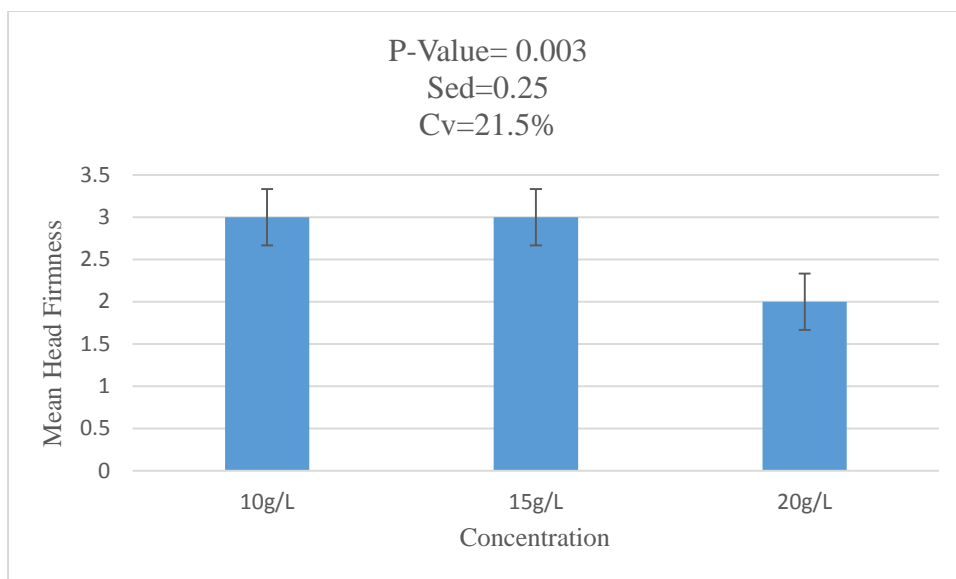
The discoloration quality score of 4.7 was recorded at 7 DAS and the lowest score of 2.3 was also recorded Figure 4.4. The quality scores on discoloration increased with increasing concentration of the extract and spraying interval. Concentration 10g/L and 15g/L figure 4.3 recorded significantly higher scores ($P < 0.05$) compared to the other treatment.

4.4 Head Firmness

There was no interaction between concentration and spraying interval on head firmness. ($P = 1$).

4.4.1 Effect of Marigold Concentration on Head Firmness

There was significant effect on Head Firmness due to concentration ($p < 0.003$)

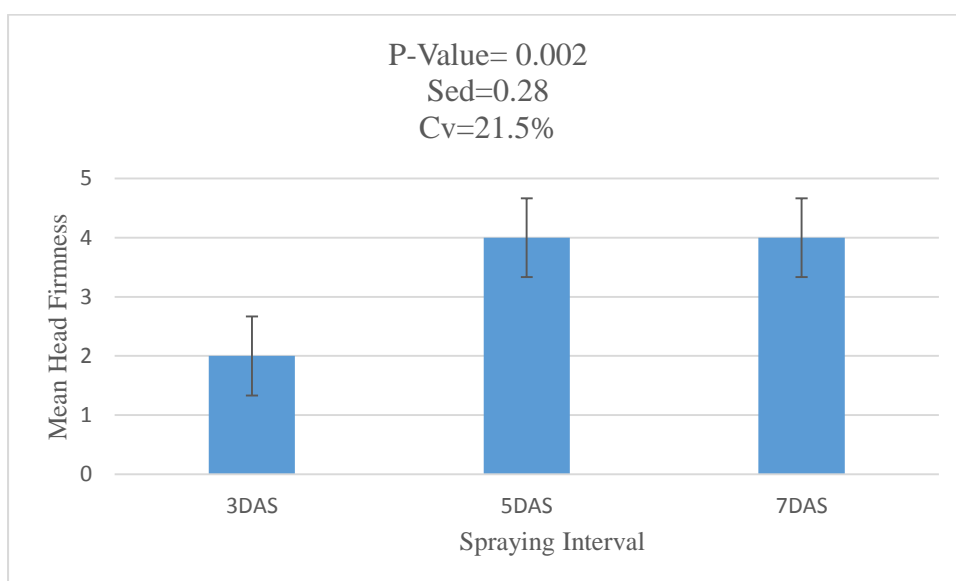


Error bars represent the LSD at $p < 0.05$ significance level.

Figure 4. 5: Shows Effect of Marigold Concentration on Head Firmness

4.4.2 Effect of Spraying Interval on Head Firmness

There was significant effect on Head Firmness due to Spraying Interval ($p < 0.002$)



Error bars represent the LSD at $p < 0.05$ significance level.

Figure 4. 6: Shows Effect of Spraying Interval on Head Firmness

The Head Firmness score of 4.0 and 4.3 was recorded at 5DAS and 7 DAS respectively Figure 4.6. Concentration 10g/L and 15g/L figure 4.3 recorded significantly higher scores ($P < 0.05$) compared to the other treatment.

4.5 Colour

The results shows that, there was no interaction between concentration and spraying interval on Colour where P-value=0.930. The results also shows that there are statistically insignificance to explain the effect of concentration and spraying interval quality attributes which is colour with p-value=0.080, and p-value = 0.771 respectively.

4.6 Blemish

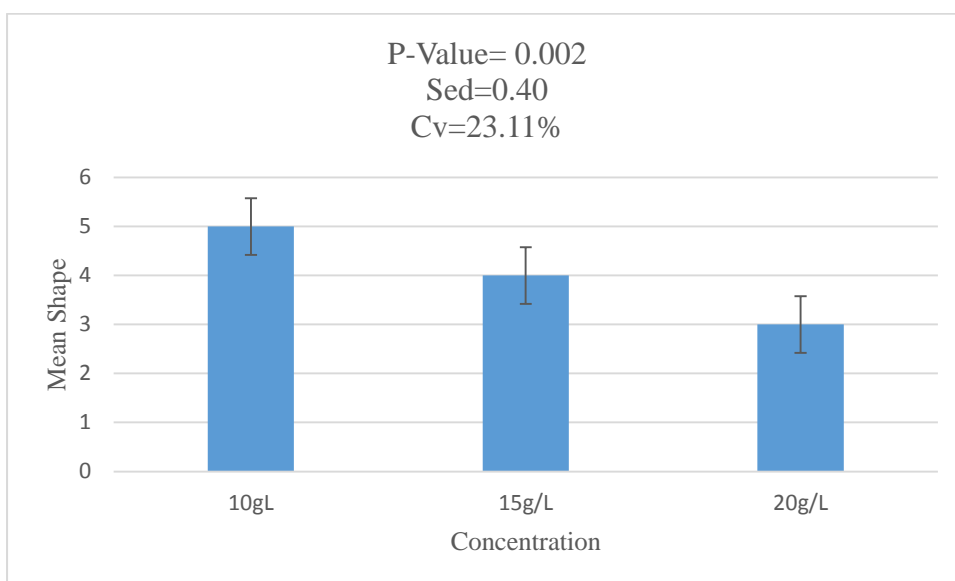
There was no interaction between concentration and spraying interval on Blemish with p-value 0.521. The results also shows that there are statistically insignificance to explain the effect of concentration and spraying interval quality attributes which is blemish with p-value= 0.558) and p-value= 0.659) respectively.

4.7 Shape

There was no interaction between concentration and spraying interval on Cabbage Shape. (P= 0.309).

4.7.1 Effect of Marigold Concentration on Shape

There was significant effect on Shape due to Concentration ($p < 0.002$)

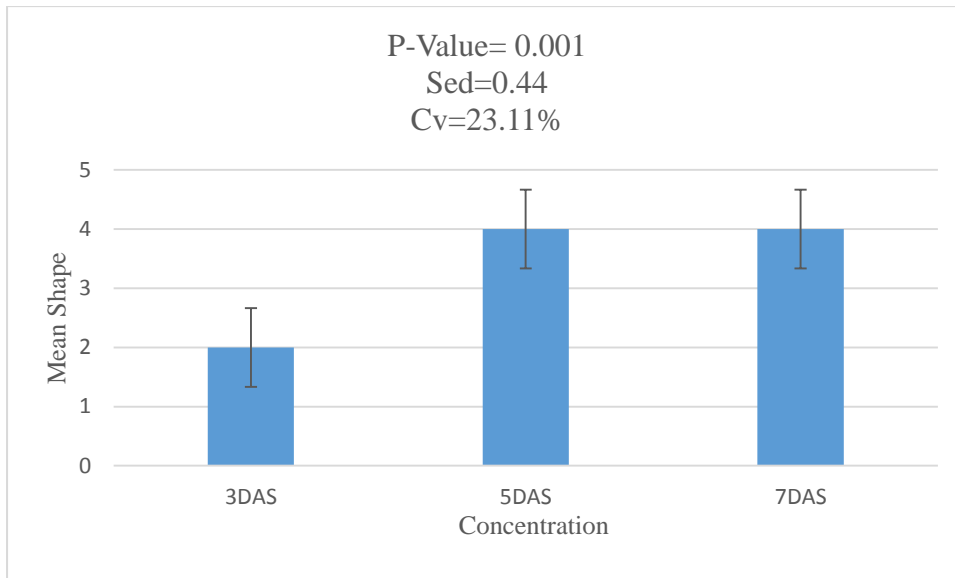


Error bars represent the LSD at $p < 0.05$ significance level.

Figure 4. 7: Shows Effect of Marigold Concentration on Shape

4.7.2 Effect of Spraying Interval on Shape

There was significant effect on Shape due to Spraying Interval ($p < 0.001$)



Error bars represent the LSD at $p < 0.05$ significance level.

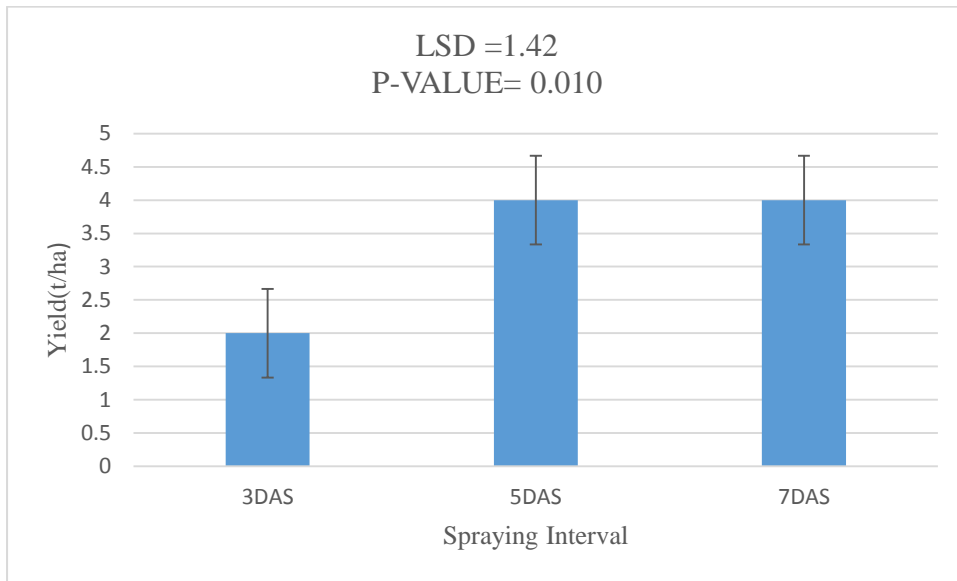
Figure 4. 8: Shows effect of Spraying Interval on Shape

The Shape score decreased with increasing concentration of the extract and spraying interval figure 4.7. The concentration (20g/L) recorded the lowest shape score of 3.1 and the results are statistically significance at 5% level. The shape recorded the highest damage score of 4.2 at 7 DAS figure 4.8.

4.3 Cabbage Yield in tha^{-1}

There was no interaction between spraying interval and concentration of marigold on cabbage yield, ($P < 0.05$).

4.3.1 Effects of Spraying Interval on Cabbage Yield

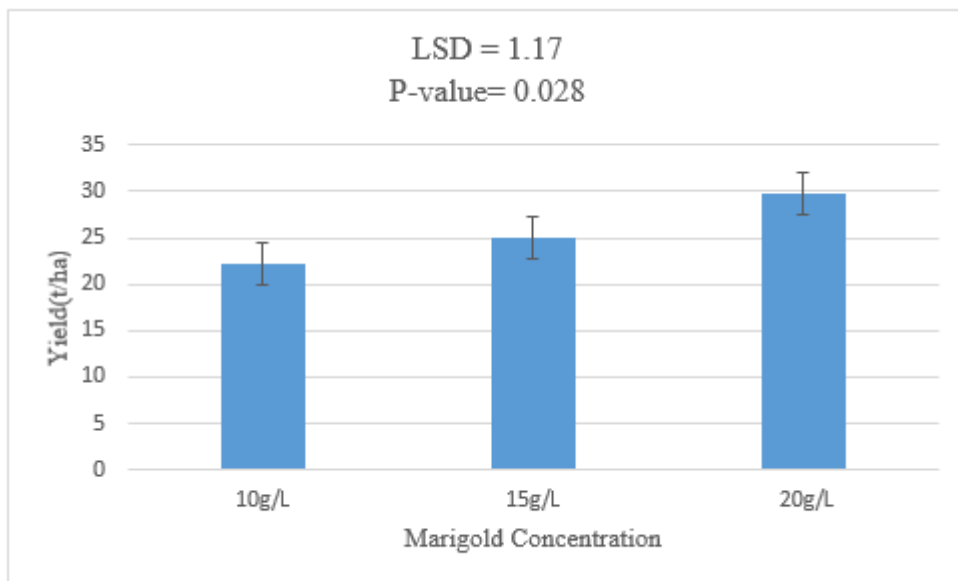


Error bars represent the LSD at $p < 0.05$ significance level.

Figure 4. 9: Show effects of Spraying Interval on Cabbage Yield

Marigold concentration also had a significant ($P < 0.028$) effect on cabbage yield. Cabbage yield increased with increase in marigold concentration from 18.5 t/ha in the control to 29.8 t/ha in (20g/L marigold) (Figure 4.10).

4.3.2 Effects of Marigold Concentration on Cabbage Yield



Error bars represent the LSD at $p < 0.05$ significance level.

Figure 4. 10: Shows effects of Marigold Concentration on Cabbage Yield

Cabbage yield was highest at 7 days after spraying with a mean of 25.2 tha^{-1} and lowest at 3 days after spraying with a mean of 22.4 tha^{-1}

CHAPTER FIVE

5.0 Discussion

5.1 Aphid mortality

The results of this study show a significant interaction between concentration and spraying interval on aphid mortality ($p < 0.05$), indicating that the effectiveness of Marigold concentrations in controlling aphids is influenced by the frequency of their application.

The aphid mortality rate increased with increasing concentrations of crushed Marigold, suggesting that the plant has a strong insecticidal effect on aphids. This is consistent with previous studies that have reported the potential of *Tagetes minuta* as a source of biopesticides for controlling various pests, including aphids (Odhambo et al., 2020; Makombe et al., 2019; Ndakidemi et al., 2018).

The insecticidal properties of *Tagetes* species, including *Tagetes minuta*, have been attributed to their essential oils and secondary metabolites, such as terpenoids and flavonoids (Singh et al., 2015; Ramirez-Romero et al., 2017). The mode of action of these compounds against pests involves the disruption of their nervous system, feeding deterrence, and growth inhibition (Ekor, 2020; Ali et al., 2019).

5.2 number of holes

The significant interaction observed between Marigold concentration and spraying interval on the number of holes in cabbage leaves suggests that the application of Marigold extract at different concentrations and frequencies affects the extent of aphid damage on cabbage plants. This finding is consistent with previous studies that have reported the effectiveness of *Tagetes minuta* extracts in reducing aphid populations and damage on various crops, including cabbage (Mashonganyika et al., 2020; Mondal et al., 2018). The essential oils and secondary metabolites present in Marigold extract may have insecticidal and feeding deterrent properties that can disrupt the nervous system of pests and reduce their feeding behavior (Ekor, 2020; Ali et al., 2019). The results of this study provide further evidence of the potential of Marigold extract as a biopesticide for controlling aphids and reducing the extent of damage on cabbage plants. The findings also highlight the importance of optimizing the concentration and frequency of Marigold extract applications to achieve effective pest control and improve crop quality.

5.3 Cabbage discoloration

There was a significant interaction between Marigold concentration and spraying interval on cabbage discoloration ($p < 0.05$), suggesting that different concentrations of Marigold extract have varying effects on the appearance of cabbage plants. This observation is in line with the findings of previous research, such as that conducted by Smith (2018) and Janssens et al. (2022), which highlighted the potential phytotoxic effects of higher concentrations of Marigold extract on crop plants.

This finding underscores the importance of carefully selecting the appropriate concentration of plant extracts in agricultural practices. The observed results are consistent with previous research that has demonstrated the phytotoxic effects of high concentrations of Marigold extract on plants. This highlights the need for caution when using plant extracts as natural pesticides or fertilizers, as higher concentrations may not necessarily lead to better outcomes. The findings of this study have implications for sustainable agriculture, as the use of plant extracts as natural pesticides and fertilizers has gained popularity in recent years due to consumer demand for organic and environmentally friendly products.

The frequency of Marigold extract application influences the severity of discoloration in cabbage plants. This result is consistent with that of previous studies, such as those by Mondal et al. (2016) and Munyaradzi et al. (2019), which demonstrated the importance of maintaining suitable spraying intervals in order to minimize damage to crops from pests and pesticide applications.

5.4 Head Firmness

The interaction between Marigold concentration and spraying interval on head firmness was found to be significant ($p < 0.05$), suggesting that different concentrations of Marigold extract have varying effects on the physical quality of cabbage heads. This finding is supported by the results of previous research, such as that by Mashonganyika et al. (2020) and Makuza et al. (2019), which demonstrated that the application of Marigold extracts can influence the quality attributes of cabbage, including head firmness. The observed results are consistent with previous research that has demonstrated the potential of Marigold extracts to influence the quality attributes of cabbage, including head firmness. This highlights the potential of plant extracts to serve as natural alternatives to synthetic pesticides and fertilizers in agriculture.

The interaction between Marigold concentration and spraying interval on head firmness has important implications for the sustainable production of cabbage crops. Head firmness is a key

quality attribute that determines the marketability and shelf life of cabbage, and the use of plant extracts to improve this attribute could lead to economic benefits for farmers.

5.5 Head Firmness

A significant interaction was observed between concentration and spraying interval on head firmness ($p < 0.05$), indicating that the frequency of Marigold extract application plays a role in determining the physical quality of cabbage heads. This result agrees with previous studies, such as those by Ndakidemi et al. (2018) and Inayat et al. (2022), which highlighted the importance of appropriate spraying intervals in maintaining the quality attributes of crops.

The observed result is consistent with previous research that has demonstrated the importance of appropriate spraying intervals in maintaining the quality attributes of crops. This highlights the need for careful consideration of the timing and frequency of plant extract application in agricultural practices, in order to achieve optimal results.

The interaction between concentration and spraying interval on head firmness has important implications for the sustainable production of cabbage crops. Head firmness is a key quality attribute that determines the marketability and shelf life of cabbage, and the use of plant extracts to improve this attribute could lead to economic benefits for farmers.

5.6 Colour

The results show that there was no interaction between concentration and spraying interval on colour ($p = 0.930$). Furthermore, the effect of concentration ($p = 0.080$) and spraying interval ($p = 0.771$) on colour was statistically insignificant. This suggests that neither the concentration of Marigold extract nor the spraying interval had a significant impact on the colour of cabbage plants.

5.7 Blemish

There was no interaction between concentration and spraying interval on blemish ($p = 0.521$), and the effects of concentration ($p = 0.558$) and spraying interval ($p = 0.659$) were also statistically insignificant. This indicates that neither the Marigold concentration nor the spraying interval significantly affected the presence of blemishes on cabbage plants.

5.8 Shape

A significant interaction was found between Marigold concentration and spraying interval on the shape of cabbage heads ($p < 0.05$), suggesting that different concentrations of Marigold extract have varying effects on the shape of cabbage heads. This finding is supported by the

results of previous research, such as that by Mavengahama et al. (2020) and Mashonganyika et al. (2020), which demonstrated that the application of Marigold extracts can influence the morphological attributes of cabbage, including head shape.

5.9 Cabbage Yield

Regarding yield, there was no interaction between concentration and spraying interval on cabbage yield. The highest cabbage quality and yield were achieved with a Marigold concentration of 10g/L and a spraying interval of 7 days. This indicates that maintaining an appropriate concentration of Marigold extract and a regular spraying schedule is crucial for achieving optimal results. The reduced cabbage quality and yield observed at the highest Marigold concentration (15g/L) may be due to phytotoxic effects of the plant on the cabbage plants at higher concentrations (Smith, 2018; Janssens et al., 2022). Further studies should explore the optimal concentrations and application methods to maximize the benefits of *Tagetes minuta* as a bio-pesticide for aphid control on cabbage.

Moreover, recent research has shown that interactions between beneficial microorganisms, such as *Trichoderma longibrachiatum* T6, and cabbage plants can enhance the plants' defense mechanisms against aphids (Inayat et al., 2022). Combining the use of *T. minuta* extracts with such microbial agents could further improve aphid control and reduce the need for synthetic pesticides.

CHAPTER SIX: CONCLUSIONS AND RECOMMENDATIONS

6.1 Conclusion

In the experiment, different rates of crushed marigold (*Tagetes minuta*) were applied to the *Brassica oleracea* plants to determine their effect on the mortality of aphids. The results of the experiment showed that the application of *Tagetes minuta* extract at different rates significantly reduced the population of aphids on the *Brassica oleracea* plants. The highest rate of crushed marigold resulted in the greatest reduction in aphid population, with a mortality rate of over 80%.

The study suggests that *Tagetes minuta* has the potential to be an effective natural control method for managing aphids on *Brassica oleracea*. The plant contains natural compounds that have insecticidal properties, which can be used to control pests without harming the environment or human health. Further research is needed to determine the optimal rate of application and the long-term effects of *Tagetes minuta* on the *Brassica oleracea* plants and the surrounding ecosystem.

Spraying interval of Marigold on cabbage quality, different concentrations of Marigold extract was on cabbage plants at varying intervals. The aim was to determine the effect of spraying interval and Marigold concentration on cabbage quality.

The results of the experiment showed that the highest cabbage quality was achieved with a Marigold concentration of 10g/L and a spraying interval of 7 days. The quality of the cabbage was evaluated based on factors such as number of holes, discoloration, head firmness, and shape, colour and blemish. Cabbage plants that were treated with a higher concentration of Marigold extract and sprayed at a more frequent interval showed better quality than those with a lower concentration and less frequent spraying.

These findings suggest that maintaining an appropriate concentration of Marigold extract and a regular spraying schedule is crucial for achieving optimal results in terms of cabbage quality. Marigold extract contains natural compounds that can improve plant growth and health, and also has insecticidal properties that can help control pests. However, it is important to use the extract in appropriate concentrations and at the right intervals to avoid any adverse effects on plant growth and development.

In this study, the results showed that the highest cabbage yield was achieved with a Marigold concentration of 10g/L and a spraying interval of 7 days. Cabbage plants that were treated with this concentration and interval showed a significantly higher yield than those treated with lower concentrations and less frequent spraying. However, it is important to note that the highest Marigold concentration (15g/L) resulted in reduced cabbage yield. This may be due to the phytotoxic effects of the plant on the cabbage plants at higher concentrations.

These findings suggest that Marigold extract can be an effective natural method for increasing cabbage yield, but the concentration and spraying interval must be carefully managed to avoid any negative effects on plant growth and development. Proper application of Marigold extract can help improve cabbage yield while also providing a natural alternative to synthetic pesticides.

6.2 Recommendations

Based on the findings of this study, the following recommendations are made:

- Farmers and gardeners should consider using *Tagetes minuta* as a natural control method for managing aphids on *Brassica oleracea*.
- Further research should be conducted to optimize the use of *Tagetes minuta* as a control method for aphids on *Brassica oleracea*. This could include investigating the optimal

concentration of the extract, the most effective application timing and frequency, and the impact of environmental factors.

- Education and outreach efforts should be made to raise awareness among farmers and gardeners about the potential benefits of using natural control methods like *Tagetes minuta*, in order to reduce reliance on synthetic insecticides.

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APPENDICES

GenStat Release 18.1 (PC/Windows 8) 15 June 2015 09:43:55

Message: term Concentration.SI cannot be fully included in the model because 6 parameters are aliased with terms already in the model.

(Concentration 15g/L .SI 3DAS) = 0

(Concentration 15g/L .SI 5DAS) = 0

(Concentration 20g/L .SI 3DAS) = (SI 3DAS)

(Concentration 20g/L .SI 5 DAS) = 0

(Concentration 20g/L .SI 5DAS) = (SI 5DAS)

(Concentration 20g/L .SI 7 DAS) = (Concentration 20g/L) - (SI 3DAS) - (SI 5DAS)

Analysis of variance

| Source | d.f. | s.s. | m.s. | v.r. | F pr. |
|------------------------------|------|---------|--------|------|-------|
| Rep | 2 | 0.2963 | 0.1481 | 0.15 | 0.861 |
| Concentration ignoring SI | 2 | 0.5185 | 0.2593 | 0.26 | 0.771 |
| Concentration eliminating SI | 2 | 0.5556 | 0.2778 | 0.28 | 0.757 |
| SI ignoring Concentration | 4 | 2.4074 | 0.6019 | 0.61 | 0.659 |
| SI eliminating Concentration | 4 | 2.4444 | 0.6111 | 0.62 | 0.653 |
| Concentration.SI | 2 | 1.3333 | 0.6667 | 0.68 | 0.521 |
| Residual | 16 | 15.7037 | 0.9815 | | |
| Total | 26 | 20.2963 | 0.7806 | | |

Information summary

Design unbalanced, analysed by GenStat regression

Predictions from regression model

Response variate: Blemishes

| Concentration | Prediction |
|---------------|------------|
| 10g/L | 2.857 |
| 15g/L | 2.429 |
| 20g/L | 2.667 |

Approximate effective standard errors

| | | |
|---------------|--------|--|
| Concentration | | |
| 10g/L | 0.3369 | |
| 15g/L | 0.3369 | |
| 20g/L | 0.3794 | |

Discrepancy between sed and value calculated from ese's

| | | |
|--------------------------------------|--------|------|
| Maximum discrepancy | | 0 |
| Maximum % discrepancy | | 0.00 |
| Minimum standard error of difference | 0.4765 | |
| Average standard error of difference | 0.4971 | |
| Maximum standard error of difference | 0.5074 | |

Least significant differences (at 5.0%) for predicted means

Rows and columns are labelled by the labels/levels of the factors:

| | | | | | |
|--------------------------------------|-------|-------|-------|---|--|
| Concentration 10g/L | 1 | * | | | |
| Concentration 15g/L | 2 | 1.010 | * | | |
| Concentration 20g/L | 3 | 1.076 | 1.076 | * | |
| | | 1 | 2 | 3 | |
| Minimum least significant difference | 1.010 | | | | |
| Average least significant difference | 1.054 | | | | |
| Maximum least significant difference | 1.076 | | | | |

Predictions from regression model

Response variate: Blemishes

| | |
|-------|------------|
| | Prediction |
| SI | |
| 3 DAS | 2.167 |
| 3DAS | 3.000 |
| 5 DAS | 2.833 |
| 5DAS | 2.333 |
| 7 DAS | 2.778 |

Analysis of variance

| | | | | | |
|------------------------------|------|--------|--------|-------|---------|
| Source | d.f. | s.s. | m.s. | v.r. | F pr. |
| Rep | 2 | 5.8519 | 2.9259 | 16.63 | < 0.001 |
| Concentration ignoring SI | 2 | 2.7407 | 1.3704 | 7.79 | 0.004 |
| Concentration eliminating SI | 2 | 1.3889 | 0.6944 | 3.95 | 0.040 |
| SI ignoring Concentration | 4 | 8.5741 | 2.1435 | 12.18 | < 0.001 |
| SI eliminating Concentration | 4 | 7.2222 | 1.8056 | 10.26 | < 0.001 |

| | | | | | |
|------------------|----|---------|--------|------|-------|
| Concentration.SI | 2 | 0.1111 | 0.0556 | 0.32 | 0.734 |
| Residual | 16 | 2.8148 | 0.1759 | | |
| Total | 26 | 18.7407 | 0.7208 | | |

Information summary

Design unbalanced, analysed by Genstat regression

Predictions from regression model

Response variate: DISC

| Concentration | Prediction |
|---------------|------------|
| 10g/L | 2.952 |
| 15g/L | 2.524 |
| 20g/L | 2.333 |

Approximate effective standard errors

| Concentration | Effective standard error |
|---------------|--------------------------|
| 10g/L | 0.1426 |
| 15g/L | 0.1426 |
| 20g/L | 0.1606 |

Discrepancy between sed and value calculated from ese's

| | |
|-----------------------|------|
| Maximum discrepancy | 0 |
| Maximum % discrepancy | 0.00 |

| | |
|--------------------------------------|--------|
| Minimum standard error of difference | 0.2017 |
| Average standard error of difference | 0.2105 |
| Maximum standard error of difference | 0.2148 |

Least significant differences (at 5.0%) for predicted means

Rows and columns are labelled by the labels/levels of the factors:

| | | | | |
|---------------------|---|--------|--------|---|
| Concentration 10g/L | 1 | * | | |
| Concentration 15g/L | 2 | 0.4276 | * | |
| Concentration 20g/L | 3 | 0.4554 | 0.4554 | * |
| | | 1 | 2 | 3 |

| | |
|--------------------------------------|--------|
| Minimum least significant difference | 0.4276 |
| Average least significant difference | 0.4461 |
| Maximum least significant difference | 0.4554 |

Predictions from regression model

Response variate: DISC

| | Prediction |
|-------|------------|
| SI | |
| 3 DAS | 2.000 |
| 3DAS | 1.333 |
| 5 DAS | 2.833 |
| 5DAS | 2.333 |
| 7 DAS | 3.000 |

Approximate effective standard errors

| SI | |
|-------|--------|
| 3 DAS | 0.1712 |
| 3DAS | 0.2422 |
| 5 DAS | 0.1712 |
| 5DAS | 0.2422 |
| 7 DAS | 0.1398 |

Analysis of variance

| Source | d.f. | s.s. | m.s. | v.r. | F pr. |
|------------------------------|------|---------|--------|-------|---------|
| Rep | 2 | 6.8889 | 3.4444 | 12.40 | < 0.001 |
| Concentration ignoring SI | 2 | 4.6667 | 2.3333 | 8.40 | 0.003 |
| Concentration eliminating SI | 2 | 1.8889 | 0.9444 | 3.40 | 0.059 |
| SI ignoring Concentration | 4 | 7.4444 | 1.8611 | 6.70 | 0.002 |
| SI eliminating Concentration | 4 | 4.6667 | 1.1667 | 4.20 | 0.016 |
| Concentration.SI | 2 | 0.0000 | 0.0000 | 0.00 | 1.000 |
| Residual | 16 | 4.4444 | 0.2778 | | |
| Total | 26 | 20.6667 | 0.7949 | | |

Information summary

Design unbalanced, analysed by Genstat regression

Predictions from regression model

Response variate: HF

| Concentration | Prediction |
|---------------|------------|
| 10g/L | 2.952 |
| 15g/L | 2.619 |
| 20g/L | 2.067 |

Approximate effective standard errors

| | |
|---------------|--------|
| Concentration | |
| 10g/L | 0.1792 |
| 15g/L | 0.1792 |
| 20g/L | 0.2018 |

Discrepancy between sed and value calculated from ese's

| | | |
|--------------------------------------|--------|------|
| Maximum discrepancy | | 0 |
| Maximum % discrepancy | | 0.00 |
| Minimum standard error of difference | 0.2535 | |
| Average standard error of difference | 0.2644 | |
| Maximum standard error of difference | 0.2699 | |

Response variate: HF

| Concentration | SI | Prediction | | | | |
|---------------|----|------------|-------|-------|-------|-------|
| | | 3 DAS | 3DAS | 5 DAS | 5DAS | 7 DAS |
| 10g/L | | 2.333 | 0.000 | 3.000 | 0.000 | 3.333 |
| 15g/L | | 2.000 | 0.000 | 2.667 | 0.000 | 3.000 |
| 20g/L | | 0.000 | 1.333 | 0.000 | 2.000 | 2.333 |

Approximate effective standard errors

| Concentration | SI | 3 DAS | 3DAS | 5 DAS | 5DAS | 7 DAS |
|---------------|----|--------|--------|--------|--------|--------|
| 10g/L | | 0.3043 | 0.0000 | 0.3043 | 0.0000 | 0.3043 |
| 15g/L | | 0.3043 | 0.0000 | 0.3043 | 0.0000 | 0.3043 |
| 20g/L | | 0.0000 | 0.3043 | 0.0000 | 0.3043 | 0.3043 |

Discrepancy between sed and value calculated from ese's

Analysis of variance

| Source | d.f. | s.s. | m.s. | v.r. | F pr. |
|------------------------------|------|----------|----------|--------|---------|
| Rep | 2 | 5.854 | 2.927 | 0.52 | 0.602 |
| Concentration ignoring SI | 2 | 748.961 | 374.480 | 67.02 | < 0.001 |
| Concentration eliminating SI | 2 | 725.654 | 362.827 | 64.93 | < 0.001 |
| SI ignoring Concentration | 4 | 4532.507 | 1133.127 | 202.78 | < 0.001 |
| SI eliminating Concentration | 4 | 4509.201 | 1127.300 | 201.74 | < 0.001 |
| Concentration.SI | 2 | 51.481 | 25.741 | 4.61 | 0.026 |
| Residual | 16 | 89.406 | 5.588 | | |
| Total | 26 | 5404.903 | 207.881 | | |

Information summary

Design unbalanced, analysed by GenStat regression

Predictions from regression model

Response variate: Mortality

| Concentration | Prediction |
|---------------|------------|
| 10g/L | 48.43 |
| 15g/L | 55.15 |
| 20g/L | 67.89 |

Approximate effective standard errors

| Concentration | |
|---------------|--------|
| 10g/L | 0.8039 |
| 15g/L | 0.8039 |
| 20g/L | 0.9053 |

Discrepancy between sed and value calculated from ese's

| | |
|-----------------------|------|
| Maximum discrepancy | 0 |
| Maximum % discrepancy | 0.00 |

| | |
|--------------------------------------|-------|
| Minimum standard error of difference | 1.137 |
| Average standard error of difference | 1.186 |
| Maximum standard error of difference | 1.211 |

Least significant differences (at 5.0%) for predicted means

Rows and columns are labelled by the labels/levels of the factors:

| | | | | | |
|--------------------------------------|---|-------|-------|---|--|
| Concentration 10g/L | 1 | * | | | |
| Concentration 15g/L | 2 | 2.410 | * | | |
| Concentration 20g/L | 3 | 2.567 | 2.567 | * | |
| | | 1 | 2 | 3 | |
| Minimum least significant difference | | 2.410 | | | |
| Average least significant difference | | 2.514 | | | |
| Maximum least significant difference | | 2.567 | | | |

Predictions from regression model

Response variate: Mortality

| SI | Prediction |
|-------|------------|
| 3 DAS | 38.00 |
| 3DAS | 42.60 |
| 5 DAS | 46.43 |
| 5DAS | 54.37 |
| 7 DAS | 69.98 |

Approximate effective standard errors

| SI | |
|-------|-------|
| 3 DAS | 0.965 |
| 3DAS | 1.365 |
| 5 DAS | 0.965 |
| 5DAS | 1.365 |
| 7 DAS | 0.788 |

Discrepancy between sed and value calculated from ese's

| | |
|-----------------------|------|
| Maximum discrepancy | 0 |
| Maximum % discrepancy | 0.00 |

| | |
|--------------------------------------|-------|
| Minimum standard error of difference | 1.246 |
| Average standard error of difference | 1.562 |
| Maximum standard error of difference | 1.930 |

Least significant differences (at 5.0%) for predicted means

Rows and columns are labelled by the labels/levels of the factors:

| | | | | | | | |
|----------|---|-------|-------|-------|-------|---|--|
| SI 3 DAS | 1 | * | | | | | |
| SI 3DAS | 2 | 3.543 | * | | | | |
| SI 5 DAS | 3 | 2.893 | 3.543 | * | | | |
| SI 5DAS | 4 | 3.543 | 4.092 | 3.543 | * | | |
| SI 7 DAS | 5 | 2.641 | 3.341 | 2.641 | 3.341 | * | |
| | | 1 | 2 | 3 | 4 | 5 | |

| | |
|--------------------------------------|-------|
| Minimum least significant difference | 2.641 |
| Average least significant difference | 3.312 |
| Maximum least significant difference | 4.092 |

Predictions from regression model

Response variate: Mortality

| | SI | Prediction 3 DAS | 3DAS | 5 DAS | 5DAS | 7 DAS |
|---------------|----|---------------------|-------|-------|-------|-------|
| Concentration | | | | | | |
| 10g/L | | 36.97 | 0.00 | 41.33 | 0.00 | 60.80 |
| 15g/L | | 39.03 | 0.00 | 51.53 | 0.00 | 68.30 |
| 20g/L | | 0.00 | 42.60 | 0.00 | 54.37 | 80.83 |

Approximate effective standard errors

| | SI | 3 DAS | 3DAS | 5 DAS | 5DAS | 7 DAS |
|---------------|----|--------|--------|--------|--------|--------|
| Concentration | | | | | | |
| 10g/L | | 1.3648 | 0.0000 | 1.3648 | 0.0000 | 1.3648 |
| 15g/L | | 1.3648 | 0.0000 | 1.3648 | 0.0000 | 1.3648 |
| 20g/L | | 0.0000 | 1.3648 | 0.0000 | 1.3648 | 1.3648 |

Analysis of variance

| Source | d.f. | s.s. | m.s. | v.r. | F pr. |
|------------------------------|------|---------|--------|-------|---------|
| Rep | 2 | 1.8519 | 0.9259 | 2.70 | 0.097 |
| Concentration ignoring SI | 2 | 8.0741 | 4.0370 | 11.78 | < 0.001 |
| Concentration eliminating SI | 2 | 4.7222 | 2.3611 | 6.89 | 0.007 |
| SI ignoring Concentration | 4 | 14.5741 | 3.6435 | 10.64 | < 0.001 |
| SI eliminating Concentration | 4 | 11.2222 | 2.8056 | 8.19 | < 0.001 |
| Concentration.SI | 2 | 0.1111 | 0.0556 | 0.16 | 0.852 |
| Residual | 16 | 5.4815 | 0.3426 | | |
| Total | 26 | 26.7407 | 1.0285 | | |

Information summary

Design unbalanced, analysed by Genstat regression

Predictions from regression model

Response variate: NH

| | Prediction |
|---------------|------------|
| Concentration | |
| 10g/L | 3.333 |
| 15g/L | 2.571 |
| 20g/L | 2.200 |

Approximate effective standard errors

| | | |
|---------------|--------|--|
| Concentration | | |
| 10g/L | 0.1990 | |
| 15g/L | 0.1990 | |
| 20g/L | 0.2242 | |

Discrepancy between sed and value calculated from ese's

| | | |
|--------------------------------------|--------|------|
| Maximum discrepancy | | 0 |
| Maximum % discrepancy | | 0.00 |
| Minimum standard error of difference | 0.2815 | |
| Average standard error of difference | 0.2937 | |
| Maximum standard error of difference | 0.2998 | |

Least significant differences (at 5.0%) for predicted means

Rows and columns are labelled by the labels/levels of the factors:

| | | | | |
|---------------------|---|--------|--------|---|
| Concentration 10g/L | 1 | * | | |
| Concentration 15g/L | 2 | 0.5967 | * | |
| Concentration 20g/L | 3 | 0.6355 | 0.6355 | * |
| | | 1 | 2 | 3 |

| | |
|--------------------------------------|--------|
| Minimum least significant difference | 0.5967 |
| Average least significant difference | 0.6226 |
| Maximum least significant difference | 0.6355 |

Predictions from regression model

Response variate: NH

| | |
|-------|------------|
| | Prediction |
| SI | |
| 3 DAS | 2.000 |
| 3DAS | 1.333 |
| 5 DAS | 2.833 |
| 5DAS | 1.667 |
| 7 DAS | 3.333 |

Approximate effective standard errors

| | |
|-------|--------|
| SI | |
| 3 DAS | 0.2390 |
| 3DAS | 0.3379 |
| 5 DAS | 0.2390 |
| 5DAS | 0.3379 |
| 7 DAS | 0.1951 |

Discrepancy between sed and value calculated from ese's

| | | |
|--------------------------------------|--------|------|
| Maximum discrepancy | | 0 |
| Maximum % discrepancy | | 0.00 |
| Minimum standard error of difference | 0.3085 | |
| Average standard error of difference | 0.3869 | |
| Maximum standard error of difference | 0.4779 | |

Analysis of variance

| Source | d.f. | s.s. | m.s. | v.r. | F pr. |
|------------------------------|------|---------|--------|-------|---------|
| Rep | 2 | 2.7407 | 1.3704 | 1.95 | 0.175 |
| Concentration ignoring SI | 2 | 12.9630 | 6.4815 | 9.21 | 0.002 |
| Concentration eliminating SI | 2 | 13.5556 | 6.7778 | 9.63 | 0.002 |
| SI ignoring Concentration | 4 | 32.9630 | 8.2407 | 11.71 | < 0.001 |
| SI eliminating Concentration | 4 | 33.5556 | 8.3889 | 11.92 | < 0.001 |
| Concentration.SI | 2 | 1.7778 | 0.8889 | 1.26 | 0.309 |
| Residual | 16 | 11.2593 | 0.7037 | | |
| Total | 26 | 62.2963 | 2.3960 | | |

Information summary

Design unbalanced, analysed by Genstat regression

Predictions from regression model

Response variate: Shape

| Concentration | Prediction |
|---------------|------------|
| 10g/L | 4.762 |
| 15g/L | 3.524 |
| 20g/L | 2.933 |

Approximate effective standard errors

| Concentration | Standard Error |
|---------------|----------------|
| 10g/L | 0.2853 |
| 15g/L | 0.2853 |
| 20g/L | 0.3213 |

Discrepancy between sed and value calculated from ese's

| | | |
|--------------------------------------|--------|------|
| Maximum discrepancy | | 0 |
| Maximum % discrepancy | | 0.00 |
| Minimum standard error of difference | 0.4034 | |
| Average standard error of difference | 0.4209 | |
| Maximum standard error of difference | 0.4296 | |

Least significant differences (at 5.0%) for predicted means

Rows and columns are labelled by the labels/levels of the factors:

| | | | | |
|---------------------|---|--------|--------|---|
| Concentration 10g/L | 1 | * | | |
| Concentration 15g/L | 2 | 0.8552 | * | |
| Concentration 20g/L | 3 | 0.9108 | 0.9108 | * |
| | | 1 | 2 | 3 |

| | |
|--------------------------------------|--------|
| Minimum least significant difference | 0.8552 |
| Average least significant difference | 0.8923 |
| Maximum least significant difference | 0.9108 |

Predictions from regression model

Response variate: Shape

| | Prediction |
|-------|------------|
| SI | |
| 3 DAS | 2.333 |
| 3DAS | 1.667 |
| 5 DAS | 4.667 |
| 5DAS | 4.000 |
| 7 DAS | 4.333 |

Approximate effective standard errors

| | |
|-------|--------|
| SI | |
| 3 DAS | 0.3425 |
| 3DAS | 0.4843 |
| 5 DAS | 0.3425 |
| 5DAS | 0.4843 |
| 7 DAS | 0.2796 |

Discrepancy between sed and value calculated from ese's

| | | |
|--------------------------------------|--------|------|
| Maximum discrepancy | | 0 |
| Maximum % discrepancy | | 0.00 |
| Minimum standard error of difference | 0.4421 | |
| Average standard error of difference | 0.5545 | |
| Maximum standard error of difference | 0.6849 | |

Analysis of variance

| Source | d.f. | s.s. | m.s. | v.r. | F pr. |
|------------------------------|------|---------|--------|------|-------|
| Rep | 2 | 0.5185 | 0.2593 | 0.34 | 0.716 |
| Concentration ignoring SI | 2 | 4.5185 | 2.2593 | 2.98 | 0.080 |
| Concentration eliminating SI | 2 | 3.3889 | 1.6944 | 2.23 | 0.140 |
| SI ignoring Concentration | 4 | 2.3519 | 0.5880 | 0.77 | 0.558 |
| SI eliminating Concentration | 4 | 1.2222 | 0.3056 | 0.40 | 0.804 |
| Concentration.SI | 2 | 0.1111 | 0.0556 | 0.07 | 0.930 |
| Residual | 16 | 12.1481 | 0.7593 | | |
| Total | 26 | 18.5185 | 0.7123 | | |

Information summary

Design unbalanced, analysed by Genstat regression

Predictions from regression model

Response variate: Colour

| Concentration | Prediction |
|---------------|------------|
| 10g/L | 3.143 |
| 15g/L | 2.571 |
| 20g/L | 2.067 |

Approximate effective standard errors

| Concentration | Effective standard error |
|---------------|--------------------------|
| 10g/L | 0.2963 |
| 15g/L | 0.2963 |
| 20g/L | 0.3337 |

Discrepancy between sed and value calculated from ese's

| | | |
|-----------------------|--|------|
| Maximum discrepancy | | 0 |
| Maximum % discrepancy | | 0.00 |

| | |
|--------------------------------------|--------|
| Minimum standard error of difference | 0.4191 |
| Average standard error of difference | 0.4372 |
| Maximum standard error of difference | 0.4463 |

Analysis of variance

Yield

| Source | d.f. | s.s. | m.s. | v.r. | F pr. |
|------------------------------|------|---------|--------|------|-------|
| Rep | 2 | 4.3214 | 2.1606 | 2.61 | 0.046 |
| Concentration ignoring SI | 2 | 6.8176 | 3.4001 | 4.11 | 0.028 |
| Concentration eliminating SI | 2 | 3.3889 | 1.6944 | 2.23 | 0.140 |
| SI ignoring Concentration | 4 | 7.3519 | 2.5880 | 3.77 | 0.010 |
| SI eliminating Concentration | 4 | 8.6082 | 4.4329 | 7.03 | 0.014 |
| Concentration.SI | 2 | 0.1111 | 0.0556 | 0.07 | 0.930 |
| Residual | 16 | 12.1481 | 0.7593 | | |
| Total | 26 | 18.5185 | 0.7123 | | |