RABBIT URINE AND SUNFLOWER OIL REINFORCED TEPHROSIA-BASED FORMULATION FOR COWPEA INSECTPESTS MANAGEMENT IN SINGIDA, TANZANIA

Agricola Matle

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ABSTRACT

Insect pests are a major problem in cowpea production. Synthetic pesticides are used to control; however, are non-friendly to human and diversity of organisms. These negative effects raise farmers' interest in botanical pesticides due to less harmful effects on the ecosystem. *Tephrosia vogelii* extracts, rabbit urine, and sunflower oil were assessed for synergistic effects against cowpea field's insectpest's including aphids, pod borer, leaf miner, and beneficial insects. The ingredients were combined in ratio of 10% (w/v) *Tephrosia Vogelii* (T), 50% (v/v) rabbit urine (U), and 10% (v/v) of sunflower oil (O), while unsprayed plots and synthetic pesticide (Lambda-cyhalothrin 2.5 EC) were control negative and positive, respectively. The experiment was laid down in randomized complete block design (RCBD) with three replications. Spraying was done weekly where abundances for insects were recorded a day before next schedule. Results showed significant difference $P \leq 0.001$ in insect pest counts between treatments. The positive control exhibited smaller mean number (4.3 ± 0.3 d, 4.7±0.3 a and 5.0±0.6 a), followed by combined formulation OUT at a mean (11.0 ± 0.6 c, 8.0±0.6 b, and 4.3±0.3 a) for aphid, leaf miner, and pod borer respectively compared to negative control and individual ingredients (O, U and T) evidencing synergy. The results also indicated the combined formulation (Oil, rabbit urine and Tephrosia) OUT showed comparable yield of (846.1 kg/ha) with control positive (794.6 kg/ha) while uncombine formulation and negative control showed lower yield of 520.6 kg/ha, 611.1 (kg/ha), 662.2 kg/ha, and 483.3 kg/ha respectively. Based on the results, OUT-formulation exhibits synergy for managing cowpea's insectpest's, however study on cost-benefit is recommended.

DECLARATION

I, Agricola Matle do hereby declare to the Senate of the Nelson Mandela African Institution of Science and Technology that this dissertation is my original work that has neither been submitted nor being concurrently submitted for degree award in any other institution.

Agricola Matle Date

The above declaration is confirmed by:

Prof. Ernest R. Mbega Date

Prof. Paul Kusolwa Date

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CERTIFICATION

The undersigned certify that they have read the dissertation titled "*Rabbit Urine and Sunflower Oil Reinforced Tephrosia-Based Formulation for Cowpea Insectpests Management in Singida, Tanzania*" and recommended for partial fulfillment of the requirements for the Degree of Master of Science in Biodiversity and Ecosystem Management of the Nelson Mandela African Institution of Science and Technology.

Prof. Ernest R. Mbega Date

Prof. Paul Kusolwa Date

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DEDICATION

I dedicate this dissertation to my parents, Mr. and Mrs. Panga for their support to my studies.

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LIST OF ABBREVIATIONS ANSD SYMBOLS

ANOVA test	Analysis of Variance
DDT	Dichlorodiphenyltrichloroethane
Eos	Essential Oils
HPR	Plant Host Resistance
HSD	Honestly Significant Difference
IITA	International Institute of Tropical Agriculture
LiSBE	School of Life Science and Bio-Engineering
O	Sunflower Oil
RCBD	Randomized Complete Block Design
SSA	Sub- Saharan Africa
T	*T. vogellii* Extract
TPHPA	Tanzania Plant Health and Pesticide Authority
U	Rabbit Urine
UASG	Ursolic Acid and Stearoyl Glucoside
IPM	Integrated Pest Management

CHAPTER ONE

INTRODUCTION

1.1 Background of the Problem

Cowpea *Vigna unguiculata* (L.), (Fabaceae) is among the major human food sources (Bozokalfa *et al.*, 2017). The crop has its origin in Africa and was introduced to the rest of the world such as India approximately 2000 to 3500 years ago (Kébé *et al.*, 2017). It is among the most extensively grown and consumed grain legumes in the world (Fang *et al.*, 2007). Cowpea is an important source of vegetables and grains, containing protein, zinc, iron, and phenolic compounds that are significant for human health (Carvalho *et al.*, 2012). Cowpea requires an annual rainfall between 350 to 500 mm and a temperature between 30-35°C (El Naim & Jabereldar, 2010). In Tanzania cowpea is grown in coastal regions, Mwanza, Dodoma, Tabora, Singida, and Shinyanga (Hella *et al.*, 2013).

Cowpea production is negatively affected by various conditions like poor soil fertility, drought, diseases, and insect pest (Chelav & Khashaveh, 2013), among these challenges, insects infestations is a major challenging factor reducing cowpea yield (Oyewale & Bamaiyi, 2013). Small holder cowpea growers depend on the extensive use of synthetic pesticide[i]s for insect pests' management. However, the chemicals are known to cause environmental pollution and threatening human health and the ecosystem (Edwards, 2013). For instance, *Azadirachta indica* has excellent insecticidal results for insect control but it affects the nervous system, acetylcholine receptors, and voltage gated-sodium pump in human beings and animals (Rani *et al.*, 2021). Synthetic Pyrethroids and DDT leads to several effects on human health such as skin and eye irritation, headache, and dizziness (Beard & Collaboration, 2006)

The application of various pesticidal plants in managing pests have reported as an environmentally friendly approach in managing insectpest's (Lengai *et al.*, 2020). *Tephrosia vogelii*, *Vernonia amygdalina*, *Tithonia diversifolia* and *Lantana camara* are scientifically proven for their effectiveness against insectpest's (Mkindi *et al.*, 2015). Another study by Alao *et al.* (2011) reported that *Tephrosia vogelii* and *Petiveria alliacea* at 20% and 10% v/v significantly protected cowpea's pods and grains. Rotenoids from *T. vogelii* was an active component against most of the sucking and biting insect pests. Fermented rabbit urine helps to manage insects including aphids, moths, leaf miners, caterpillars, and as organic fertilizer in cucumber (Okonji *et al.*, 2022). Other study reported that rabbit urine significantly reduced the survival of fall armyworm a major cereal pests during their instar larvae stage (Kemunto *et al.*, 2022). The use of plant oils against insect pests have been studied. Plant bio-pesticides based on essential oils (Eos) possess repellency, and insecticidal properties (Ibrahim, 2019). Some plants essential oils not only repel insects but also exhibit insecticidal actions on insects by contact (Isman, 2000).

However, despite their environmental friendliness, organic pesticides just like other botanical pesticides are proven to be are less effective compared to synthetic (Damalas & Koutroubas, 2020; Musa *et al.*, 2022). Combining two or more botanical pesticides may result into synergy (Oparaeke *et al.*, 2005). Combination of secondary metabolites on insectpest's hinder their infestation on crop than single compound (Rattan, 2010).

Since rabbit urine, *T. vogelii* extract and sunflower oil have been reported to have insecticidal properties, their combination may result in synergy. This study aimed at investigating the effects of a combination of *T. vogelii*, rabbit urine, and sunflower oil as agroecological friendly formulation for cowpea insect's pest management.

1.2 Statement of the Problem

Cowpea production is highly affected by insect pests. The use of synthetic pesticides is a popular practice among farmers, though they are expensive, disrupt ecological systems, and dangerous to human health and non-targeted beneficial insects in areas where they are administered (Rani *et al.,* 2021). These negative impacts lead to the utilization of botanical insecticides to be used as an alternative to synthetic pesticides since they are easily biodegradable and less toxic with less harm to the environment and humans (Miresmailli *et al.,* 2014). However, botanical pesticides are found to be less effective as compared to synthetic pesticides (Das, 2014), necessitating further research for improved performance. Combining pesticidal plant extracts is one of the methods for optimizing the efficacy of botanical pesticides (Stevenson *et al.,* 2017; Gaffar *et al.,* 2020). Secondary metabolites (natural products) found in bacteria *Escherichia coli* when mixed with neem extract has been found to increase crop protection against insectpest's and nematodes (Copping & Duke, 2007).

This research, therefore, investigated the synergistic effects of combining *T. vogellii* extract, rabbit urine, and sunflower oil in managing cowpea insect pests without harming beneficial insects.

1.3 Rationale of the Study

Cowpea production is negatively affected by various conditions like poor soil fertility, drought, diseases, and insect pest (Chelav & Khashaveh, 2013), among these challenges, insects infestations is a major challenging factor reducing cowpea yield (Oyewale & Bamaiyi, 2013). Small holder cowpea growers depend on the extensive use of synthetic pesticide s for insect pests' management. However, the chemicals are known to cause environmental pollution and threatening human health and the ecosystem (Edwards, 2013). Research reported that rabbit urine significantly reduced the survival of fall armyworm a major cereal pests during their instar larvae stage (Kemunto *et al.*, 2022). The use of plant oils against insect pests have been studied. Plant bio-pesticides based on essential oils (Eos) possess repellency, and insecticidal properties (Ibrahim, 2019). Some plants essential oils not only repel insects but also exhibit insecticidal actions on insects by contact (Isman, 2000). However, despite their environmental friendliness, organic pesticides just like other botanical pesticides are proven to be less effective compared to synthetic (Damalas & Koutroubas, 2020; Musa *et al.*, 2022). Combining two or more botanical pesticides may result into synergy (Oparaeke *et al.*, 2005). This research, therefore, investigated the synergistic effects of combining *T. vogellii* extract, rabbit urine, and sunflower oil in managing cowpea insect pests without harming beneficial insects.

1.4 Research Objectives

1.4.1 General Objective

Assessing the synergistic effect of rabbit urine and sunflower oil on tephrosia-based formulation for cowpea insectpest's management

1.4.2 Specific Objectives

(i) To assess the effects of the Tephrosia-based formulation on cowpea insect pest abundance.

(ii) To assess the effects of Tephrosia-based-formulation on beneficial insects.

(iii) To assess the efficacy of Tephrosia-based formulation on cowpea yield, and yield component.

1.5 Research Questions

(i) How do Tephrosia-based-formulation influence the abundance of cowpea insect pests?

(ii) How does Tephrosia-based-formulation affect beneficial insects?

(iii) How does Tephrosia-based formulation affect cowpea yields, and yields' components?

1.6 Significance of the Study

The study is an effort to enhance the performance of organic pesticides which should in turn encourage agroecological farming for the sustenance and wellbeing of the ecosystem. Moreover, the outcome of the research has the potential to minimize the production cost for cowpea small scale farmers through the use of home-made pesticides from naturally occurring and locally available pesticidal materials. The use of bio based-formulation in insect pests' management can potentially improve productivity while protecting beneficial insects.

1.7 Delineation of the Study

The present study focused on investigation of synergistic effect of rabbit urine and sunflower oil on Tephrosia-based formulation for cowpea insectpest's management. Therefore, the effects of the Tephrosia-based formulation was investigated on cowpea insect pest abundance, and the on beneficial insects. Finally the efficacy of Tephrosia-based formulation on cowpea yield, and yield component were assessed.

CHAPTER TWO

LITERATURE REVIEW

2.1 Insect Pest

Insect pests are fundamental limitation in yield which leads to massive crop losses in Africa's (Horn & Shimelis, 2020; Olaitan *et al.,* 2011) production of cowpea and the rest of the world where it is grown (Egbadzor *et al.*, 2013; Horn & Shimelis, 2020). When neglected or poorly managed insect pest's damage can lead to yield loss of 80% to 100% (Dugje *et al.,* 2009). Different insects cause damages at various phases of the crop's growth, necessitating the use of tolerant types and insecticide sprays (Olutona & Aderemi, 2019). In Tanzania damage by insectpest's on cowpea lead to lower yield of between 1.8 to 2.5 tons per ha for its varieties including black eyed cowpea (Kisetu *et al.,* 2013). According to FAOSTAT (2020), Tanzania contributed only 2% of the total African annual cowpea production of 7.1 million tones, the loss was attributed by biotic factors including insectpest's infestation (Lydia *et al*., 2020). The major insectpest's of cowpea includes Leaf miners, Whiteflies (*Bemisia tabaci*), Leafhoppers (*Empoasca sp*.), Mites (*Tetranychus spp.*), Thrips (*Megalurothrips sjostedti*), Bean leaf beetle (*Ootheca sp)*, Pod bug (*Clavigralla sp)*, Pod borer (*Maruca sp*.,) and Aphids (*Aphis craccivora*) (Oyewale & Bamaiyi, 2013).

Pests attack cowpea at three major growing stages; before flowering, during and after flowering, and post-harvest storage. Of all the mentioned insect pests, the most destructive to cowpea production are those affecting flowering and post-flowering stages which include; Aphids (*Aphis craccivora*), Pod- sucking bugs (*Anoplocnemis curvipes)*, Pod borer (*Maruca testulalis)*, Blister beetles (*Mylabris spp),* Flower thrips (*Megalurothrips sjostedti),* and Leaf miner (*Agromyzidae*) (Asante *et al.,* 2001). Attack by these insects is often so severe that farmers obtain no yield especially when grown without insecticide application.

2.1.1 Aphids (*Aphid craccivora*)

Cowpea Aphids produce by asexual means of reproduction and undergo completemetamorphosis from eggs to adults through larvae and pupa. The adult stage does more damage to cowpea plants. Aphids damage plants at every stage of growth from germination to post-flowering when pods are formed feeding on immature plants' stem tissues and leaves by sucking and on the mature plants' pods after flowering. The presence of honeydew on pods is clear evidence of aphids' feeding (Singh & Singh, 2021). The severity of aphid infection increased as plant spacing increased (Asiwe *et al.,* 2005).

Cowpea aphids damage plants directly and indirectly. Direct damage is by removing the sap leading to plants wilt and also releasing toxins into the plant as they feed. About 90% of cowpea crop yield losses are a result of direct feeding damage of aphids in non-resistance varieties and approximately 30% in resistant varieties (Allan *et al.,* 2016).

Indirectly, aphids damage cowpea by spreading plant viruses as they move from plants and paddocks (Dedryver *et al.,* 2010). These viruses include: bean yellow mosaic, alfalfa mosaic, cucumber mosaic, and pea seed-borne mosaic. Pulse crop yield losses as a result of aphids' spread of the virus account for approximately 80% (Valenzuela & Hoffmann, 2015). Cowpeas yield losses due to virus infection in Nigeria were recorded at 87% (Rao & Reddy, 2020).

Figure 1: Aphids infestation on cowpea's pods and leaves (This study, 2021)

2.1.2 Pod Borer

Pod borer belongs to the order *Lepidoptera* and the *Crambidae* family and is among the major cowpea insect pests. Its life cycle is completed within 30 -34 days at an average temperature of 28°C from egg to adult (Sonune *et al.*, 2010). The 1st, 2nd, and 3rd stage larvae feed on young cowpea leaves during development damaging foliar and fruits of the crop. Larger larvae feed on pods and consume the developing seeds inside the pod resulting in low yield (Patil *et al.,* 2017). The Pod borer results into an estimated annual economic loss approximating 30 million US Dollars in semi-arid tropics (Sharmad *et al.,* 2005)

Figure 2: Pod borer feeding on cowpea's pods (Patil *et al.*, 2017)

2.1.3 Leaf Miner

Cowpea leaf miner is among the most important insect pest species that feed on cowpea (Mahesha *et al.*, 2022). Females of this pest lay eggs on cowpea leaves and the larvae that result from them, feeding on the mesophyll tissue, which causes leaf drops (Soltan *et al.,* 2020). Infestation was particularly bad during flowering and pod-setting stages. Leaf miner feeding depends on the several factors in host plant including size and thickness of the leaf, and nutrient level beside leaf quality (AL-Ghadban *et al*., 2017). Biological control is one of the methods which was used since it is environmentally friendly. Parasitic wasp and nematodes are among the natural enemies in control of leaf miner.

2.2 Insect Pests' Management

Several techniques to control cowpea insect pests have been used to reduce the devastating effect at practically every stage of their development. The techniques involve cowpea pests in the fields and they include aphids, pod borers, leaf miners, and pod sucking bugs (Jackai & Adalla, 1997). Different methods of pests control including cultural, biological and chemical are reported to be used for management (Paddick *et al*., 2015). To minimize losses, integrated pest management (IPM), a farmer-based and knowledge-intensive management strategy, is essential to promoting natural and cultural control of pest populations (Kabote *et al.*, 2021). Previously many farmers did not know about spraying their cowpea crops rather than using cultural methods as reported by Erbaugh *et al.* (2010), this traditional way is of importance since it promotes control over natural enemies, though is not more successful to protect the damage of the crop by insect pests in the field. Due to the low yield produced by the farmers as a consequence of insect pests' destruction stated by Oyewale and Bamaiyi (2013), it was suggested that the natural management by itself cannot offer enough protection against the damage caused by the insect pests (Witzgall *et al.,* 2010). As the need for cowpea increases the production widens hence large-scale initiated in different places this led to improvement in the application of other helpful methods of cowpea insect control in the field (Tarawali *et al.,* 2002).

2.2.1 Cultural Control

Cultural methods of insect pests control involve approaches such as early planting, host plant resistance, intercropping, weed control, and crop rotation.

In the intercropping approach, cowpea is usually grown together with cereals like maize (*Zea mays*), cassava *(Manihot esculentum*), and sorghum (*Sorghum bicolar*), finger millet (*Eleusine coracana*) and other crops (Nampala *et al*., 2002). Crop density is increased while pest density is reduced via intercropping (Lithourgidis *et al.*, 2011). High crop diversity can change the insect habitat and may also disturb the insect's normal processes in its host plant (Karungi *et al.*, 2000). During intercropping, environmental factors like low temperature, high humidity, and minimum light transmission helps to reduce pest infestation. Similarly, the activity of natural enemies is thought to be higher under mixed crops (Tamò *et al*., 2012). Regardless of its advantage in insect management, this method is not the best method due to its effects on plant growth through nutrient competition and yield reduction. Thobatsi (2009), reported that there is no advantage to intercropping over solitary cropping.

(ii) Early Planting

Pests' damage has been minimized in numerous crops by planting at the right time. Prediction of host plants 'growth and insect's population outbreaks aids host plants in avoiding pest infestations (Karungi *et al.*, 2000). Early growing in the season reduced aphid infestation but it has no significant effect on thrips, legume pod borers, and pod-sucking bugs infestation (Adipala *et al.*, 1999). A variation in planting time earlier or delayed may result in the pest failing to coincide with a critical crop growth stage, thereby reducing or delaying the colonization of the crop by the pest (Murúa *et al.,* 2006).

(iii) Host Plant Resistance

Plant host resistance (HPR) is a cost-effective and environmentally friendly pest management strategy (Adipala *et al.*, 1999). The approach is well documented in pest control, especially against *A. craccivora.* Their ubiquitous and prolonged use results in the development of resistance to targeted insect pests. Michael *et al.* (2011), reported the usage of leafhopper-resistant cowpea cultivars, adopted at the International Institute of Tropical Agriculture (IITA). In Africa and Asia, several cowpea aphid-resistant cultivars have been created. In some plants, plant resistance has a successful result in suppressing pests' population or damage. The HPR is the natural quality of the plant that makes it unsuitable as food, shelter or insects also through antibiosis, non-preference, or providing the plant's ability to endure without loss in yields (Singh, 2011). Good resistance has been recognized for some pests of cowpea. The IITA has been a strong supporter of the use of plant resistance as the major means of pest management on cowpea (Singh, 2011). Regardless of the access to resistant varieties, aphids are still generally managed by synthetic insecticides because of their essential role as vectors of cowpea viral diseases. Therefore, there is still a need for the use of environmentally friendly pesticides (Biopesticide) which will both reduce insect pests as well as serve microorganisms and beneficial insects.

2.2.2 Chemical Control

This refers to the use of chemical insecticides in managing insect pests and it is the most common method for farmers due to their availability (Rai & Ingle, 2012). These availabilities offer the advantage of easy access to farmers anywhere. For years now, chemical pesticides have made a significant contribution to the fight against cowpea insect pests and diseases. Amongst the pesticides are Azodrin, Dursban, Lambda-cyhalothrin and Dimecron (Knodel, 2010).

Despite being effective in insect pest management, synthetic insecticides have many negative effects. Such as their toxicity to non-targeted creatures such as beneficial insects, wildlife, and humans (Mulla *et al.,* 2020). Mitra *et al.* (2011) reported the impacts of synthetic pesticides as they reduced bird's count. Human beings are also victims of synthetic pesticides. During application users' skin is affected, feeling difficulty in breathing, vomiting as well as farmers collapsing (Fuad *et al.,* 2012). Some synthetic insecticides such as Pyrethroids and DDT affect the nervous system, acetylcholine receptors, and voltage gated-sodium pump, in human beings (Rani *et al.,* 2021). Pesticides are also reported to enter body cells which results in some cancer in human beings (Andersson *et al.,* 2014). Leukemia, lung cancer, breast cancer, colon cancer, and kidney cancer are among the cancer results of the misuse of synthetic pesticides (Morais *et al.,* 2012). Pesticide uptake by *biota* has also been seen in studies, implying soil contamination (Zacharia, 2011). These disadvantages influenced scientists to think of alternative friendly pesticides (Biopesticides) to reduce the side effects. Pesticidal plants are so advantageous since they are very inexpensive and readily accessible, their formulations are biodegradable in the environment and have fewer hazardous harms (Bempah *et al.,* 2011).

2.2.3 Biological Control

Biological management is a substitute for chemical pesticides for sustainable Agro-ecological farming. It is a method of insect pest control using other organisms, such as predators, parasites, and herbivory, and sometimes involves an active role of human management to reduce the abundance to a level that is no longer harmful to crops. Another approach in biological control is the use of plant extracts for the synthesis of botanical pesticides.

2.3 The use of Botanical Pesticides for Insect Pest's Management

Botanical insecticides, also known as biopesticides, are readily biodegradable compounds isolated from plants (Oyewale *et al.,* 2013). Increase attention in environmental safety triggered the interest in pest control approaches through eco-friendly plant-based pesticides (Ngeba *et al* 2022). Predators' biological diversity is maintained by botanical insecticides (Amoabeng *et al.,* 2013), therefore, making their usage in farming a long- term pest's control strategy. Their effects are not always as long-lasting as chemical pesticides (Ebenezer, 2010). Moreover, they are generally pest-specific and have less insect development resistance possibility (Isman, 2006).

In Africa, many pesticidal plants are naturally accessible as they grow in the field and within farm boundaries, hence there is no or less cost for growing the plants. Despite its availability, the knowledge of the use of botanical pesticides is not well recognized in the markets among farmers (Stevenson *et al.,* 2017). About 2000 plant species are identified as possessing insecticidal properties (Kamaraj *et al.,* 2008), yet just a handful have undergone scientific evaluation. Across many countries in Sub- Saharan Africa (SSA) pesticide plants are widely distributed through their use tends to be restricted (Sola *et al.,* 2014). Indigenous Africans have long used insecticides derived from plants and other organisms for a variety of reasons, which include but not limited to insect pests' management (Deng *et al.,* 2009). *Capsicum frutescens, Tagetes spp., Nicotiana tabacum, Cypressus spp., Tephrosia vogelii,* and other plants like I*ndica Azadirachta Aloe spp., Eucalyptus spp., Musa spp., Moringa oleifera, Tithonia diversifolia, Lantana Camara,* *Phytollacca dodecandra, Vernonia amygdalina,* and *Musa spp* have been employed by farmers (Mugisha-Kamatenesi *et al.,* 2008). Priyanka *et al.* (2013), reported that, *Lantana Camara* is one of the pesticidal plants from the family *Verbenaceae.* Its leaves and flowers contain compounds like *ursolic* acid and *stearoyl glucoside* (UASG) (Kazmi *et al.,* 2017), which are associated with toxicity includes *triterpenoids* in a polar phase. *Lantana* has been utilized to keep insect pests out of stored grains (Rajashekar *et al.,* 2013). Studies indicate that it leads to decreased ability to move, dehydration, and constipation in mice, as well as a congested heart and lung, *nephrosis*, overall reproduction disfunction, and teratology (Kumar *et al.,* 2016).

2.3.1 Challenges to the Adoption of Botanical Pesticides

(i) Standardization of Unstable Bio-Active Ingredients and Residual Decomposition

Botanical pesticide active components are very susceptible to decomposition under various conditions because they are mostly in aqueous solutions where they are prone to chemical degradation and microbial activity (Liu *et al.,* 2017). Although easy decomposition, hence short half-life, would be a desirable quality for environmental friendliness, due to quick residue degradation, it leads to preparation and preservation complications. In this regard, an ideal pesticide should be chemically stable enough to allow long shelf-life but easily degradable after application to avoid environmental persistence where it may cause unnecessary harm to the environment (Vurro *et al.,* 2019). In addition, variations in the concentration of the active components in pesticide plant chemotypes and the growing conditions result in difficulty in the standardization of botanical pesticides (product standardization) which further complicates preparation.

(ii) Supply and Accessibility

The market supply of botanical pesticides is affected by inconsistent product due to standardization complications as compared to chemical pesticides. This therefore limits farmers' accessibility to such botanical pesticides as compared to chemical pesticides in managing insect pest (Sola *et al.,* 2014). It proposes that there is more room to grow a significant business that fits both domestic and worldwide demand for more environmentally friendly pest management.

(iii) Methods of Extraction of Complex Structure

The methods of extracting the complex bio-active components from pesticide plants can be knowledge-intensive and require a steep learning curve. The knowledge is lacking among farmers in developing countries compared to developed countries (Rates, 2001). Smallholder farmers who form the majority in the developing economies lack the appropriate technologies for the "Do-It-Yourself" approach of preparation for "village-level collective", and to reach the level of "private-sector industry" (Isman, 2017). The transitional gap between theory and practice still exists since research has been heavily weighted towards discoveries known as basic research as opposed to technological transfers or applied research.

2.3.2 Weakness of Botanical Pesticides

The preceding discussion presents the strengths of botanical pesticides over chemical pesticides. However, there are some weaknesses of botanical pesticides as follows:

(i) Slow Response

The effects of botanical pesticides on the insect pests are generally slow and most farmers have a perception that the application and use of botanical pesticides are not effective. Most botanical products have minimal information on their applicability, efficacy, and safety among farmers (Foerster *et al.,* 2001). Most farmers are discouraged by the high-frequency spraying and short residual period of botanical pesticides for effectiveness and this is a weakness that leads to poor adoption among farmers as an alternative to pest management.

(ii) The Bio-Active Compound of Plants

The exploitation of pesticidal plants is complicated by phytochemical variation (Sarasan *et al*., 2011) and it has been stated that they are not as effective as synthetic chemical pesticides because their pesticidal action is modest, varied, and often unknown. Different botanical plants sometimes have more than one bio-active ingredient with different levels of toxicity to non-target organisms (Islam *et al.,* 2011). This, therefore, requires complex modifications and optimization to targeted insect pests' management through their combination. One of the examples of pesticidal plants is pyrethrum which is less effective under UV light. Pyrethrum is nature-based insecticide derived from *Tanacetum cinerariifolium* flowers (*Asteraceae*) also it has been utilized in the control of field's pests such as bean aphids and storage pests (Kareru *et al.,* 2013). Natural pyrethrum contains six entomotoxic compounds cinerin, pyrethrin, and jasmolin (Manda *et al.,* 2020). It disrupts the insects' peripheral and central nervous systems resulting in paralysis in a similar way to DDT and other synthetic chemical pesticides (Davies *et al.,* 2007). Though pyrethrin is known to be very fast in knocking down and causing paralysis in insects, it is easily metabolized by insects which makes them recover instead of dying. Pyrethrin is also easily broken down by sunlight ultraviolet light and hence less effective for outdoor use. This UV effect on pyrethrin has led to the limited commercial production adoption.

2.4 Modification of Botanical Pesticides

Although botanical pesticides are less detrimental to human's health and environmentally, they still need to be improved to work as effectively as synthetic insecticides for insect pest management. Plant materials, on the other hand, are frequently used inefficiently, necessitating optimization and modification (Sola *et al.,* 2014). To handle this challenge, around the mid to late 20th century, research was actively conducted on biopesticides, to improve their efficiency through modifications using approaches such as additions of novel compounds like steroids and synergies (Wakeil, 2013).

2.4.1 Structural Modification

Some of the pesticidal Phyto-compounds (the active compounds in the botanical pesticides) have been used in the synthesis of chemical pesticides resulting in a nearly similar mechanism of action against insect pests. Among them, is the pyrethrum plants where pyrethrin was used as a template to synthesize pyrethroids as a chemical alternative which is not purely natural as pyrethrin (Kareru *et al.*, 2013). Despite being synthetic, pyrethroids exhibit most of the benefits of its natural template pyrethrin. It is more effective and less susceptible to UV light.

Neem as a pesticidal plant also undergoes structural modification. The structural modification of *Azadirachta indica* (gum neem) with urea in the presence of calcium chloride evaluates it as a pharmaceutical suspending agent (Ogunjimi & Alebiowu, 2014).

2.4.2 Combining Pesticidal Plants Extracts for Improving their Efficiency

Pesticidal plants contain a number of active components with different in chemical composition (Sola *et al.,* 2014). Studies reported that some botanicals have low efficacy and can be improved through combination with other plant species (Foluke & Abiodun, 2020). Some of these combinations involve neem a plant with pesticide properties, commonly used by farmers. Neem oil as a product of the neem tree (*Azadirachta indica*) is among the most important compounds in the present approach to pest management reference. It contains a number of insecticidal characteristics like repellency, toxicity, and growth disruption for various insect species. Neem acts as a repellent, preventing insects from starting to eat and then stopping due to the presence of a deterring effect (Wakeil, 2013). Neem is suspected to affect normal development by interfering with chitin synthesis as a growth regulator (ref). Neem oil is reported to exhibit greater toxicity in combinations with other botanicals like Karanja oils from *Militia pinnata* trees on aphids and mites (Kumar *et al.,* 2007). Nzanza and Mashela (2012) noted that fermented extract of neem and wild garlic managed whiteflies and aphids on tomato more effectively than either plant extract applied individually.

Likewise, research by Alao *et al.* (2011) proved that the combination of *T. vogelii* and *Petiveria alliacea* at 20% attained the same efficacy as a positive control (Deci) in controlling insect pests including aphids. Research on the combination of *T. Vogelii*, *T. diversifolia,* and *L. javanica* extracts showed the same effectiveness as the synthetic pesticide (Lambda-cyhalothrin) in crop yields and pest regulations. According to Oparaeke *et al*. (2000), the extract mixture of four plant extracts including cashew nutshell, African pepper, garlic bulb and chili pepper at 10:10% w/w reduced the number of flower bud thrips, pod borer and pod sucking bugs on cowpea. This synergistic effect also improved the grain yield by 4-5 times compared to untreated control.

This study assessed the synergistic effect of *Tephrosia* and rabbit urine formulation in 10% v/v sunflower oil for agro-ecological-based insect pests management and improved cowpea yields. The study involved assessing the efficacy of formulation on insect abundance, pollinators, yield, and yield components.

2.5 Toxicity of the Selected Plants and Rabbit Urine

2.5.1 *Tephrosia vogelii*

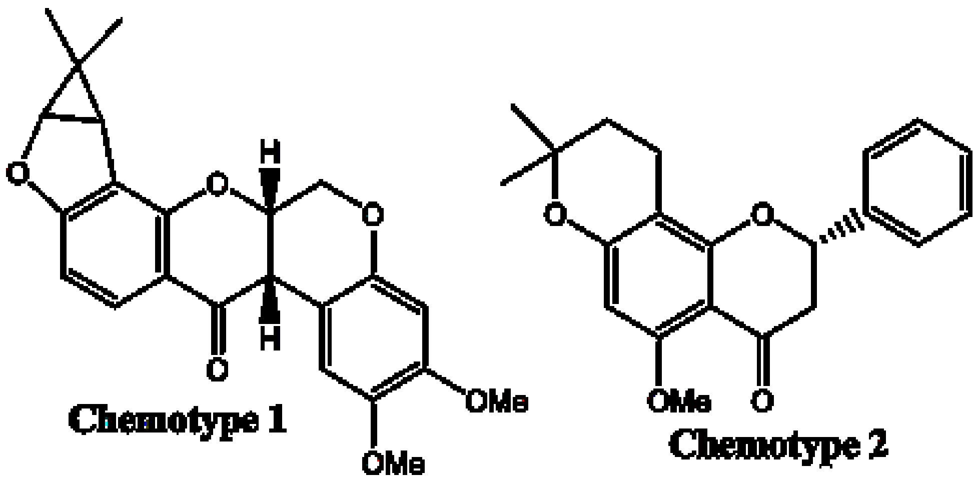
(i) Taxonomy and Habitat Distribution

Tephrosia is a soft woody branching herb species in the family of *Fabaceae*, known by its common name fish poison bean (Bruschi *et al.,* 2014) and Utupa in Swahili. It is a tropical African native plant that grows in a range of territories like savannah vegetation, forest edges, and shrubland (Mwaura *et al.,* 2012). It may be found up to 2100 m altitude in climates with an annual rainfall of roughly 850-2650 mm and typical annual temperatures of 12.5-26.2°C (Muyobela *et al.,* 2016).

(ii) Pesticidal Property

The pesticidal property of *T. vogelii* is attributed to its chemical composition of rotenoids deguelin, tephrosin, and rotenone (Belmain *et al.,* 2012). The rotenoids deguelin is a widely abundant class of flavonoids that is very toxic to insects and it is the most used botanical pesticide ingredient in Sub-Saharan Africa (Mkindi *et al.,* 2019). The rotenoids interfere with the electron transportation during the oxidation process from NADPH to NADP+ thereby blocking the mitochondrial oxidation of Krebs cycle intermediates. Hence, inhibiting oxygen supply to the cells and causing suffocation due to cellular respiratory depression (Zhang *et al.,* 2020).

There are different varieties of *T. vogelii* with unique chemotypes that differ in their chemical compositions (Belmain *et al.,* 2012). It has been found that the varieties with chemotype 1 possess insecticidal properties for use as botanical pesticides. However, those plant varieties with chemotype 2 have inactive flavonoids that are unresponsive to insects (Mkindi *et al.,* 2019). Figure 3 shows the chemical structure of chemotype 1 and chemotype 2 varieties and a picture of the *T. vogelii* plant.



(a) (b)

Figure 3: Chemical structure of flavonoid found in Chemotype 1(a) and (b) Chemotype 2 (Mkindi *et al.,* 2017)

Figure 4: A picture of Tephrosia vogelii plant (Mwaura *et al.,* 2012)

2.5.2 Rabbit Urine

Rabbit urine is one of the by-products of its metabolism. It is also used in crops as organic fertilizer. The presence of potential nutrients such as nitrogen, phosphorous, potassium, and calcium in rabbit urine promotes its efficiency compared to commercial foliar-fed fertilizers and contributed to the improved crop yield (Mutai, 2020). In other cases, farmers also use rabbit urine in managing insect pests in crops (reference). Corbeels *et al.* (2019) reported that fermented rabbit urine spray has been found to help in the management of insects and other pests, including aphids, moths, leaf miners, caterpillars, and mites. The pungent smell due to ammonium compounds in rabbit urine exhibits repellency characteristic of insect pests. These pesticidal properties were used as a major key to be used in this study.

Figure 5: Picture of rabbit urine in a white bucket on the left and rabbit on right (This study author's photograph, 2021)

2.5.3 Sunflower Oil

Oils extracted from plants possess pesticidal properties due to their ability to block insects' spiracles which results in suffocation. Numerous researchers have also documented the use of plant's oils against insect pests. On various insect species, plant bio-pesticides based on essential oils (Eos) possess poisonous, repellent, and insecticidal properties (Ibrahim, 2019).

Figure 6: A picture of sunflower oil extracted from sunflower seed (This study, 2021)

Sunflower oil at 10 mL significantly reduces the lifetime of cowpea storage pests (Rajapakse & Van Emden, 1997). Plant oils and powders obtained from different parts of various plants have also been used to protect cowpea from damage in storage. For instance, groundnut oil and palm oil are effective in the protection of infested cowpea by weevil *C. maculatus* leading to low emergence of adult weevils.

CHAPTER THREE

MATERIAL AND METHODS

3.1 Study Site

The experiment of the current study was conducted in Singida Region (Mandewa-rural Singida). The region is in northern portion of Tanzania's central plateau, between latitudes 30° 52' and 70° 34' south, longitude 33° 27' and 35° 26' east (Salaam, 2017). The climate in the region is semi-arid, with annual rainfall ranging from 500 to 800 mm. The temperature in the region varies from 15°C to 30°C, depending on the altitude of the area.

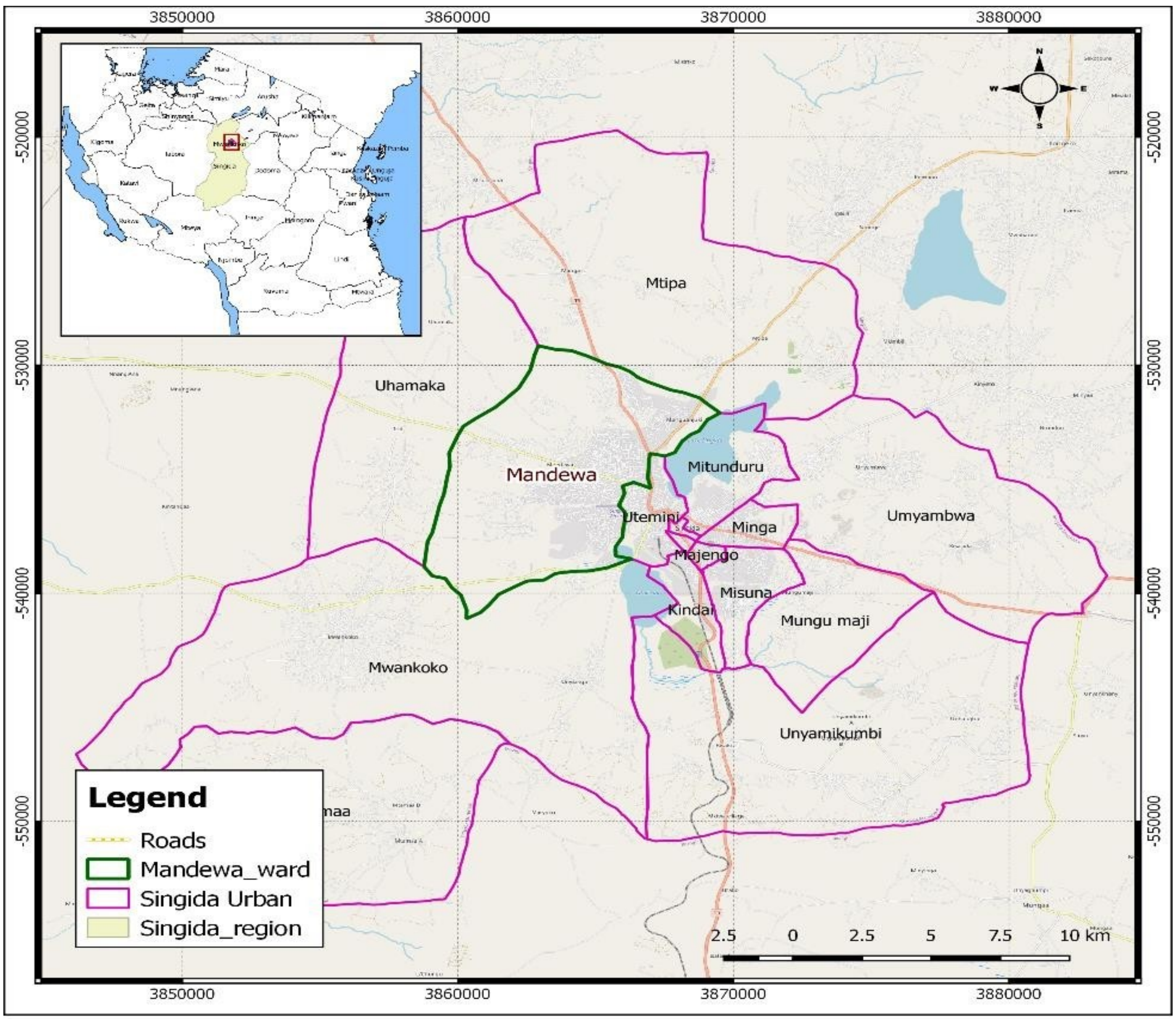


Figure 7: A map showing the study site

3.2 Materials Collection and Insecticide Formulation

The *T. vogelii* leaves were randomly picked from uncultivated farms in rural Singida district. To reduce the loss of volatile components, the leaves were dried in the shade for 7 days (Sejali & Anuar, 2011). The dried leaves were then pulverized with an electric grinder, put into 1 kilogram plastic bags, and kept in dry environment with no light. Local rabbit keepers and local industries in Singida provided rabbit urine and cold-pressed sunflower frying oil, respectively. To obtain an extract, the *T. vogelii* powder was steeped in an aqueous solution containing 1% liquid soap (Stevenson *et al.,* 2012). The soap was used to enhance the extraction efficiencies of non-polar chemicals plant's material (Belmain *et al.,* 2012). After allowing the combination to sit for 24 hours, the extract was filtered through a clean cloth.

Various formulations were tested in this investigation; including *T. vogellii* extract (T), rabbit urine (U) and sunflower oil (O) were prepared as stock solutions at concentrations of 10%, 50% and 10%, respectively. To attain the same spraying volume, 100 mL of each ingredient was diluted with distilled water to make 1000 mL water. The formulations O, U, T were applied individually while OUT was obtained in a manner that the concertation of the individual ingredient's solutions would be 10, 50, and 10% combination to make a liter. Dish detergent (liquid soap) was added to the formulation to break the layers between liquid and oil.

3.3 Experimental Setup

The experimental setup was a six-treatment, three-replicate randomized complete block design (RCBD). Using a manual hoe, 3 m × 3 m experimental plots were formed as shown in Fig. 8, with 1 m alleyways between to avoid pesticide drift and inter-plots. Seed variety black-eyed cowpea (*Dolichos melanophthalmus*) locally known as "Kunde nyama" was grown at 50 cm and 20 cm apart and within rows, with two plants per hole at 3-4 cm deep, trimmed to one when plants were one week old from germination. In randomized complete block design, the formulations were applied to the cowpea plots. There were five plant rows and eight plant columns which makes a total of forty (40) plants per plot. Weed removal was carried out with a hand hoe.

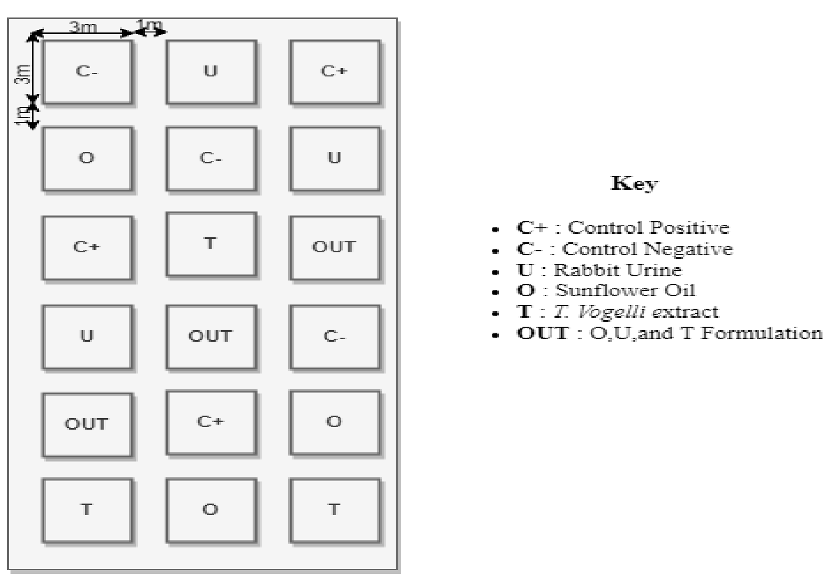


Figure 8: Design layout of plots

3.4 Data Collection

During data collection, targeted cowpea insect pests were collected and identification was done under entomologist. The efficacies of the formulation on cowpea insect pests' abundances, beneficial insects, yield and yield components were assessed.

3.4.1 Assessment of Insect Abundance on Cowpea Plants

The formulation's effectiveness was measured by the reduction in insect abundance and increase in yield. The abundance of insects was measured once per week, a day prior to the next treatment schedule. Eighteen plants were randomly selected from each treatment, and each plant was examined for the targeted insect, and its abundance was recorded. Individually, large insects, leaf miners, and pod borers were counted. Aphid abundance, on the other hand, was measured using a categorical index, in which only big colonies with more than fifty individual aphids were counted (Tembo *et al.,* 2018).

Insect identification was carried out at TPHPA under the supervision of an entomologist. Each treatment was compared to the control positive (synthetic pesticide) Lambda-cyhalothrin and the control negative (with water only).

3.4.2 Assessment of the Effects of Formulation on Beneficial Insects

The presence or absence of pollinator insects was assessed for the randomly selected eighteen plants in each plot. Beneficial insects (bees, butterflies, and ladybird beetles) were identified both before and after the pesticide formulation is applied to ascertain if they are affected or not. Sweep nets were used to gather flying insects. Assessed and recorded insects were the butterflies, bees and ladybird beetle who feeds on aphids.

3.4.3 Assessment of the Efficacy of Formulation on Growth and Yield of Cowpea

To evaluate the influence of the formulations on crop yield, yield parameters such as the number of pods, 100 seed weight, and weight of the yield per plot, expressed in kg/ha, were utilized. For the selected plants, the number of pods was physically counted and recorded. The weight of 100 seeds (g/100 seed) and all seeds per plot (g/plot) was measured and documented after the pods dried under sunlight. Later, the total weight of seed per plot was converted to kilograms per hectare (kg/ha).

The growth parameters recorded were plant's height, leaf's area, and the counts of branches for every plant which were taken since plants were one to four weeks from germination. The measurement was done using a ruler for the height and leaf area (leaf width ×leaf length), and the number of branches was counted manually per plant for the selected plants for each plot.

3.5 Data Analysis

GenStat Statistical program was used and analysis of variance (ANOVA) test was performed to determine differences in insect abundance after data transformation, and yield component among treatments, and Tukey's post-hoc Honestly Significant Difference (HSD) test was employed to differentiate the means at the 95% confidence range.

CHAPTER FOUR

RESULTS AND DISCUSSION

4.1 Results

4.1.1 Insect Pests' Abundance

The results show that there is a significant difference (P≤0.001) in insect abundance reduction. During flowering, the number of pests was low in positive control plots, with mean numbers of (4.3 ± 0.3d, 4.7±0.3a and 5.0a±0.6a), followed by the OUT formulation (11.0 ± 0.6c, 8.0±0.6b 8, and 4.3a±0.3a), compared to the mean abundance of control negative plots (24.7± 0.6a 19.7±1.5e and 12.0±0.6c) for aphid, leaf miner, and pod borer respectively. In general, the OUT formulation had more insect pests than the synthetic pesticides, but fewer than the negative control and individual chemicals (O, U, and T). The abundances for Aphids are listed in Tables 1a&b, whereas leaf miners and pod borers are listed in Tables 2 and 2, respectively.

Table 1a&b: Aphid abundance reduction trend recorded weekly per treatment on cowpea

1a

Treatment	WK9	WK10	WK11	WK12	WK13	WK14	WK15
C-	23.3±2b	24.7±0.7b	34.7±0.7a	37.0±0.6a	34.0±0.6a	37.0±0.6a	36.3±0.9a
C+	4.0±0.6e	5.7±0.9e	5.3±0.3f	6.3±0.3e	4.0±0.6e	4.0±0.6f	3.0±0.6d
O	32.7±0.3a	31.0±0.6a	29.0±0.6b	27.3±1.4b	28.7±1.2b	32.0±1.2b	32.0±1.5a
U	20.3±0.9bc	22.0±0.6b	21.3±0.9c	24.0±0.6b	21.3±0.9c	20.3±0.9c	20.7±1.2b
T	15.3±0.3c	17.0±0.6c	16.7±0.9d	19.0±0.6c	18.3±0.9c	16.0±0.6d	16.3±0.9b
OUT	9.3±0.9d	12.0±0.6d	11.0±0.6e	14.0±0.6d	10.0±0.6d	10.0±0.6e	10.0±0.6c
Mean	17.5	18.9	19.7	21.3	19.4	19.9	19.7
CV	10.7	6.1	4.0	6.0	6.1	6.1	8.8
LSD	3.4	2.1	2.3	2.1	2.1	2.1	3.2
P. value	<.001	<.001	<.001	<.001	<.001	<.001	<.001

1b

Treatment	WK 2	WK 3	WK 4	WK 5	WK 6	WK 7	WK 8
C-	4.0±1.2a	8.6±0.7b	11.0±0.6a	15.0±0.6a	18.3±0.8a	24.7± 0.6a	23.0 ± 0.6b
C+	3.3 ± 0.8a	5.3±0.7a	5.6 ± 0.7c	5.7 ± 0.8d	5.3 ± 0.3d	4.3 ± 0.3d	6.0 ± 0.6e
O	4.0±1.2a	8.0±0.6ab	9.0±0.6ab	12.3±0.8abc	17.0±0.6ab	24.0±1.2a	30.3±0.9a
U	6.7±1.2a	8.0±0.6ab	11.0±0.6a	13.7±0.3ab	14.3±0.7bc	12.7±0.3bc	17.7±1.2c
T	4.0±0.6a	5.3±0.7a	7.6±0.3bc	11.0±0.5bc	12.7±0.8c	14.0±0.6b	16.0±0.6c
OUT	4.6±0.8a	7.0±0.6ab	7.0±0.8bc	10.0 ± 0.6c	12.0 ± 0.6c	11.0 ± 0.6c	11.3±0.7
Mean	4.4	7.1	8.6	11.3	13.3	15.1	17.4
CV	37.1	15.5	11.9	11	7.5	6.4	8.1
LSD	1.9	1.9	1.9	2.2	1.7	1.7	3.4
P.value	0.273	0.013	<.001	<.001	<.001	<.001	<.001

The values presented are means ± SE and significance different at P.value, Means followed by the same letter in a column are not significantly different

Table 2: Analysis of variance on the Abundance of leaf miner recorded weekly per treatment on cowpea

Treatment	WK2	WK3	WK4	WK5	WK6	WK7	WK8	WK9	WK10
C-	4.3±1.2a	4.3±0.9a	7.0±0.6ab	15.0±1.2c	16.0±1.2d	19.7±1.5e	14.0±1.2b	17.0±1.2c	20.0±1.2c
C+	3.0±1a	3.0±0.6ab	5.0±0.6a	4.0±0.6a	3.3±0.3a	4.7±0.3a	4.3±0.9a	3.7±0.7a	5.3±0.9a
O	9.0±0.6b	4.3±0.9ab	10.0±1.2b	16.7±0.9c	15.0±0.6d	11.7±0.9c	15.7±1.5bc	13.3±0.9bc	11.3±1.5b
U	3.7±0.3a	4.7±0.3ab	8.0±1.2ab	10.0±0.6b	13.3±0.3cd	16.3±0.9d	19.0±0.6c	16.7±0.9c	14.0±0.6b
T	5.0±0.6a	5.7±0.3b	5.7±0.9a	8.0±0.6b	10.7±0.3bc	10.0±0.6bc	7.3±0.9a	11.0±0.6b	9.3±0.9ab
OUT	2.7±0.3a	5.0±0.6ab	8.7±0.9ab	11.0±0.6b	8.0±0.6b	8.0±0.6b	8.0±0.6a	5.7±0.9a	6.0±0.6a
Mean	4.6	4.5	7.4	10.8	11.1	11.7	11.4	11.2	11.0
CV	25.9	16.2	18.8	12.9	10.1	9.0	15.2	13.2	13.2
LSD	2.2	1.3	2.5	2.5	2	1.9	3.1	2.7	2.6
P.value	<.001	0.022	0.011	<.001	<.001	<.001	<.001	<.001	<.001

The values presented are means ± SE and significant differences at P.value, Means followed by the same letter in a column are not significantly different

Table 3: Pod borer abundance recorded weekly per treatment on cowpea

Treatment	WK5	WK6	WK7	WK8	WK9	WK10
C-	10.0±0.6c	15.0±0.6d	12.0±0.6c	12.0±0.6c	14.0±0.6e	14.0±0.6d
C+	3.7±0.3a	4.3±0.3a	5.0a±0.6a	2.7±0.8a	3.0±0.6a	2.3±0.3a
O	6.0±0.6b	7.3±0.9c	10.0±0.6bc	11.0±0.6c	10.3±0.9d	10.0±0.6c
U	7.3±0.3b	5.3±0.3abc	5.7a±0.9a	6.0±0.6b	6.0±0.6bc	7.6±0.3b
T	5.6±0.3ab	7.0±0.6bc	6.7±0.9a	5.0±0.6ab	7.7±0.3cd	6.3±0.3b
OUT	5.0±0.6ab	5.0ab±0.6ab	4.3a±0.3a	4.0±0.6ab	3.3±0.3ab	4.0±0.3a
Mean	6.3	7.3	7.3	6.6	7.4	7.3
CV	14.1	11.1	16.6	14.0	13.9	10.3
LSD	1.6	1.5	2.2	1.7	1.9	1.34
P.value	<.001	<.001	<.001	<.001	<.001	<.001

The values presented are means ± SE and significant differences at P.value, Means followed by the same letter in a column are not significantly different

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4.1.2 Beneficial Insects' Abundances

The results showed that the number of ladybird beetles, butterflies, and bees was high in plots treated with OUT formulation compared with those treated with control positive (synthetic pesticides). The results showed a significant difference of $P \leq 0.001$ across treatments indicating the variation between plots treated with bio-based formulation and karate as a synthetic pesticide.

Table 4: The pollinators and ladybird beetles' abundance in cowpea plots recorded during flowering per treatment

Treatment	Butterflies	Bees	Ladybird beetle
C-	5.3±1.7a	6.6±0.6ab	4.6±2.8ab
C+	5.0±2.0a	6.3±0.9ab	4.3±3.2a
O	6.0±1.0a	4.6±2.6a	6.3±1.2bc
U	7.3±0.3ab	6.0±1.2a	7.3±0.2c
T	8.0±1.0ab	8.3±1.0b	10.6±3.1d
OUT	10.3±3.3b	11.7±4.3c	12.0±4.4d
Mean	7.0	7.3	8.0
CV	20.0	10.5	8.3
LSD	3.0	1.3	1.13
P.value	<.008	<.001	<.001

The values presented are means ± SE and significant different at P.value, Means followed by the same letter in a column are not significantly different

4.1.3 Yield and Yield Component

Figure 9 presents the quantity of pods per plant varies depending on the formulation used, with the negative control showing the worst results. The number of pods in the plots sprayed with the OUT formulation, which included 50%, 10%, and 10% of rabbit urine, *Tephrosia vogelii*, and sunflower oil (OUT), respectively, did not differ significantly from the plots sprayed with synthetic pesticide (lambda-cyhalothrin).

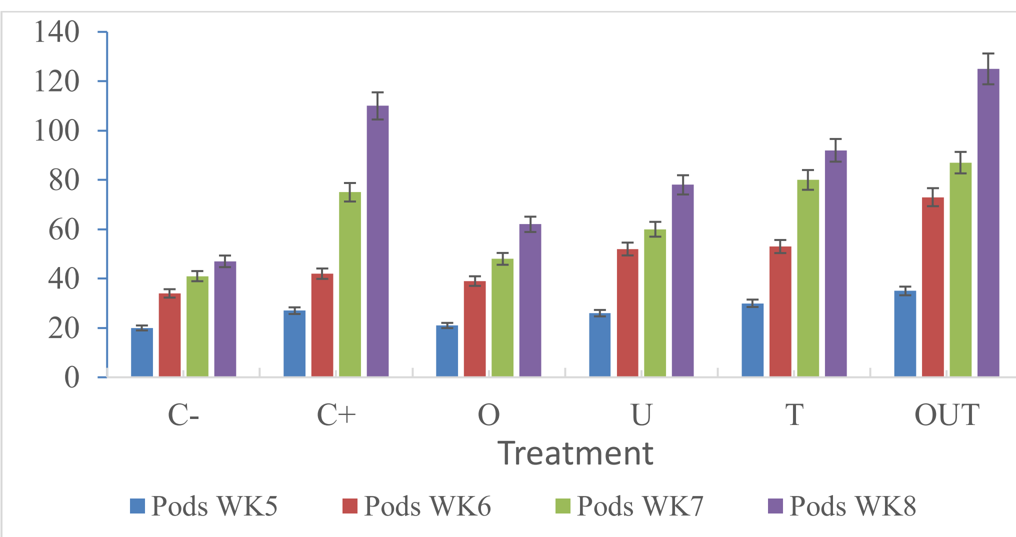

Figure 9: Cowpea's number of pods in weekly bases under different treatment at Singida, Tanzania 2021

Figure 10 shows the results for the 100 seed weight. The 100 seed weight in grams for the formulations under study ranged from 60 to 120 g, with the OUT formulation producing results comparable to the synthetic pesticide, OUT = 118 g/100 and C+= 120g/100. One-way ANOVA revealed significant variations in average 100 seed weight across treatments (C+, C-, T, U, O, and OUT) ($F_{8, 27} = 1269$, $p \leq 0.001$)

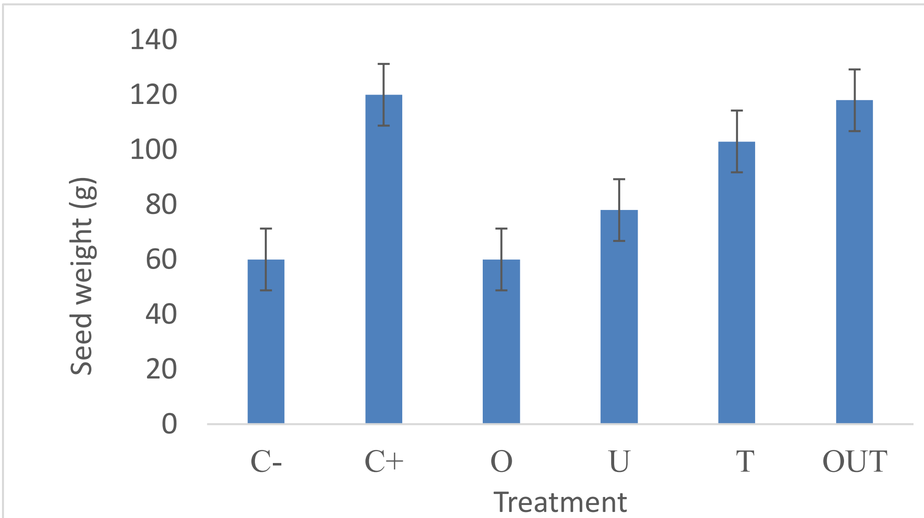

Figure 10: **100 seed weight per treatment under different treatment at Singida, Tanzania 2021**

Figure 11 presenting seed yield in (kg/ha). Results showed that OUT was comparable to the positive control (synthetic insecticides) Fig. 12. The OUT sprayed areas yielded the highest grain yield (846.1 kg/ha), followed by C+ (794.6 kg/ha), and T (662.2 kg/ha), with C- plots yielding the lowest yield (483.1 kg/ha). Although the uncombined treatments O, U, and T outperformed the control negative, they were still well below the synthetic insecticide and OUT formulation. The OUT combination outperformed formulations made from its constituents and other combinations, demonstrating O, U, and T synergy. Although the abundance of insect pests on crops treated with the OUT formulation was much higher than the synthetic control, crop production parameters showed comparably equal results.

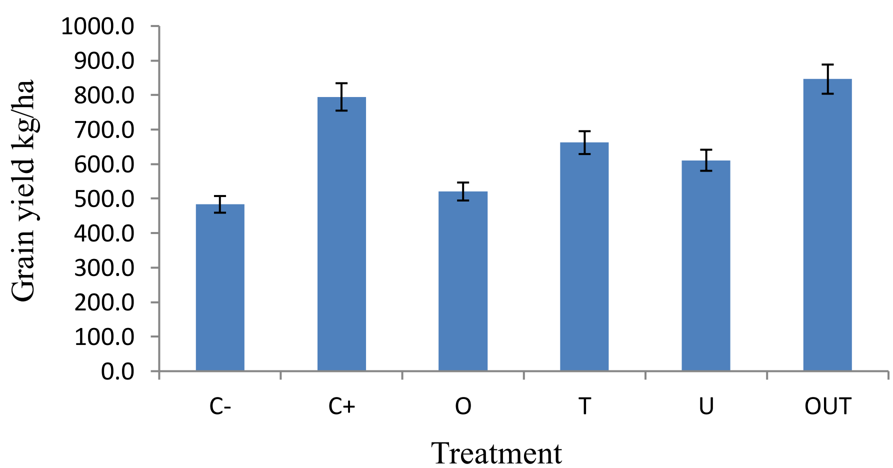

Figure 11: **Cowpea yield in kg/ha under different treatment at Singida, Tanzania 2021**

Figure 12 presents plant's height as one of the parameters determining growth promotion influenced by the formulation, the plant's height, were recorded for four weeks consequently. Selected eighteen plants from each plot were used as a sample to represent all the plants in specific plots. Results showed the plants treated with combined formulation OUT giving the differences in height from week one to week four ranging from 8 cm to 36 cm, followed by positive control 8 cm to 24 cm compared to negative control and individual applied formulation T, U and O.

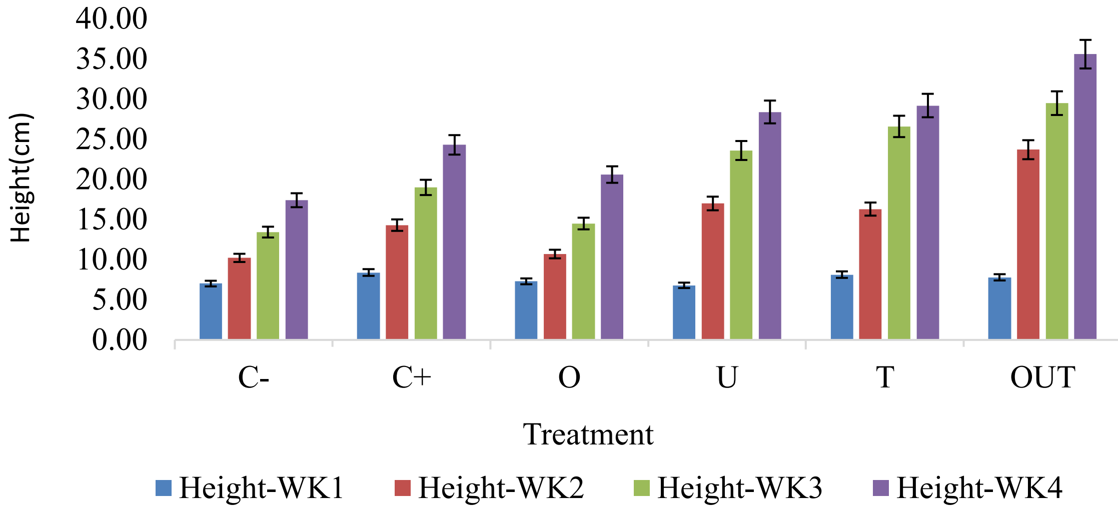

Figure 12: **Cowpea Plant's height in weekly bases under different treatment at Singida, Tanzania 2021**

Figure 13 shows plant's leaf area (leaf width×legth). Difference in leaf area recorded for four weeks consequently showed the combined formulation giving large leaf area compared to control positive and other uncombined treatment.

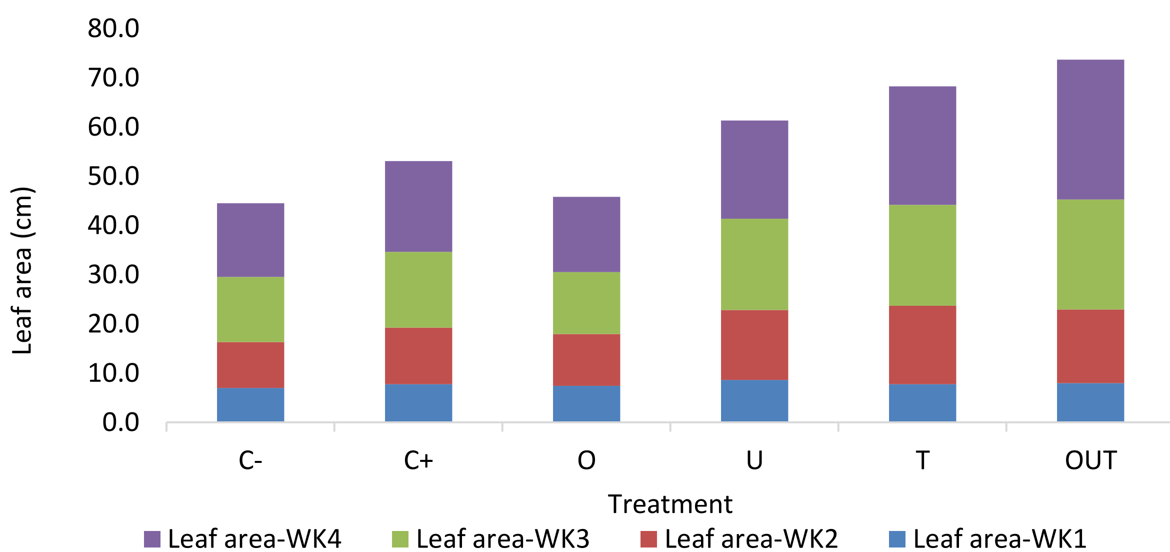

Figure 13: **Cowpea plant Leaf area in weekly bases under different treatment at Singida, Tanzania 2021**

Figure 14 presents number of branches from the selected eighteen plants per plot. From the results the number of branches differ across treatments where by OUT formulation giving higher number of branches compared to uncombined formulation and control positive synthetic pesticide.

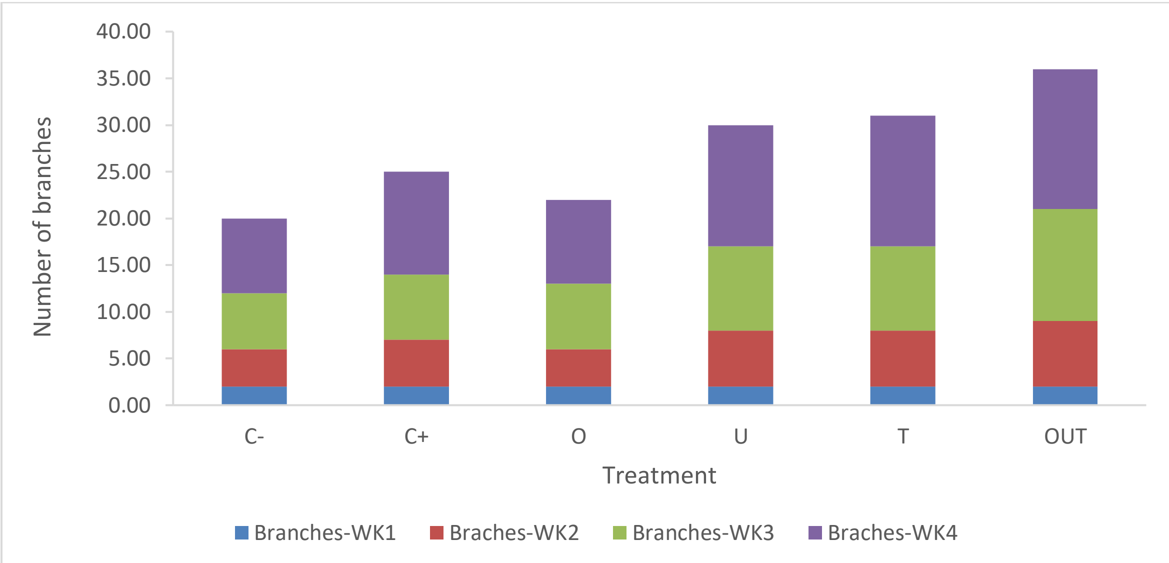

Figure 14: Cowpea's number of branches in weekly bases under different treatment at Singida, Tanzania, 2021

4.2 Discussion

4.2.1 Insect Abundance

The results of this investigation showed that mixing *T. vogelii* extract, rabbit urine, and sunflower oil to generate an anti-insect immersion (with a detergent) improves performance synergistically when compared to uncombined formulations. The number of times performance improves when the combination formulation is used demonstrates the synergistic effect. The OUT reduced aphid, pod borers, and leaf miner abundance by up to 2-3 times when compared to the control negative. Despite the gains, the control positive formulations performed somewhat better (lower aphid abundance) than the OUT formulations and much better than the uncombine formulations. Previous research has found significant differences in the decrease of insect pests on cowpeas between the positive control and botanical extract formulations (Tembo *et al*., 2018). The diverse mechanisms of action of the many bioactive components in the OUT formulation could explain its better efficacy in this investigation study.

While Oil suffocates insects by plugging spiracles, rabbit urine contains ammonium chemicals that give it a distinct strong repellency (Mohammed, 2016), and insects are repelled by this odour. According to Corbeels *et al.* (2019), fermented rabbit urine spray has been shown to help control insects and other pests such as aphids, moths, leaf miners, caterpillars, and mites. A phytochemical study of *T. vogelii*, on the other hand, revealed the existence of the rotenoids deguelin, tephrosin, and rotenone with deguelin being the most common (Stevenson *et al.,* 2012). Retinoids are the only flavonoids that are known to be very poisonous to insects (Mkindi *et al.,* 2019). Rotenoids impede the mitochondrial oxidation of Krebs cycle intermediates by interfering with electron transport in the oxidation of NADPH to NADP+ by cytochrome b. This reduces the cellular respiration of insects. When rotenone is applied to insect cells, oxygen absorption drops dramatically (Zhang *et al.,* 2020).

Individual plants in *T. vogelii* may differ greatly in rotenone, deguelin, rotenone, and tephrosin content due to various chemotypes (Belmain *et al.,* 2012). The chemotype 1 *T. vogelii* plant contains insecticidal properties, however, the flavonoids in chemotype 2 were ineffective against insects (Mkindi *et al.,* 2019). In other studies, T. vogelii was shown to have a significant difference when used alone at 10% concentration, in this study, demonstrating efficacy in conjunction with sunflower oil and rabbit urine in terms of minimizing insect damage and boosting yield (Karungi *et al.,* 2000; Mkindi *et al.,* 2017).

The decreased abundance seen with the positive control could be related to lambda-strong cyhalothrin's affinity for its receptor and the high efficiency of its method of action. Lambda-cyhalothrin works by entering the insect cuticle and disrupting neuronal conduction by keeping the sodium channel activation gate open, causing nerve fiber overexcitation, paralysis, and death (Altieri, 2002). This study is in line with the study done by Olaitan and Abiodun (2011) who prove the combined use of two plant extracts, *Tephrosia vogelii*, and *Petiveria alliacea*, as a pesticide to dramatically reduce the population of insect pests when compared to the control. While according to other studies (Tembo *et al.,* 2018), the efficacy of combining *Lippias javanica*, *T. vogelii*, and *T. diversifolia* in reducing insect pest count on cowpea was found to be comparable to synthetic pesticides, and this study's results showed that the efficacy of combining *T. vogelii*, sunflower oil, and rabbit urine on insect pests' management was found to be comparable to the synthetic pesticide.

4.2.2 Beneficial Insects

This study revealed the less detrimental effects of formulation on beneficial insects /pollinators during flowerings. Research by Tembo *et al.* (2018) reported the fewer effects of botanical pesticides on spider and ladybird beetles which is in line with this study. The pollinators like bees and butterflies contributed to the high yield of cowpea (Latif *et al.,* 2019). The existence of the ladybird beetle in plots sprayed with a bio-based formulation indicates the presence of an ample amount of pollen during flowering due to the preservation of beneficial insects. The ladybird beetles feeding on flower pollens is a sign of an abundance of flowers produced hence the higher number of pods produced (Riddick, 2017). This study is in line with the study done by Singh *et al.* (2017) reported that the abundance of bees and houseflies resulted into a significant difference in all quantitative parameters including a number of pods and seed weight.

4.2.3 Yield and Yield Components

Although plots treated with OUT formulation exhibited slightly high insect abundance compared to the synthetic treatment, cowpea grain yields were equivalent. Despite the slightly high insect abundance on plots treated with OUT formulation compared to the synthetic treatment, cowpea grain yields were equivalent. The yield metrics of the OUT treated plots, like the pods' counts, 100 seed weight, and grains' yields in kilograms per hectare, are comparable to those of the positive control. Having rabbit urine as one of the formulation ingredients has added effects to the comparable yield due to its nutrient composition. According to Mutai *et al.* (2020), rabbit urine contains potential nutrients such as 1.05% nitrogen, 0.01% phosphorus, 0.85% potassium, and 0.12% calcium, which has a superior nutrient composition than even commercial foliar-fed fertilizers. Furthermore, OUT's capacity to retain the diversity of beneficial insects necessary for pollination may have contributed to its comparable yield despite higher insect abundance (Wezel *et al.,* 2014). This study's findings are consistent with those of Alao *et al.* (2011) that reported that when compared to positive control and singly applied formulations, the combination of *Tephrosia vogelii* and *Petiveria alliacea* at 10% reduced pod damage and boosted grain quality.

Apart from insect pest control, a large number of pods in the OUT formulation is attributable to the contribution of plant's extracts to plant's nutrition as a foliar fertilizer; thus, its application in crop protection has maintained crop production as supported by Mkindi *et al.* (2017) and Kayange *et al.* (2019), reported that sprayed plots with *T. vogelii* and *T. candida* had the highest pod number at higher extraction concentration.

CHAPTER FIVE

CONCLUSION AND RECOMMENDATIONS

5.1 Conclusion

This study examined the potential of combining *T. vogelii* extract, rabbit urine, and sunflower oil. As a result of the synergistic impact, mixing *T. vogelii* with rabbit and sunflower oil at 10%, 50%, 10%, respectively, boosts pesticidal effectiveness. Despite somewhat higher insect abundance on crops treated with the OUT combination than on crops treated with synthetic insecticides (karate), crop yields were equal, indicating that the OUT combination is a viable alternative to synthetic pesticides. The study showed that botanical extract mixtures might be used to develop synergistically improved biopesticides for ecologically friendly insect-pest management with less detrimental effects on pollinators, hence to be used in the broader context of agro-ecological farming.

5.2 Recommendations

From the outcome of this study, it is recommended that:

(i) Further studies should be conducted to investigate the efficacies of the *Tephrosia*-based formulation at different concentrations for insect management on cowpea.

(ii) The effects of the *Tephrosia*-based formulation on other field crops should be studied.

(iii) Studies have to be done in different regions to assess the efficacy of the formulation on cowpea insect pest management on different climatic conditions.

(iv) Further study has to be done on the cost benefit analysis of the formulation to be used by small scale farmers for agro-ecological farming.

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RESEARCH OUTPUTS

(i) Publication

Agricola, M., Paul, K., Mashamba, P., & Ernest, R. M. (2022), Effects of *Tephrosia vogelii* and rabbit urine formulation on insect pests and yields of cowpea Singida, Tanzania. *Journal of Biodiversity and Environmental Sciences,* 3, 63-70.

(ii) Poster Presentation