# DOLICHOS (*Lablab purpureus* L.) GENOTYPES AND FIELD MARGIN VEGETATION EFFECTS ON BEAN APHIDS AND THEIR NATURAL ENEMIES

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**EGERTON UNIVERSITY**
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## DECLARATION AND RECOMMENDATION

### Declaration

This thesis is my original work and has not been presented in this university or any other for the award of a degree.

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### Recommendation

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# DEDICATION

This thesis is dedicated to my late mother, Judith Karimi, my late son, Crown Jasiri Mutugi, and my wife, Sophida Wairimu Mwangi, to whom I acknowledge my success to. They have been a valuable resource and consistent source of inspiration in my social and educational pursuits.

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# ABSTRACT

Dolichos bean (*Lablab purpureus* L.) is a drought tolerant multipurpose legume used for food/feed and soil fertility management. Despite these benefits, dolichos remains underutilized and unexploited in terms of area under cultivation and efforts towards its genetic enhancement particularly resistance towards insect pests and diseases.There is need for selection of tolerant/resistant genotypes that can be integrated with other natural pest regulation strategies for the management of insect pests specifically the bean aphid. A field and cage exclusion experiments were conducted at the Agronomy Research and Teaching field of Egerton University, Njoro campus, during the 2019 and 2020 crop growing seasons to determine the effect of dolichos genotypes and field margin vegetation (FMV) on abundance and diversity of natural enemies. The treatments consisted of 18 dolichos genotypes which were planted in the presence or absence of margin plants. The margin plants consisted of four weed species: black jack (*Bidens pilosa* L.), Mexican marigold (*Tagetes minuta* L.), goat weed (*Ageratum conyzoides* L.) and gallant soldier (*Galinsoga parviflora* Cav.). The treatments were laid out in a randomized complete block design (RCBD) with four replicates. Data collected on aphid abundance, percent incidence and damage severity, natural enemy populations, yield and yield components of dolichos were subjected to analysis of variance using Statistical Analysis System (SAS) version 9.2. Results showed significant seasonal variations in bean aphid infestation levels ($P<0.001$). Further results showed significant interactive effects between dolichos genotypes and FMV on aphid abundance and percent incidence ($P<0.001$). The presence of FMV showed, low aphid abundance (1.24) in 2019 cropping season compared to the 2020 (2.59) season. The lowest aphid abundance was observed on genotypes Machakos I (1.76) and Brown Rongai (1.75). Among the natural enemies captured, the most abundant were Tachnid flies (6.19), hoverflies (1.0), parasitic wasps (2.93) and ladybird beetles (1.07).Genotype Echo-Cream had low hoverfly abundance while Machakos-II had high abundance of ladybirds across the two seasons. Genotype Brown - Rongai had the highest yield of 0.97 t/ha while Machakos - I and HA-4 had the lowest yields of 0.19 t/ha in the two seasons. These findings highlight the potential of integrating bean aphid tolerant dolichos genotypes and field margin vegetation in suppressing bean aphid infestation and supporting natural enemies of bean aphids in dolichos production

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# LIST OF ABBREVIATIONS AND ACRONYMS

| | |
|---|---|
| ANOVA | Analysis of variance |
| DAG | Days after germination |
| EC | Emulsifiable concentrate |
| FMV | Field margin vegetation |
| GAPs | Good agricultural practices |
| Ha | Hectare |
| NEs | Natural enemies |
| RCBD | Randomized Completely Block Design |
| WP | Wettable powder |

# CHAPTER ONE

# INTRODUCTION

## 1.1 Background information

Dolichos (*Lablab purpureus* L.) also known as hyacinth bean is a multipurpose crop grown for food, forage, soil improvement and cover crop. The young leaves and pods are consumed as vegetables and fodder while the dry grains are consumed as pulse and livestock feed (Ewansiha *et al*., 2016;Habib *et al*., 2017). It is a rich source of proteins, minerals and vitamins (Trinidad *et al*., 2010). It is also used in traditional cropping systems due to its nitrogen-fixing capacity, cover crop for or green manure (Kouris-Blazos & Belski, 2016). Dolichos thrives under diverse conditions including arid, semi-arid and humid areas making it suitable for climate smart agriculture for improved food and nutritional security and rural livelihoods.

Despite all these benefits, dolichos is regarded as an orphaned crop that remains neglected in terms of utilization, area under cultivation and efforts towards its genetic enhancement (Khoury *et al*., 2014;Maass *et al*., 2010;Massawe *et al*., 2015;Vaijayanthi *et al*., 2019). In Kenya, the grain yield of dolichos ranges between 0.8 to 0.9 tonnes ha$^{-1}$ compared to the yield potential of 3 tonnes ha$^{-1}$. The low yield is attributed to pests such as mites, aphids pod borers and sucking bugs (Letting *etal*.,2022) among other factors. Aphids are among the most serious insect pests on dolichos causing yield loss of up to 37% bean (Munyasa, 2013).

Several aphid species have been reported in dolichos where the nymphs and adult aphids suck sap from leaves, petioles inflorescences tender stems, and pods (Blackman & Eastop, 2000). The affected leaves curl and the crop remains stunted, but in severe infestations, the affected crop withers and dries up (Sharma *et al*., 2010). Abundance and infestation of aphid are correlated to characteristics of host plants and prevailing weather patterns (Amin *et al*., 2017). Aphid infestation level varies among the crop species and different growth stages of the same species.

The use of synthetic pesticides to reduce aphid damage and yield losses caused by insect pests is widespread among smallholder farmers (Stevenson *et al*., 2017) because they are readily available and considered affective. The over-reliance, continuous and indiscriminate use of synthetic insecticides poses serious risks such as development of insecticidal resistance, resurgence of pests, environmental pollution and disruption of the eco-system equilibrium (Chopkar *et al*., 2020). These created the impetus for research to explore the use of environmentally friendly approaches such as bio pesticides, host plant resistance and biological control (Jahan *etal*., 2013).

There is an urgent need for insect repelling and insect-resistant genotypes to reduce the number of insect pests as well as increase their tolerance to injury (Kinyua *et al*., 2008). Rizwan *et al.* (2021) found out that genotypes with high soluble sugars and proteins content were more susceptible to the sucking insect pests. Tolerance is a resistance where a plant can resist or recover from damage caused by the pest population (Smith, 2007). According to Mondal *et al*. (2017) aphid ingestion of sufficient volume of phloem sap was limited in genotypes with less water content of vine and leaflet, turgor pressure in the vascular system, and a high compaction of the vine, and there was an antibiotic effect of phloem sap that discouraged aphid fecundity.

Biological control on the other hand involves natural enemies (NEs) that parasitize or predate on insect pests (Bale *et al*., 2008; Gurr *et al*., 2018). The use of natural enemies for biological control is long-term and cost-effective compared to conventional methods (Amoabeng *et al*., 2021). Non-crop plants support NEs by providing nectar and pollen for efficient biological control (Kishinevsky *et al*., 2017). Inclusion of field margin plants can therefore maximize natural enemies' contribution to pest management. Pesticidal plants such as *Tagetes spp*, *Capsicum frutescens, Lantana camara, Tephrosia vogelii* have been reported to exhibit strong anti-insect properties (Mugisha-Kamatenesi *et al*., 2008; Sharma, 2007). The effectiveness of these plants is due to chemical compounds which have toxic and repellant properties to insect pests and attract natural enemies (Karani, 2017). Field margin vegetation is more economical method of controlling aphids and a good practice for conservation of natural enemies and pollinators (Mkenda *et al*., 2019). Field margin vegetation (FMV) creates a niche for supplementary food, multiplication and refuge of natural enemies which are key components of natural pest regulation (Forsythe, 2019). Previous studies have shown the influence of field margin plants on different natural enemies including syrphids, tachinids and spiders (Amaral *et al*., 2016; Fusser *et al*., 2018). In an experiment conducted to determine the importance of field margin vegetation to natural enemy populations, results showed that field margin vegetation harbored more natural enemies than insect pests implying that the margin plants have an important role in enhancing biocontrol agents for conservation biological control (Mkenda *et al*., 2019). Ndakidemi *et al*. (2022) evaluated how field margin plants support natural enemies in common bean and revealed that in cage experiments, the most NEs were observed interacting with *Euphorbia heterophylla* than *Bidens pilosa*, *Tagetesminuta*, and *Hyptissuaveolens*, implying that field margin plants help natural enemies by providing nectar and pollen. The study also found out that, lady beetles and assassin bugs were most abundant in plots with *Bidenspilosa* margins, hoverflies in plots with *Tagetesminuta* and *Partheniumhysterophorus* margins, and lacewings in plots with *Bidenspilosa* borders. The loss of natural habitats surrounding croplands, natural enemies are forced to disperse from decreasing and more distant non-crop reservoirs.

Host plant resistance, along with natural enemies, are an important component of pest management in various ecosystems (Sharma, 2007). However, combining the two strategies necessitates a thorough evaluation of their individual and combined effects on insect pests and natural enemies, as well as their overall impact on crop yield. There is mounting evidence that the qualities of pest-infested plants influence the behaviour of natural enemies. Plant defense traits can influence both the numerical and functional responses of natural enemies (Mitchell *et al*.,2016; Pappas *et al*.,2017; Peterson *et al*.,2016.). This study aimed to investigated the potential contribution of genotypes and natural field margin vegetation on crop damage and abundance of bean aphids and their natural enemies in dolichos.

### 1.2 Statement of the problem

Dolichos is an important crop with diverse benefits that include human, animal feed and improvement of soil health. Common production constraints of dolichos are susceptibility to pests and diseases which lower the yield. nutrition and economic value of the crop. The black bean aphid, (*Aphis fabae* S.) is among the major pests that attack the dolichos crop and contribute to significant yield losses. Aphids are multivoltine insects with high reproduction and dispersal capacity hence can rapidly increase their incidence and damage severity on crops. They are also important vectors for several disease-causing viruses in crops. Small-holder farmers often rely on the application of synthetic insecticides to manage these pests. Although the use of synthetic insecticides is the most effective and commonly used strategy, it is not cost-effective and ecologically sound. Pesticides pose high risks of developing insecticide resistance, adverse effects on non-target organisms and environmental pollution. The use of aphid-resistant genotypes and conservation of natural enemies using non-crop habitats in the form of field margin vegetation provides a viable natural pest regulation among the smallholder farmers.

## 1.3 Objectives

### 1.3.1 Broad objective

To contribute to improved food and nutrition security through the integration of host plant resistance and conservation of natural enemies using field margin vegetation in the management of aphids in dolichos lablab beans.

### 1.3.2 Specific objectives

i. To determine the effect of dolichos genotypes and field margin vegetation on bean aphids' abundance, incidence and severity on dolichos.
ii. To determine the effect of dolichos genotypes and field margin vegetation on the abundance and diversity of natural enemies of bean aphids infesting dolichos.
iii. To determine the effect of dolichos genotypes and field margin vegetation on growth yield and yield components of dolichos.

## 1.4 Hypotheses

i. Field margin vegetation and dolichos genotypes have no significant effect on the bean aphid abundance, incidence and severity on dolichos.
ii. Field margin vegetation and dolichos genotypes have no significant effects on the abundance and diversity of natural enemies of bean aphids infesting dolichos.
iii. Field margin vegetation and dolichos genotypes have no significant growth, yield and yield components on dolichos.

## 1.5 Justification

Climate change poses a significant challenge for the attainment of Sustainable Development Goals (SDG) 2 which aims at zero hunger by 2030. Orphan crops possess inherent properties of stress tolerance and nutrition content and can therefore offer solutions to boost the sustainability of agriculture and ensure food and nutritional security for future generations Dolichos bean is among the orphaned legume crops that is drought tolerant and highly nutritious and can be a major source of cheap proteins for human food and animal feed. Its production however remains neglected particularly in research and development.

Dolichos production is highly constrained by its susceptibility to insect pests that lower the quality and quantity. A review study by Khan *et al*. (2020) revealed that the adverse effects by harmful insects, reduces the lablab bean production by 20-45%. The bean aphids are among the major insect pests of dolichos can cause yield loss of about 37% to 100% (Khan *et al*.,2018; Ochilo *et al*., 2011). Over-reliance on insecticide and particularly contact foliar applications have been considered to be ineffective because of the feeding nature of the aphid and are also detrimental to natural enemies. There is an urgent need for a concerted efforts to undertake research and development that will identify sustainable pest management for the management of bean aphids in legumes including dolichos. Host plant resistance and conservation biological control are two methods that have attracted much attention as alternatives to synthetic pesticides and component of integrated pest management programmes (Mandal *et al*.,2018).

Despite significant efforts in recent years to understand the mechanisms of insect resistance in grain legumes, as well as screening and selecting aphid-tolerant and aphid-resistant genotypes, there has been limited success with lablab (Kujur *et al*., 2017). While lablab is recognized for its genetic diversity, there are similar impediments to widespread adoption of insect-resistant cultivars in other legumes (Maass *et al*., 2010; Whitbread *et al*., 2011). Inadequate seed production and distribution efforts, as well as a lack of investment in research and development, are among the obstacles. Identification and selection of genotypes that not only withstand aphid attacks but also attract natural enemies will offer a cheaper and sustainable ecologically friendly alternative

The use of natural enemies as for biological control is more long-term and cost-effective as compared to conventional methods (Amoabeng *et al*., 2021). As a result of the loss of natural habitats surrounding croplands, natural enemies are forced to disperse from decreasing and smaller and more distant farther out non-crop reservoirs. Field margin vegetation (FMV) creates a niche for supplementary food, the multiplication and harbouring refuge of natural enemies which are key in components of natural pest regulation (Forsythe, 2019). Sustainable alternatives that are compatible with natural enemies for managing bean aphids such as use of biopesticides, bioinsecticides and highly selective and reduced application of biological and synthetic insecticides pesticides have been demonstrated (Halder & Srinivasan, 2011; Shinde & Narangalkar, 2018; Stevenson *et al*., 2017).

The use of aphid-resistant genotypes and non-crop habitat manipulation can help to conserve natural enemies. However, the individual and combined effects of the two strategies on insect pests and the associated natural enemies and crop yield requires thorough evaluation. This study investigated the additive effects and potential contribution of dolichos genotypes and field margin vegetation to suppress bean aphid infestation, support natural enemy population and improve the yields of dolichos.

# CHAPTER TWO

# LITERATURE REVIEW

## 2.1 Botany of dolichos(*Lablab purpureus*)(L)

Dolichos bean is an herbaceous legume that belongs to Fabaceae family and is commonly known as Field bean, Hyacinth bean, Indian bean and "Njahii" in Kenya, grown as an annual crop. Dolichos belongs to the family of Fabaceae, genus Lablab and species purpureus (Al-Snafi, 2017). It is a bushy, semi-erect, climbing and twining plant that has a tap root which have many laterals and adventitious roots that are well developed (Maass *et al*., 2010). The semi-erect type has thick stem that can grow up to 3 feet while the climbing type has cylindrical stem which twins to a length of 6 meters. However, some forms of dolichos bean stems are dwarf and bushy. Lablab leaves are alternate and trifoliolate which are hairy, with ovate leaflets that are 5-15cm by 4-15cm in size. The upper surface is smooth while the underside has short hairs (Moteetee & Van, 2012).

It produces pink, purple, or white-coloured flowers, in groups of between four and five. The plant produces pods that are variable in shape usually flat, inflated, smooth, pubescent, and papery. The pods are also crescent-shaped and can be either green, purple, or whitish, with their length being at least 5cm to 20cm. Each pod contains 3 to 6 seeds, which vary in their sizes and colour. Seeds can be white, cream, pale brown, dark brown, red, black, or mottled depending on the variety. The hilum is white approximately 10mm long and 7mm wide.

## 2.2 Importance of dolichos bean

Dolichos lablab is grown as a pulse crop as well as a vegetable crop (Mondal *et al*., 2017) mainly in Africa, Asia, and the Caribbean (Moteetee & Van, 2012). It is harvested as a pulse crop for dry seed, while the green pod is consumed as a vegetable (Muthomi *et al*., 2014.It is used for human consumption and livestock feed in various parts of the world (Ewansiha *et al*., 2007). In Asian countries, it is used as a source of proteins in diets cooked in various forms as green pods, dry seeds and leaves as green vegetables (Miah *et al*., 2017; Sarma *et al*., 2010). Dolichos seeds are cooked with maize, crushed and fried, or added to soups in Africa. In Kenya, the *Kikuyu* community use dolichos seeds to prepare a traditional dish called *mukimo*, which is eaten on important occasions such as weddings, circumcisions (Kilonzi *et al*., 2017). Dolichos has a high protein content of 31.3% hence stimulates milk production in lactating mother (Al-Snafi, 2017).

Dolichos lablab leaves are palatable hence used as forage, hay and silage. The leaf protein content varies from 21% to 38% and the seed protein ranges between 20% to 28% (Maass *et al*., 2010). The crop does not only enrich the soil in nitrogen but it has deep roots which brings to the surface other elements which have not been available to annual crops. In an experiment conducted to determine the effect of green manure on maize yield showed that a plot on which dolichos had been grown the previous year and incorporated into the soil yielded 4.15 kg ha$^{-1}$ against 2.42 kg ha$^{-1}$ for same plot without green manure. Dolichos lablab is a drought tolerant crop because it has taproot system which can penetrate to more than 2 metres below the soil surface thus enabling it to sustain growth on residual soil moisture (Pengelly & Maass, 2001).

Lablab produces 2,600 kg ha$^{-1}$ of biomass with an average nitrogen content of 64 kg ha$^{-1}$ (Ewansiha *et al*., 2007) hence effectively used as a green manure to improve soil fertility alongside nitrogen fixation through its root nodules (Karuma *et al*., 2011). Mendonça *et al*. (2017) compared the effect of different legume crops on biological nitrogen fixation and found dolichos contributed more to nitrogen fixation and consequently uptake by coffee plants. Similarly, Ewansiha *et al*. (2012) found out that intercrop of maize and dolichos bean resulted in 70% more grain than soil alone. This difference in yields was attributed to improved soil organic matter supply and increased nitrogen, phosphorus and potassium content. Most dolichos varieties have high potentials of improving livestock feed and generate residues that promise to be dominant crop-livestock integration resources (Smith *et al*., 1997). Ogedegbe *et al*. (2016) evaluated the performance of several herbaceous legumes as potential cover crops found that dolichos was the second most crucial soil protecting crop after *Mucuna pruriens.* These studies show that adequate dolichos incorporation is useful in ensuring the provision of essential nutrients for human and livestock production as well as soil fertility improvement (Mallikarjuna *et al*., 2021).

## 2.3 Ecological requirements

Dolichos bean is adapted to wide areas under diverse climatic conditions such as arid, semi-arid and humid regions(Morris, 2009). It is a drought tolerant legume adaptive to a range of climatic conditions with temperatures ranging from 18°C to 30°C, however it can grow at low temperatures of 3°C for short periods (CABI, 2019). Lablab is frost susceptible, but tolerates very light frost is known to damage the leaves, although they do not kill the plant entirely (Maass *et al*., 2010). The rainfall requirement ranges between 750 mm to 2500 mm per annum with rainfall <500mm the crop will grow but loses leaves during prolonged dry periods. The crop grows optimally under shady or water stressed conditions with minimum rainfall averaging 500-800mm per annum. However, it cannot stand in waterlogged areas. Soil texture, as long as drainage is good, the crop is able to grow in all types of soils (CABI, 2019). The plant does well in soils with a pH range of between 5 and 7.8 and does well in low-altitude regions. Dolichos bean is a short-day plant because requires long periods of darkness for its growth and development Compared to cowpea (*Vignaunguiculata*) and common beans, it is considered to be more drought resistant (Maass *et al.*, 2010).

## 2.4 Dolichos beanproduction

Dolichos bean is grown in small regions of East Africa (Uganda, Tanzania and Kenya) and West Africa - Cameroon and Nigeria (Forsythe, 2019). There has been a significant reduction in production when farmers reported approximately 10% less land area dolichos (Raghu *et al*., 2018). In Kenya, dolichos production is for domestic consumption with imports from Tanzania to bridge the gap (Boit *et al*., 2018). However, the exact amount of dolichos being produced in Kenya remain unknown due to informal, undocumented trade and is estimated to be only 20, 000ha with 0.4 tons/ha of grains with low domestic demand (Forsythe, 2019). The market survey revealed that the low demand is due to changing eating habits, long cooking times of grains and poor palatability of the common black seeded variety (Ngure *et al*., 2021). Dolichos is mainly grown in the Eastern, Rift Valley, Coast, and Central parts of Kenya, (Nahashon *et al*., 2016) in regions that receive 750 mm to 2500 mm rainfall per annum (Esther *et al.*, 2012).

## 2.5 Production constraints

The production of lablab has been on a decline due to biotic factors, variation in climate patterns, poor marketability, less awareness of its nutritional value, poor seed quality and poor storage facilities (Ogedegbe *et al*., 2012). Lablab production has been affected majorly by biotic factors resulting in low production and consumption. Insect pests such as pod borers (*Helicoverpa armigera* and *Maruca vitrata*), aphids (*Aphis sp.*), sucking bugs, leaf miners, flower thrips and mites affects lablab normal growth consequently resulting in reduced yield (Forsythe, 2019; Kamotho, 2015; Khan *et al.*, 2018; Nahashon *et al*., 2016). Previous studies have reported that bruchid (*Callosobruchu ssp*.) beetles are the major storage pest which leads to a yield loss of 90% of the stored legume grain (Chawe *et al*., 2019; Ebinu *et al*., 2016). Several studies have reported that farmers cultivating lablab obtain yields of 800-900 kg ha$^{-1}$, which is extremely below the expected yield potentials of 2700-

3000 kg ha$^{-1}$ due to pest infestations (Kinyua *et al*., 2008). This has led to preference shifting to alternative legumes common beans and French beans, which cannot survive under extreme environmental conditions (Reddy *et al*., 2017).

Aphids are the most destructive pest on dolichos lablab because the nymphs and adults suck the cell sap from tender shoots, inflorescence and pods resulting in yellowing, curling and drying up of tender pods (Golvankar *et al*., 2019). The pest attacks the dolichos bean from seedling to maturity stage, with the highest population seen at flowering to the pod formation stage (Golvankar *et al*., 2019). Cowpea aphid infestation exude honey dew which spreads on the leaves and twigs and produces black sooty mould caused by fungus *Capnodium spp.* which impedes photosynthesis thus reducing yield (Soffan & Aldawood, 2014). Apart from reduction of yield, infestation by the black bean aphid (*Aphis fabae*) stunt the growth of faba bean (*Vicia faba*) leading to a decrease in shoot fresh and dry weight, leaf area, and plant height. In an experiment conducted to determine incidence of aphids on dolichos lablab variety HA-4 the results revealed that nymphs and adults of coreid bug, *Riptortus pedestris* were observed at later stages of lablab growth and were found sucking the sap from the pods resulting to sap sucking, brown spots being observed on the pod and shriveling of seeds (Prasad *et al*., 2011). In a different study pod borer, *Helicoverpa armigera* bored into the flower buds and tender pods while the later instars damaged mature and developing pods making circular holes on pods and feeding on individual seeds of pod leading to pod damage of 20.43% on 80 days old crop.

## 2.6 Overview of aphids

### 2.6.1 Aphid biology

Aphid species can sometimes demonstrate higher reproductive patterns; they may vary from gradual to compulsory sexual reproduction. In aphids, the ancestral lifespan, gradual parthenogenesis, involves numerous lifetimes of apomicic parthenogenesis by dioecious females consisting of a single sexual progeny composed of males and egg-laying females (Dixon, 2019). Nevertheless, transformations to compulsory parthenogenesis which is reduced sexual progeny are prevalent, and both genetic modalities may coincide in the same organisms or in the same community (Dedryver *et al*., 2001; Delmotte *etal*., 2001; Vorburger *et al*., 2003).

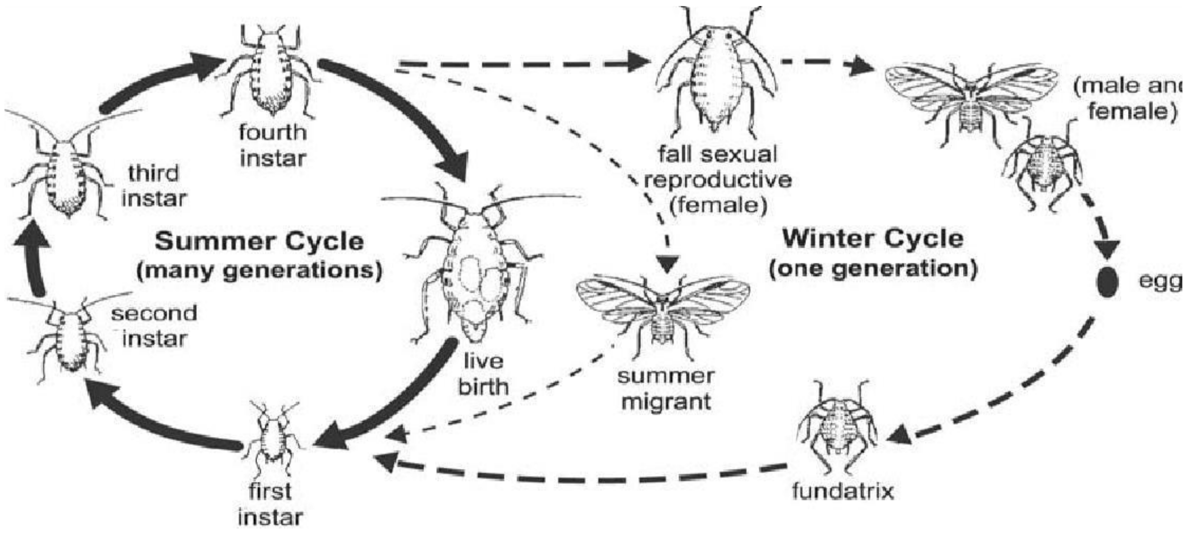

**Figure 2.1:** Aphids lifecycle. Source: Kang *et al*. (2008).

### 2.6.2 Aphid Ecology and foraging behaviour

Aphids (*Aphis fabae*) belongs to the family Aphidae. Aphids are characterized by a wingless body with small to medium size, pale yellowish, green body color. The winged aphids are characterized by; a black patch in the dorsal part of their abdomen. Their sizes vary from 1.0 to 2.0 mm length (Blackman & Eastop, 2000). Aphis use various visual cues to locate the host plant. The winged aphids use their visual cues while flying to locate the plants. During their flight, the winged aphids use preferential colour displays to patch on yellow surfaces while most polyphagous species use responsive cues to the green wavelength (Nottingham *et al*., 1991). Besides, the aphids use semio-chemicals in the nature of olfaction's to detect the host plants. They are capable of detecting volatiles from the plants such as the isothiocyanates, nitriles, monoterpenes, benzaldehydes and green leaves volatiles (Johnson & Agrawal, 2005).

Aphids use the genetic information from the plants like pubescence, waxiness, glandular trichomes, and epidermal thicknesses to detect the suitable plant to probe (Johnson *et al*., 2006). From the distal end of its proboscis, a ring of hair that senses the plant traits and texture is used to get the most appropriate section to insert their stylets (Smith *et al*., 2014). When the aphids insert its stylet, the aphid performs various cell punctures where it decides whether to feed on the phloem available or not. The phloem sap is composed of water, carbohydrates from the sucrose, proteins, lipids, amino acids, mRNA, hormones, secondary compounds and inorganic ions (Smith *et al*., 2014). Once it commits to feeding on the phloem in the section patched, they produce saliva like compounds that sieve the elements for some time and then start ingesting the sap (Smith *et al*., 2014).

### 2.6.3 Crop Damage by aphids

Aphids affect plant growth and crop yield both directly and indirectly. The direct damage is affected by sucking sap, and in the process, nutrient sinks in the plants are drained, especially when the pest population is high (Figure 2.2). This in-turn causes yield losses estimated to be between 40 and 60%. The indirect damage caused by the pests is by acting as vectors by transmitting viral diseases. Aphids are the only known vectors of Barley Yellow Dwarf Virus disease. Both the direct and indirect damage is linked directly to their populations on the host plant. Practices that reduce the net reproduction of pests are one way to improve the management of bean aphids. A decline in the aphids' populations will reduce direct damage to the crop and consequently lower their local spread to other crops. A low population of aphids minimizes crowding, an essential stimulus for wing development, and subsequent dispersal (Nyaanga, 2008).

**Figure 2.2:**Aphid infested leaf

## 2.7 Management of aphids

### 2.7.1 Chemical control

Inorganic chemical natural nicotine, extracted from tobacco leaves which was sold as sulphate was used prior to Second World War and was considered the only effective insecticide against aphids (Dedryver *et al.*, 2010). It kills aphids that come into contact with the product and its action is not long-lasting. In 1939, the first organochlorinated insecticide, was discovered. After the war, other organochlorinated compounds were used to control aphids in the late 1940s, organophosphophates in the 1950s, carbamates in the 1960s, and pyrethroids in the 1970s (Dedryver *et al*., 2010). Organophosphates, carbamates, pyrethroids, cyclodienes, and neonicotinoids are the most commonly used insecticides (Guo *et al.*, 2017). Over the years due to indiscreet usage of insecticides on crops, aphids are becoming resistant their effect (Kaleem Ullah *et al*., 2023).

Insecticides have been used as an alternative short-term strategy to control aphids, however they are expensive leading to increase in cost of production, pollute the environment and destroy non-targeted beneficial insects such as predators, parasitoids and pollinators (El-Wakeil *et al*., 2014). Cruz *et al*. (1984) found out that spraying Diazinon 60 EC, Dimethoate 40 EC, Malathion 57 EC at 2.5 ml $l^{-1}$ of water at every 10 days reduced damage by *A. craccivora* on country bean. In a different study conducted to evaluate efficacy of different insecticides on beans, it was evident that application of Dimethoate 40EC, Deltamethrin 20EC and Pirimicarb 50 WP controlled aphid's population and increased yields by 27.23, 26.01 and 23.90%, respectively (Li *et al*., 2021).

Management of aphids is challenging because of their short life cycles and extremely high reproductive rates (Mkindi *et al.,* 2017; Mkindi *et al.,* 2020). A wide range of insecticides with various formulations are used to control aphids (Jackai & Daoust, 1986). However, inorganic control measures have been used such as spraying soap-water suspension at the rate of 25 ml liquid detergent per litre of water or spraying with the extract of Neem (*Azadiracta indica*) seed kernel. In a field trial conducted to determine efficacy of insecticidal soap kaolin on citrus groves showed that insecticides soap and kaolin foliar spray reduced aphid density for one week (Smaili *et al*., 2014). In a different study, three organically permitted products kaolin, mineral oil, and insecticidal soap were used to control *Myzus persicae* in a peach orchard (Karagounis *et al*., 2006). According to this study, kaolin is effective for aphid control in peach orchards and it has low-side effects on natural enemies and it could be used in organic peach orchards. All products had good control in the first year but were less effective the following year.

### 2.7.2 Host Plant Resistance

Host plant resistance forms an important modality in managing pests and conserving natural enemies in an agroecosystem (Giles *et al*.,2002). Host plant resistance or defense towards the infesting insects can be classified as constitutive or induced defense (Chen, 2008). Constitutive defense refers to the chemical and physical barriers that have been developed naturally by the plant. Once the aphids set to feed or oviposit on the plant surfaces, the plant leaf surfaces develop the initial physical defense (Walling, 2008). The plants produce hormones that result in transpiration reduction in the affected leaf, whilst the surface wax in the leaf provides vital component that protects the plant from the biotic stress (Jenks *et al*., 1994). The insects further encounter trichomes present in the leaf surface which form the initial defensive mechanisms. The trichomes can be glandular or non-glandular based on the species of the plant (Wagner *et al*., 2004). The relationship between the insect and the plant is determined by three types of resistance, particularly antibiosis, antixenosis and tolerance (Awais Iqbal *et al*., 2018;Koch *et al*., 2016). Antibiotic plant traits have a negative impact on pest biology by increasing mortality, decreasing growth, longevity, and fecundity (Smith, 2007). Antixenosis, also known as non-preference, is a host-expressed trait that has a negative impact on insect behavior since the insects have a preference for susceptible hosts over antixenotic hosts (Mondal *et al*., 2017).

Other plant characteristics that are activated when the insects infest the plant resulting in production of anti-nutritional and toxic compounds are referred to as direct resistance (Chen, 2008). Of these compounds produced during the inducible defense include the polyphenol oxidases and lectins that significantly reduce the population of the aphids. Plants with elevated snowdrop lectin levels have adverse impacts on growth and development of aphids (Gatehouse *et al*.,1996). Studies have shown that accumulation of the phenolic metabolites confers resistance of aphids such as the bird cherry oat aphids (*Rhopalosiphum padi*), cowpea aphid (*Aphis craccivora*) in the wheat and cowpea (*Vigna unguiculata*), respectively (Smith & Boyko, 2007). Application of the cowpea phenolic flavonoids on the faba bean (*Vicia faba*) leaves have been shown to reduce reproduction and multiplication of the black bean aphids (*Aphis fabae*) (Lattanzio *et al*.,2000). Plant also forms defense resistance to the insects indirectly by producing plant volatiles that involve tritrophic interactions between the aphids and the plants (Lattanzio *et al*., 2000). Additionally, the plants produce volatiles that attract the natural enemies to the infesting insect for instance the aphids such as the parasitoid wasps, lacewings, coccinellid beetles and hoverflies (Hatano *et al*., 2008).

### 2.7.3 Natural Enemies

Natural enemies (NEs) such as predators, parasitoids and pollinators are used for biological pest control and pollination hence are important in the ecosystem (Dainese *et al*., 2019). However, there is increasing concern about the widespread decline of NEs (Sánchez-Bayo & Wyckhuys, 2019). While many factors and their interactions may contribute to arthropod declines, landscape characteristics such as fertilizer and pesticide application, crop rotation, tillage practices, and the composition of the field surroundings are viewed as key drivers (Bakker *et al*., 2022; Jacobsen *et al*., 2022; Seibold *et al.*, 2019). Some studies have shown the negative effects of chemical pesticide application on the NEs of pests. Thus, field margins can be used to mitigate the negative effects of insecticides on populations of NEs (Bakker *et al*., 2021; Mkenda *et al*., 2015). A few studies have found populations of NEs is enhanced through intercropping (Azimi *et al*., 2015; Caballero-López *et al*., 2012; Clem, 2021; Tiroesele *et al*., 2019). An experiment conducted on common bean showed a subtle effect of intercropping versus mono-cropping on the natural enemy numbers: while overall populations were higher in intercropped systems, no individual taxon was more abundant in intercropped fields (Ndakidemi *et al*., 2022). This study suggests that intercropping alone as a method to support NEs populations may not result to improved pest management benefits and needs to be combined with other agro ecological interventions.

Previous study has reported variations in hereditary resistance to these enemies such as resistance of pea aphids (*Acyrthosiphon pisum*) to *Aphidius ervi* wasp (Oliver *et al*.,2005). Even though the variability was assumed to convey genotypic disparities between aphids, the other prospective cause of resistance to *A. Ervi* is an attack with the optional bacterial symbiont *Hamiltonella* protection (Oliver *et al*.,2005). Natural enemy diversity is more and more common in diverse environments containing greater quantities of natural habitat, and the resulting change in pest management in these ecosystems is not well known (Bianchi *et al*., 2006). Broad enemies demonstrate similar good reactions to landscape ambiguity, while specialized enemies, on the other hand, react more intensely to small - sized landscape ambiguity. Enemy reaction to native surroundings often appears to occur at higher dynamic levels than for specialized enemies, indicating that land management approaches to improve natural pest management will vary considerably based on whether the predominant enemies are specialists or generalists (Chaplin-Kramer *et al*., 2011).

### 2.7.4 Margin diversity

Diversified cropping can be traced back to early man and is still a vital farming practice in modern agriculture, given the fact that it has been practiced over the years. In recent years, controlling insect pests using the cropping as mentioned above system has been the focus of agricultural research (Chun *et al*., 2016). It affects the populations of natural enemies, parasitic, and also predation rates of the populations. Its effects on the natural enemies are credited to disturbing their orientation, foraging, and dispersal behaviours (Chun *et al*.,2016). Landscape composition directly impact the pest population by impairing its mobilization, mortality rate or fecundity, or ability to affect its real predators. In a study to determine an increase in arthropod diversity within farms with field margins, the results showed that during late spring, the margins provided a reliable, ecosystem for organisms that normally would not have lasted the entire season throughout the agricultural land with the existence of crop ecosystems, while the winter period provided refuge to several species of arthropods, such as predators effective in arable farms (Dennis *et al*., 1993).

Field margins have effects on the species diversity and density of plant pest prey in neighbouring plants during the early summer when population of the pest could be suppressed. Study by Veres *et al.*(2013) suggested that an improvement in the percentage of natural habitats regions across the landscape can lead to the development of these landscapes. They also recommended that consideration should be given to the intent of the specified semi-natural regions, as natural habitats provide pests. Grasslands, for instance, could be springs of leafhoppers and cereal thrips early in the growing season (Letourneau *et al*., 2009) and woodlands can sometimes deliver as nesting areas for pests like pollen beetle, or it may enhance nesting in other semi-natural places by altering microclimate situations.

In any planting season, plants for natural predators are both acceptable and unfit due to management activities or inconsistencies in the food supply (Samu & Szinetar, 2002). Few large-scale studies have measured the enemy effects on pest species, reducing our understanding of the complexities of the environment on pest management. Experimental studies should concentrate on making the connection between field or plant margins intensity and pest management from natural predators more discrete. A diverse or complex landscape is correlated to a higher abundance of natural enemies (Bianchi *et al*., 2006; Drapela *et al*., 2008; Gardiner *et al*., 2009a, 2009b; Schmidt *et al*., 2008; Werling & Gratton, 2008). A rise in the abundance of natural enemies is thought to augment pest control and the documentation of a positive correlation done on the relationship between landscape diversity and parasitism rates of predation in many systems nevertheless, pest population reduction is not guaranteed (Bianchi *et al*., 2006, 2008; Boccaccio & Petacchi 2009; Gardiner *et al*., 2009a; Thies *et al*., 2008).

In some systems, there is a positive correlation with pest densities indicating that pest densities may, at times, drive parasitoids (Costamagna *et al*., 2004; Thies *et al*., 2005). According to Kremen & Chaplin (2007), local diversification, from within field or around field hedgerow sources, can improve natural pest control. Therefore, it is important to evaluate whether these little, on-farm diversification approaches may lead in suppression of pests in large scale intensified systems. Although farmers have minimal control on intense modification in large scale farming, implementation of these strategies on local farms can result in huge advantages that can be trickled down to these farms (Morandin & Kremen, 2013).

# CHAPTER THREE
# MATERIALS AND METHODS

## 3.1 Study site

The study was conducted at the Agronomy Research and Teaching field of Egerton University situated in Nakuru county Kenya during the May to December2019 and 2020 growing seasons. The field lies at latitude 0° 22' S, longitude 35° 55’ E and an altitude of 2238m and is classified under lower highland 3 within the Kenya Highlands (Jaetzold *et al*., 2012). The area experiences annual precipitation of about 1200 mm per annum with a bimodal distribution of long rains (April –August) and short rains (October- December). The average maximum temperature of this location is 22°C with a minimum of 17°C. The weather data recorded during the study periods are presented in Table 3.1. The soils in the experimental site are predominantly mollic andosols which are well-drained dark reddish clays.

## 3.2 Field Experiment

### 3.2.1 Dolichos genotypes

A total of 18 genotypes sourced from different counties were used in the study; Machakos I – EUD1, Machakos II – EUD2, Machakos III – EUD3, Machakos Kiboko – EUD4 Eldo-KTL-Black I – EUD5, CIAT 22759 – EUD6, Echo Cream – EUD7, Brown Rongai – EUD8, Black Rongai – EUD9, CPI 81364 – EUD10, DL1002 – EUD11, Kikuyu Mkt – EUD12, Tx – 24 - EUD13, Q6880B – EUD14, Eldo KT Cream – EUD15, Kikuyu X-Meru – EUD16, Eldo KT- Black II–EUD17, HA – 4 – EUD18.

**Table 3.1:** Mean monthly Rainfall, Relative humidity and Temperature data recorded during study period- June to December,2019 and May to November 2020

| | **2019** | | | | **2020** | | | |
|---|---|---|---|---|---|---|---|---|
| | **Temperature** | | **Rainfall** | **Relative Humidity** | **Temperature** | | **Rainfall** | **Relative Humidity** |
| | **%** | | **Mm** | **%** | **%** | | **mm** | **%** |
| Month | **Max.** | **Min.** | | | **Max.** | **Min.** | | |
| May | 24.7 | 15.5 | 60.4 | 60 | 23.7 | 14.4 | 91.6 | 65 |
| June | 22.4 | 14.6 | 232.4 | 79 | 22.7 | 13.2 | 93.5 | 74 |
| July | 22.2 | 13.5 | 146.7 | 70 | 21.4 | 16.3 | 113.9 | 71 |
| August | 22.5 | 13.1 | 76.4 | 69 | 21.3 | 18.2 | 119.2 | 76 |
| September | 24.7 | 13.3 | 89.7 | 65 | 22.6 | 15.1 | 96.4 | 73 |
| October | 22.9 | 14.1 | 161.6 | 70 | 24.7 | 17.4 | 85.2 | 64 |
| November | 22.8 | 14.4 | 114.8 | 70 | 23.9 | 15.1 | 49.7 | 61 |
| December | 21.1 | 14.3 | 223.6 | 75 | 24.3 | 16.8 | 51.2 | 63 |

Source: Egerton University Engineering Meteorological Station, 2019 and 2020.

### 3.2.2Experimental design and treatment application

The field experiment was conducted during May-December 2019 and March-November 2020 growing seasons.The eighteen dolichos genotypes were planted in experimental units measuring 3.0 m x 2.5 m in a factorial arrangement within a Randomized Completely Block Design (RCBD) with four replicates, with a plant population of 40 per plot. Each genotype was planted in the presence or absence of field margin vegetation (FMV).To enhance the field margin vegetation, seeds from four plant species (*Bidens pilosa, Tagetes minuta, Ageratum conyzoides and Galinsoga parviflora*) that our earlier work has identified as having a positive influence on natural pest regulation (Amoabeng *et al*., 2020; Ndakidemi *et al*., 2022; Obanyi *et al*., 2023) were mixed in equal proportions and randomly planted. The FMV seed mixtures were sown two weeks before the dolichos crop was planted. In plots that had no field margin vegetation, the ground was left bare throughout the growing period by hoeing out any vegetation around the dolichos crops. The margin species were planted 0.5m from the outer row of dolichos as a strip 0.5m in width. At planting each plot received an equivalent rate of 60 kg $ha^{-1}$ of N.P.K (23:23:0), to supply 13.8 kg N $ha^{-1}$ and 13.8 kg $P_2O_5$ $ha^{-1}$. Within the experimental plots, mechanical weeding and inter-cultivation operations were done twice, at 21 days after planting (seedling stage) and 49 days after planting (early to late vegetative stage). Throughout the growing season no pesticides were applied to prevent confounding effects on crop growth, bean aphids and natural enemies.

### 3.2.3 Data Collection

Data collection on aphid abundance, severity of crop damage and aphid incidence was conducted at four distinctive dolichos growth stages; seedling, early vegetative, late vegetative and flowering-podding stages.

**Aphid abundance**

Aphid abundance was determined by visually observing and scoring aphid abundance from ten randomly selected plants from the inner three rows of each plot in each treatment. A categorical scale was used to assess aphid abundance, 1 = no aphids; 2 = a few scattered aphids; 3 = a few small colonies; 4 = several small colonies; 5 = large isolated aphid colonies; and 6 = large continuous colonies (Mkenda *et al*., 2015) (Table 3.2).

**Table 3. 2:** Aphid abundance assessment scale

| Number | Aphid population | Visual expression |
|---|---|---|
| **1** | No aphid infestation and damage |  |
| **2** | A few scattered aphid |  |
| **3** | A few isolated colonies |  |
| **4** | Several isolated aphid colonies |  |
| **5** | Large isolated colonies |  |
| **6** | Large continues colonies |  |

Sources: Mkenda *et al*. (2015)

### Aphid incidence

The percent aphid incidence was determined by visually examining and counting aphid damaged or aphid infested dolichos plants in each treatment. Incidence was assessed on a 0-1 scale (where; 0 = clean plant with no signs of aphids' infestation and 1 = plant with signs of aphids' infestation. Incidences were obtained by randomly sampling 20 plants from the inner five rows in each replicate plot and was expressed as percentage incidence using the following formula;

$$\text{Aphid incidence } (\%) = \frac{\text{Number of infested plants}}{\text{The total number of plants observed}} \times 100 \ldots\ldots\ldots\ldots\ldots\ldots\ldots \text{ Equation 3.1}$$

### Aphid damage severity

Damage severity on the crop was determined by visually observing and scoring the level of aphid damage in each experimental unit from ten randomly selected plants in the inner three rows. The damage severity was assessed by scoring the extent of damage using a 1 to 5 scale, where: 1= no infestation or damage, 2=light damage and infestation < 25 % plant parts damaged or infested, 3=average damage and infestation 26 % - 50 % plant parts damaged, 4=high infestation and damage 51 % - 75 % plants parts damaged showing yellowing of lower leaves and 5= severe infestation >75 % damage resulting to plants with high infestation levels with a yellow and severely curled or dead plant (Mkenda *et al.*, 2015) (Table 3.3).

**Table 3. 3:**Damage severity scoring scale

| Scale | Description of damage | Visual expression |
|---|---|---|
| 1 | No symptoms of an attack |  |
| 2 | Showing damage symptoms up to 25% |  |
| 3 | Showing damage symptoms from 26-50% |  |

Sources: Mkenda *etal*. (2015)

### Natural enemy abundance and diversity

Sampling of natural enemies was done at seedling, vegetative, flowering and podding growth stages. The population of ground dwelling natural enemies was determined using yellow pan traps placed at the crop center and field margin vegetation (Plate 3.1 A). The yellow pan traps consisted of 20cm diameter plastic plates filled with water to three-quarter level. Three droplets of soapy detergent were added to break the surface tension and prevent trapped insects from crawling or flying away. The population of flying natural enemies was determined by hanging of yellow sticky cards 30cm above the crop at the crop centre and in the field margin vegetation (Plate 3.1 B). The yellow sticky traps were of dimensions 10cm by 25cm. The traps were left in field for 48hours. Natural enemies caught by the yellow sticky traps were removed by washing out with analytical hexane. All the natural enemies' samples were transferred to 50ml falcon tubes containing 75% ethanol for preservation prior to laboratory identification. Natural enemies of bean aphids were sorted and identified to the family level under light microscope (Leica ZOOM 2000 Inc. Buffalo. NY U.S.A 14240-0123) using identification keys of Arnett and Jacques (1981). The number of natural enemies recorded on each dolichos genotype at each growth stage in each of the two field margin vegetation statuses was used to estimate species richness, abundance and diversity. The diversity of insect families was estimated using the Shannon-Weaver index (H) formula as described in equation 3.2 below.

$$H = -\Sigma p_i ln p_i \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots \text{ Equation 3.2}$$

Where $H$ is the Shannon's diversity index and pi is the proportion of individuals found in the $i^{\text{th}}$ species $ln$ is the natural logarithm of individuals found in the $i^{\text{th}}$ species.

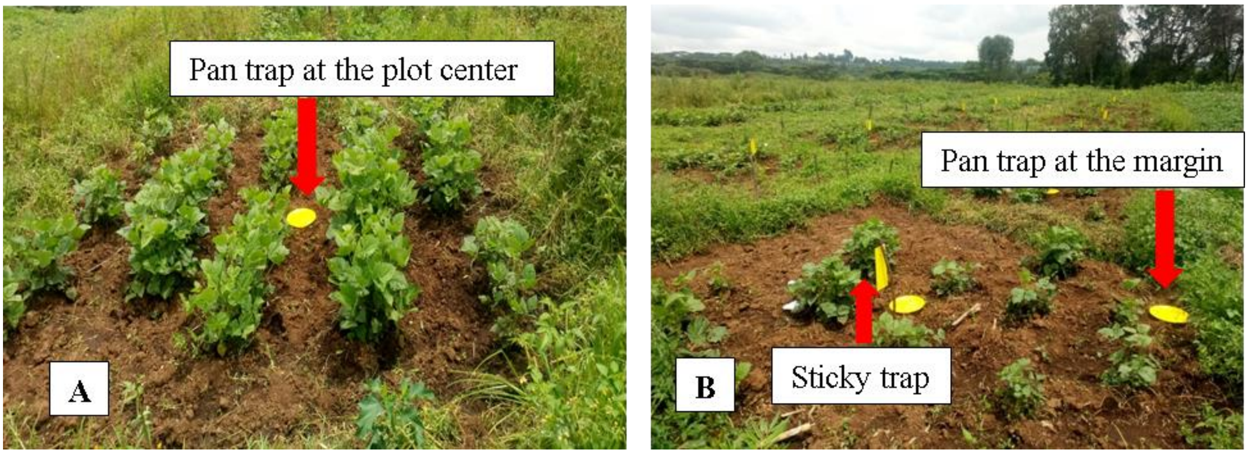

**Plate 3. 1:** Yellow sticky traps and pan traps: (A) at the crop center and (B) at the field margin vegetation

### Morphological characteristics, growth, yield and yield components of dolichos genotypes

Morphological characterization was based on visual observations of growth habits and structure of the plants. Growth, yield and yield components data was collected to ascertain variation in genotype performance in the study. Data on stand count, number of days to 50% flowering, plant height, pod length, number of pods per plant, number of seeds per pod, peduncle length, above ground biomass and grain yield were collected from each plot across the treatments. For the growth and yield aspects that were measured five plants were randomly selected within the three inner rows in each plot except for stand count that was determined by counting all the plants in each plot three weeks after planting. Number of days to 50% flowering was determined by observing each genotype from emergence to when 50% of plants flowered. Plant height was measured at physiological maturity from ground level to the tip of the main stem using a 1m wooden measuring ruler. To determine above ground biomass, destructive sampling was adopted at pod set, when the dry matter accumulation was at maximum. The plants were cut just above the soil surface fresh weight measured and taken an oven for drying at 65 °C for 24 hours. The number of pods per plant was counted in each of the five randomly selected plants from the inner middle rows. Similarly, the number of seeds per pod was determined by threshing each pod, counting the seeds and computing means. The weight of a hundred seeds was determined using a digital QZ-161 electronic kitchen scale (Comglobal Solutions, India). For grain yield, pods were harvested separately within the sampling area for each treatment. Pods were sun-dried for three days, threshed and seeds dried as the moisture content was monitored using a draminski digital moisture meter (Dramiński S.A., Poland). After attaining 13 % moisture content, grains from each treatment were weighed separately using a portable digital weighing scale (Comglobal Solutions, India) and the weight converted to tonnes ha$^{-1}$ using equation 3.3.

$$GY = \frac{GWP \times 10}{HA\ (m^2)} \quad \text{………………………………………………………………Equation 3.3}$$

Where GY=Grain yield in tons/ha, GWP= Grain weight per plot and HA= Harvest area in m$^2$

### 3.2.4Data analyses

Data on percent incidence and natural enemy counts were subjected to arcsine square root and $\sqrt{x+1}$transformation, respectively before analysis of variance. Data on aphids' abundance, severity, incidence, days to 50% flowering, yield and yield components were subjected to analysis of variance using *PROC GLM* in Statistical Analysis System version 9.2 (SAS Institute, 2011) with genotypes and field margin vegetation as fixed effects and replicate as random variable. The model used is described in the equation 3.4 below.

$$Y_{ijkl} = \mu + S_i + R_j + G_k + M_l + SG_{ik} + GM_{kl} + SGM_{ikl} + \varepsilon_{ijkl} \text{……………………Equation 3.4}$$

Where, $Y_{ijkl}$= observed performance of the $k^{th}$ dolichos genotype, in the $i^{th}$ season, on the $j^{th}$ replicate within $l^{th}$ margin. $\mu$ is the overall mean; $S_i$= effect due to $i^{th}$ season $R_j$= effect due to $j^{th}$ replicate; $G_k$= effect due to $k^{th}$genotype; $M_l$= effect due to $l^{th}$margin, SGij= effect due to interaction between $i^{th}$ season and $k^{th}$genotype; GMkl= effect due to interaction between $k^{th}$ genotype and $l^{th}$ margin; *SGMikl* = effect due to interaction between i$^{th}$ season and k$^{th}$ genotype and l$^{th}$ margin; $\varepsilon_{ijkl}$ = random error component.

Tukey's Honestly Significant Difference (HSD) was used to compare treatment means at 5% level of significance and calculations were guided by equation 3.5 below.

$$R = q\ (\alpha, p, f) X \sqrt{\frac{MSE}{r}} \text{………………………………………………………Equation 3.5}$$

Where $q(\alpha, p, f)$ is the standardised range, $\alpha$ is the significance level, $p$ is the number of treatments, $f$ is the error degree of freedom and $r$ is the number of replications.

## 3.3 Cage experiment

### 3.3.1 Design and treatment application

To validate the findings from the field experiment, a cage exclusion experiment was conducted to investigate the effect of dolichos genotypes on bean aphids and their natural enemies. The experiment was laid in a Randomized Complete Block Design (RCBD) with four replicates. Treatments involved four genotypes of Dolichos bean - 2 that are susceptible and two tolerant or resistant. These genotypes were selected from those that exhibited resistance or susceptibility in the field. The criteria for establishing the susceptible genotypes involved picking those that showed significantly high abundance and incidences of aphids across the growth stages of dolichos. Each treatment was placed in exclusion cages measuring 1x1x2m (length x width x height) set 2m apart from each other. A total of to 8 cages were set up. The cages consisted of a tubular metal frame covered with polyester netting of mesh size 1 × 0.3mm and thread thickness of 0.1mm. The netting was extended below the soil surface to a depth of about 20cm and access was via a vent held closely by white zipping in each cage unit. Each replication had 8 plants planted in pots under a cage. Standard cultural practices involved top dressing with nitrogenous fertilizer, and watering. A sentinel plant pre infested with a population of 60 aphids was introduced in the cages at the vegetative and flowering growth stages. A total of 20 *Aphidius colemani* parasitoids were released into the cages and allowed to colonize freely through the growth stages. *A. colemani* was deemed ideal in this case because it was the most abundant primary parasitoid identified to manage bean aphids' populations in dolichos from similar research in the same study system (Mkenda *et al*., 2019).

**Plate 3. 2** Field exclusion cages

### 3.3.2 Data collection

Aphid incidence was recorded upto physiological growth stage on occurrence of aphids and was determined by examining the upper and lower leaf surfaces according to Byregowda *et al*., (2015). Number of mummified aphids was determined per sampled plant and the percentage determined using the formula in equation 3.6.

$$\text{Mummification (\%)} = \frac{\text{Number of mummies}}{\text{Total number of aphids}} \text{x} 100 \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots \text{Equation 3.6}$$

The subsequent percentage mummification was determined as the proportion of mummified aphids by subtracting the number of mummies from the total aphid population and obtaining a percentage. Population density of aphids present in each genotype was recorded from eight tagged plants from branching stage up to harvest and cumulative data expressed as absolute population per plant.

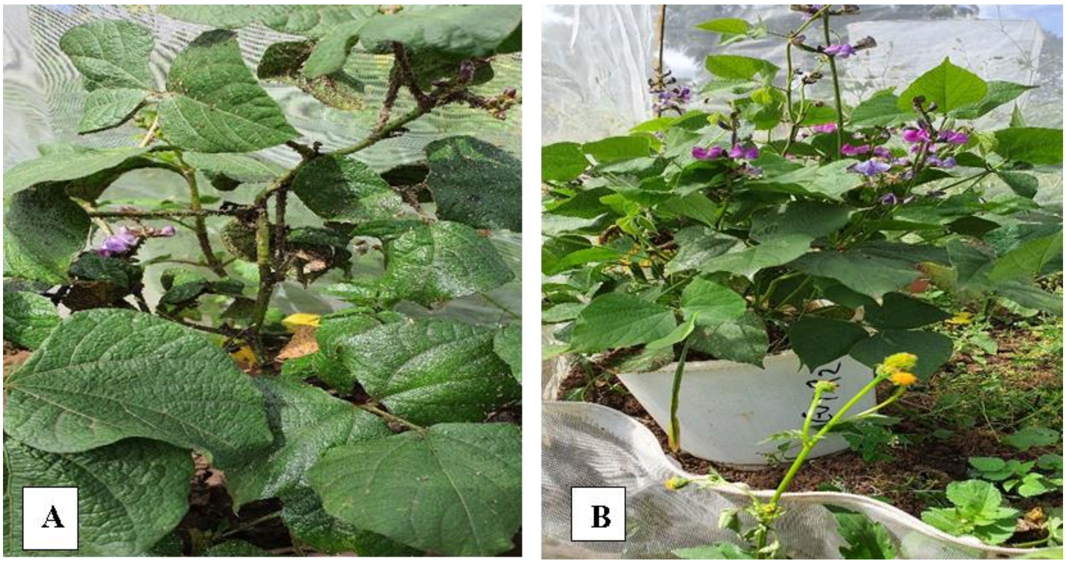

**Plate 3. 3** Aphid infestation levels on dolichos genotypes: (A) susceptible and (B) Tolerant

### 3.3.3 Data analyses

Data on aphids abundance, incidence, and mummification were first subjected to normality test using PROC UNIVARIATE in Statistical Analysis System version 9.2 (SAS Institute, 2011). Data on percent incidence were transformed using arcsine square root transformation before analysis of variance. The response variables were subjected to analysis of variance using *PROCGLM* in SAS using the following statistical model:

$$Y_{ijkl} = \mu + R_i + G_j + S_k + GS_{jk} + \varepsilon_{ijkl} \quad \text{...Equation 3.7}$$

Where, $Y_{ijkl}$= is the observation performance of the $j^{th}$ dolichos genotype, in the $i^{th}$ replicate, on the $k^{th}$ stage of growth. $\mu$ is the overall mean; $R_i$= effect due to $i^{th}$ replicate; $G_j$= effect due to $j^{th}$ genotype; $S_k$= effect due to $k^{th}$ Stage of growth, $GS_{jk}$= effect due to interaction between $j^{th}$ genotype and $k^{th}$stage of growth; $\varepsilon_{ijkl}$ = random error component.

Tukey's Honestly Significant Difference (HSD) was used to separate treatment means at 5% level of significance.

# CHAPTER FOUR

# RESULTS

## 4.1 Effect of field margin vegetation and dolichos genotypes on aphid abundance, damage severity and percent incidence

### 4.1.1 Aphid abundance

Results on aphid abundance showed that field margin vegetation (FMV) had no significant effect on aphid abundance. The highest aphid abundance was observed during 2020 cropping season in the presence of field margin (2.6) compared to the absence of FMV (2.4) (Table 4.1).Dolichos growth stage significantly affected aphid abundance over the two cropping seasons, generally peaking in early growth stages and decreasing later. In 2019, the flowering and podding stage had the highest mean abundance of 1.7. In 2020, the seedling stage had the highest mean aphid abundance (3.3) while it was lowest at flowering and podding stage (1.0) (Table 4.2). There was a significant variation at $P<0.05$ among the genotypes for aphid abundance. Aphid abundance was highest in genotypes Eldo-KT-Cream (2.08) followed by Eldo-KT-Black II (2.03). The two genotypes were however not significantly different from Machakos III (1.9), CIAT 22759 (1.9), Echo Cream (1.9), Black Rongai (1.8), CPI 81364 (1.8), DL1002 (1.8), Kikuyu Market (2.0) and Tx-24 (1.8). The lowest aphid abundance (1.75) was observed in Brown Rongai but it was not significantly different from *Machakos I* (1.76) (Table 4.3).

### 4.1.2 Aphid damage severity

Field margin vegetation (FMV) had no significant effect on aphid damage severity. The highest damage severity (2.4) was observed in plots without FMV compared to those with FMV (2.3 during the 2020 cropping season) (Table 4.1).Dolichos growth stage significantly affected damage severity. In 2019, seedling and flowering to podding growth stages had the highest damage severity of 1.1 compared to vegetative growth stage (1.0). In 2020, seedling stage had the highest damage severity (2.4) while the lowest mean was observed at flowering and podding stage (2.1) (Table 4.2). There was a significant variation at $P<0.05$ among the genotypes for damage severity. Damage severity was highest (1.79) in Eldo-KT-Creamgenotype. This was however not significantly different from HA-4 (1.57), Eldo-KT-Black II (1.75), Q6880B (1.59), Tx-24 (1.62), Machakos II (1.59), Machakos III (1.67), Machakos Kiboko (1.56), Eldo-KTL-Black (1.57) and Black Rongai (1.65). The lowest damage severity (1.51) was observed in Machakos 1 which was not significantly different from Kikuyu x Meru (1.53) (Table 4.3).

### 4.1.3 Aphid incidence

There was no significant effects noticed in aphid incidence as influenced by Field margin vegetation (FMV) (Table 4.1). However, in 2020 cropping season, the highest incidence was observed in the absence of field margin vegetation (60.6) compared to the presence of plots with (58.0). Dolichos growth stage significantly influenced aphid incidence. In 2019, flowering to podding stage had the highest aphid incidence of 31.7% and lowest at late vegetative (2.1%).

**Table 4. 1:** Effect of field margin vegetation on abundance, severity and incidence during 2019 and 2020 cropping seasons.

| | **2019** | | | **2020** | | |
|---|---|---|---|---|---|---|
| **FMV** | **Aphid abundance** | **Severity** | **Incidence (%)** | **Aphid abundance** | **Severity** | **Incidence (%)** |
| Present | $1.2^a$ | $1.1^a$ | $13.8^a$ | $2.6^a$ | $2.3^a$ | $58.0^b$ |
| Absent | $1.3^a$ | $1.1^a$ | $14.6^a$ | $2.4^b$ | $2.4^b$ | $60.6^a$ |
| **MSD** | **0.3** | **0.0** | **1.2** | **0.1** | **0.1** | **2.4** |

Means in a column followed by the same letters are not significantly different at $P<0.05$ using Tukey’s HSD test. FMV= Field margin vegetation; MSD = Minimum significant difference.

In 2020, seedling stage had the highest aphid incidence (62.6%) and lowest at late vegetative (50.1) (Table 4.2

**Table 4.2:**Abundance, severity and incidence of aphids as affected dolichos growth stage during 2019 and 2020 cropping seasons.

| | **2019** | | | **2020** | | |
|---|---|---|---|---|---|---|
| **Growth stage** | **Aphids abundance** | **Severity** | **% Incidence** | **Aphids abundance** | **Severity** | **% Incidence** |
| Seedling | $1.2^{b}$ | $1.1^{a}$ | $19.9^{b}$ | $3.3^{a}$ | $2.4^{a}$ | $62.6^{a}$ |
| Early Vegetative | $1.0^{c}$ | $1.0^{b}$ | $3.2^{c}$ | $3.0^{b}$ | $2.2^{b}$ | $58.1^{b}$ |
| Late vegetative | $1.1^{c}$ | $1.0^{b}$ | $2.1^{c}$ | $2.7^{c}$ | $2.2^{b}$ | $50.1^{c}$ |
| Flowering-podding | $1.7^{a}$ | $1.1^{a}$ | $31.7^{a}$ | $1.0^{d}$ | $2.1^{b}$ | $66.5^{a}$ |
| MSD | **0.15** | **0.04** | **1.45** | **0.11** | **0.13** | **3.16** |

Means in a column followed by the same letters are not significantly different at $P<0.05$ using Tukey's HSD test. MSD = Minimum significant difference.

There was a significant variation at $P<0.05$ among the genotypes for aphid incidence. Aphid incidence was highest in Kikuyu Market (48.6%) which was not significantly different from Eldo-KT-Cream (41.6%), Black Rongai (41.6%), Eldo-KTL-Black (41.7%) and Machakos II (43.3%). The lowest aphid incidence was observed in genotype Machakos I (20.9%) which was significantly different all the genotypes (Table 4.3).

**Table 4.3:**Abundance, severity of damage and incidence of aphids in 18 dolichos genotypes during 2019 and 2020 cropping seasons

| **Genotype** | **Aphid abundance** | **Severity** | **% Incidence** |
|---|---|---|---|
| Machakos I | $1.76^{d}$ | $1.51^{b}$ | $20.9^{f}$ |
| Machakos II | $1.82^{bcd}$ | $1.59^{ab}$ | $43.3^{ab}$ |
| Machakos III | $1.86^{abcd}$ | $1.67^{ab}$ | $36.9^{bcde}$ |
| Machakos Kiboko | $1.78^{cd}$ | $1.56^{ab}$ | $32.2^{ed}$ |
| Eldo-KTL-Black | $1.86^{abcd}$ | $1.57^{ab}$ | $41.7^{abc}$ |
| CIAT 22759 | $1.92^{abcd}$ | $1.64^{ab}$ | $38.4^{bcde}$ |
| Echo Cream | $1.97^{abcd}$ | $1.73^{ab}$ | $33.0^{cde}$ |
| Brown Rongai | $1.75^{d}$ | $1.56^{ab}$ | $30.2^{ed}$ |
| Black Rongai | $1.88^{abcd}$ | $1.65^{ab}$ | $41.6^{abc}$ |
| CPI 81364 | $1.88^{abcd}$ | $1.62^{ab}$ | $31.3^{ed}$ |
| DL1002 | $1.89^{abcd}$ | $1.62^{ab}$ | $37.5^{bcde}$ |
| Kikuyu Market | $2.01^{abc}$ | $1.76^{ab}$ | $48.6^{a}$ |
| Tx-24 | $1.85^{abcd}$ | $1.62^{ab}$ | $38.8^{bcd}$ |
| Q6880B | $1.83^{bcd}$ | $1.59^{ab}$ | $38.4^{bcde}$ |
| Eldo-KT-Cream | $2.08^{a}$ | $1.79^{a}$ | $41.6^{abc}$ |
| Kikuyu × Meru | $1.83^{bcd}$ | $1.53^{b}$ | $37.0^{bcde}$ |
| Eldo-KT-Black II | $2.03^{ab}$ | $1.75^{ab}$ | $36.3^{bcde}$ |
| HA-4 | $1.85^{abcd}$ | $1.57^{ab}$ | $34.1^{cde}$ |
| MSD $_{(0.05)}$ | 0.24 | 0.25 | 8.30 |

*Means in a column followed by the same letters are not significantly different using Tukey's HSD test at $P \leq 0.05$.

### 4.4.2 Effect of dolichos genotypes and field margin vegetation on natural enemies' species richness, abundance and diversity

Analysis of variance indicated that dolichos genotypes was significant for species richness (ANOVA: $F_{(17,\ 34)} = 2.19$; $P=0.02$) with no statistical difference for abundance (ANOVA: $F_{(17,\ 34)} =0.97$; $P=0.51$) and species diversity (ANOVA: $F_{(17,\ 34)} = 0.19$; $P=0.57$). In addition, there was a significant (ANOVA: $F_{(17,\ 34)} = 3.81$; $P<0.0001$) difference of genotype and season interaction for species richness with no statistical differences for abundance and species diversity (Appendix 2). However, across the two-cropping season, field margin vegetation showed slight variations in relation to species richness, abundance and diversity (Table 4.4). Natural enemies collected during the study periods differed significantly between the margin vegetation and crop; the majority of the natural enemies were captured within the dolichos crop than margin vegetation.

**Table 4. 4:**Bean aphid natural enemies' richness, abundance and diversity as influenced by field margin vegetation and trapping area during 2019 and 2020 cropping seasons

| | 2019 cropping season | | | 2020 cropping season | | |
|---|---|---|---|---|---|---|
| **FMV** | **Richness** | **Abundance** | **Diversity** | **Richness** | **Abundance** | **Diversity** |
| Present | $4.18^{a}$ | $12.29^{b}$ | $0.64^{a}$ | $3.36^{b}$ | $8.71^{b}$ | $0.26^{b}$ |
| Absent | $4.13^{a}$ | $17.35^{a}$ | $0.64^{a}$ | $4.77^{a}$ | $13.65^{a}$ | $0.32^{a}$ |
| **MSD** | **0.32** | **2.50** | **0.03** | **0.46** | **1.88** | **0.02** |
| **Trapping position** | | | | | | |
| Crop centre | $4.33^{a}$ | $16.72^{a}$ | $0.65^{a}$ | $4.69^{a}$ | $12.81^{a}$ | $0.31^{a}$ |
| Margin vegetation | $3.83^{b}$ | $4.49^{b}$ | $0.62^{a}$ | $2.11^{b}$ | $5.43^{b}$ | $0.23^{b}$ |
| **MSD** | **0.32** | **2.50** | **0.03** | **0.46** | **1.88** | **0.02** |

Means in a column followed by the same letters are not significantly different using Tukey's HSD test at P≤ 0.05. FMV= Field margin vegetation.

Among the natural enemy taxa collected were tachnid flies (Diptera: Tachnidae), hoverflies (Diptera: Syriphidae), Parasitic wasps (Ichneumonidae and Braconidae), ladybird (Coleoptera: coccinellidae). In 2019, tachnid flies were the most abundant as compared to other taxa while in 2020, wasps were the most abundant. Manipulating FMV was more effective for increasing richness and abundance in simplified, crop-dominated landscapes than in diverse ones for a variety of arthropods e.g.,pollinators generally (Carvell *et al.*, 2011) , such as bees (Holzchuch *et al*.,2007), butterflies (Rundlöf *et al* .,2006), and aphidophagous syrphids (Haenke *et al.,* 2001). Other studies have shown that taxon diversity and abundance respond primarily to landscape composition alone with habitat manipulation or management having a negligible influence. These contrasting results may indicate that these relationships are species- and context-dependent. Generally, in 2020, low abundance of natural enemy taxa was observed as compared to 2019 with hover flies recording the least abundance across the two years (Figure 3.1).

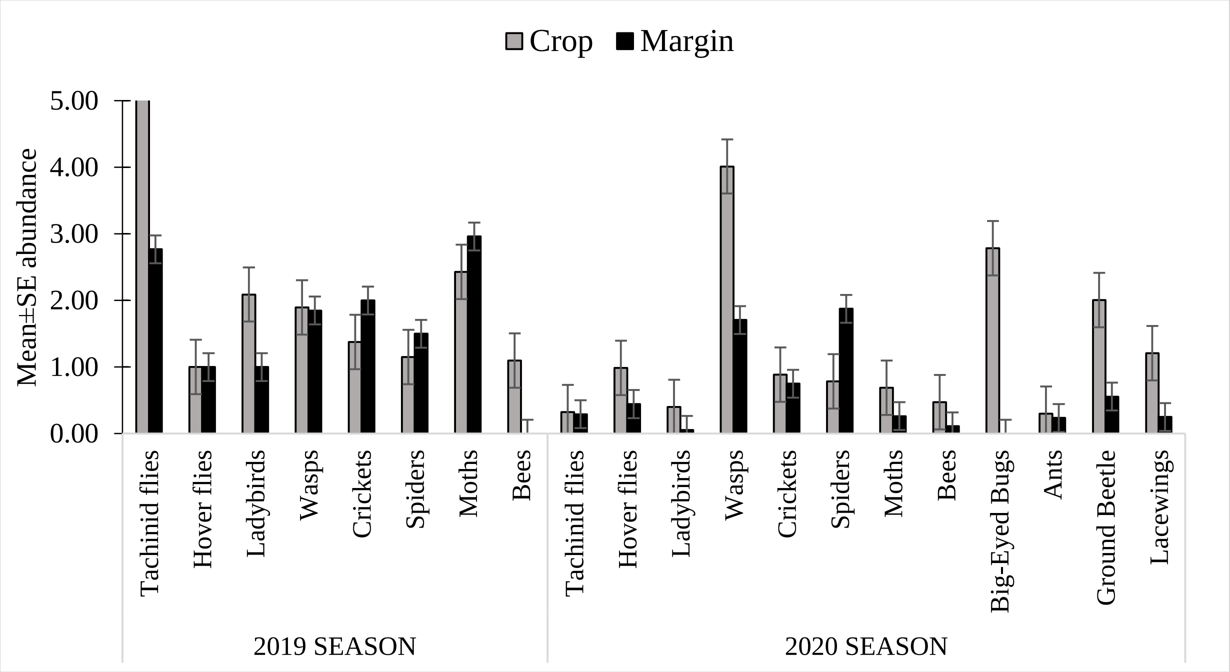

**Figure 3.1:**Abundance (mean ± SE) of natural enemies of aphids as influenced by trapping position (either at field margin vegetation or dolichos crop) during 2019 and 2020 cropping seasons

Observations also indicated that dolichos genotypes varied in supporting insect families depending on the season, Eldo-KT-black-II (11.75) had the highest tachinid flies (7.5) in 2019 season while Kikuyu market had the lowest (Figure 3.2 and 3.3). Genotypes Machakos-II (22.83) and DL1002 (21.17) had high abundance of tachinid flies in 2019 in contrast to genotypes Machakos-I (0.75), Q6880B (0.67) and Eldo-KT-Cream (1.25) which had a high abundance in 2020 (Figure 1a and c). Hoverfly abundance was highest in genotypes Machokos I (2), CIAT-22759 (3), TX-24 (3.2) and Eldo-KT-Black-II (2) in 2019 while Kikuyu-Market (1.58), Eldo-KT-Cream (1.25), Machakos-Kiboko (1.08), HA4 (1.42), Q6880B (1.42) and Eldo-KTL-Black (1.08) in 2020 (Figure 1a and c). Genotype Echo-Cream had low hoverfly abundance in 2019 and 2020 (0.97 and 1.0 respectively) seasons. A high ladybird abundance was recorded in Machakos-II (4 in 2019 and 0.75 in 2020) while Eldo-KTL-Black (0.40 in 2019 and 0.50 in 2020) registered low abundance in both seasons. Abundance of parasitic wasps were high in Machakos-I (5.83), Kikuyu-Market (4.67) and Q6880B (5.50) in 2019 and in CIAT-22759 (4.42), HA4 (5.17), Kikuyu- Meru (4.0) and CPI-81364 (3.83) in 2020 (Figure 3.2 and 3.3).

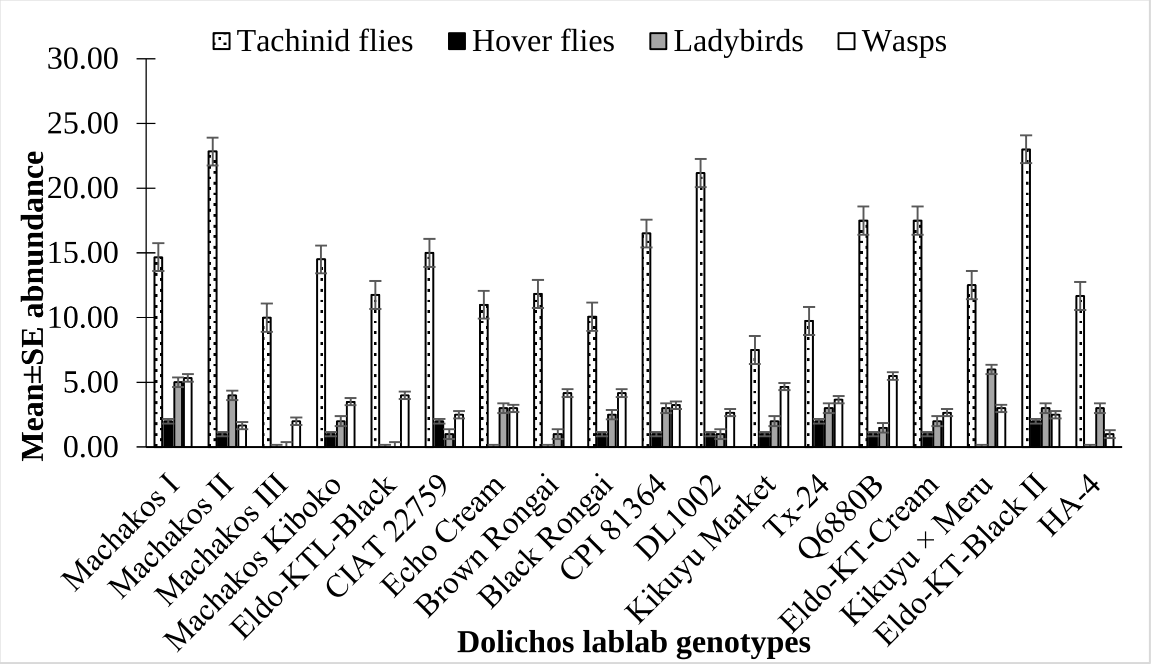

**Figure 3.2:**Abundance (mean ± SE) of natural enemies of aphids on 18 dolichos genotypes during the 2019 cropping season.

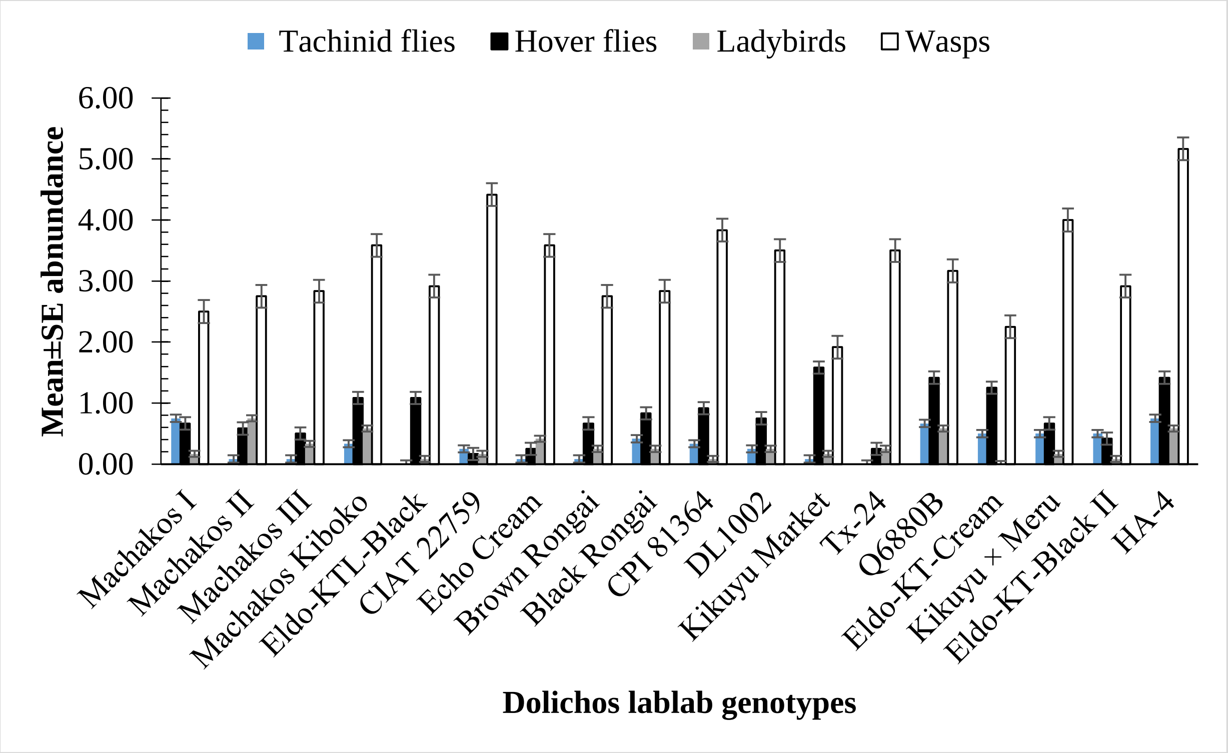

**Figure 3.3:**Abundance (mean ± SE) of natural enemies of aphids on 18 dolichos genotypes during the 2020 cropping season.

### 4.4.3 Characterization of morphology characteristics, growth, yield and yield components of dolichos genotypes

Morphological characterization was based on visual observations of growth habits and structure of the plants. Variation in major qualitative and quantitative characteristic was observed across the genotypes. Duration to maturity, presence or absence of trichomes and flower colour varied across the genotypes (Table 3.8). These traits to a great extent have been attributed to insect tolerance levels in crops. During crop establishment, majority of genotypes had high stand counts with Kikuyu-Market and Kikuyu-Meru having the highest while Echo-Cream had the lowest stand count. The highest days to flowering was recorded in CPI-81364 (103 days) while HA-4 had the lowest (79 days). In terms of growth Black-Rongai registered the highest plant height (117 cm) while *Kikuyu Meru* had the lowest (78 cm). Echo-Cream had the highest peduncle length of 4.6 while *Machakos* I had the lowest. These observations indicate slight variation among the genotypes. There was a high variation in number of pods per plant among the genotypes with DL-1002 and Eldo-KT-Cream having the highest (38) and *Machakos*-II having the lowest (26). Mean yield ranged between 0.97-0.19 t/ha with an average mean of 0.44 t/ha. Genotype *Brown-Rongai* had the highest yield (0.97 t/ha) while Machakos-I and HA-4 had the lowest yield (0.19 t/ha). *Echo-Cream* and *Machakos* II had the highest biomass of 1.5 t/ha and 1.4 t/ha respectively while Machakos III and HA4 had the lowest biomass of 0.62 t/ha and 0.63 t/ha, respectively (Table 3.9).

**Table 4. 5:**Dolichos genotypes agro-morphological characteristics

| Genotype | Maturity | Seed Colour | Flower Colour | Leaf qualities | Purpose of use |
|---|---|---|---|---|---|
| Machakos I | Mid | Black | Purple | Pubescent | Dual purpose |
| Machakos II | Mid | Brown-spotted | Purple | | Dual purpose |
| Machakos III | Mid | Brown | White | | Grain variety |
| Machakos Kiboko | Late | Black | Purple | | Grain variety |
| Eldo-KTL-Black I | Late | Black | Purple | Pubescent | Grain variety |
| CIAT 22759 | Late | Black | Purple | | Forage variety |
| Echo Cream | Late | Cream | White | | Forage variety |
| Brown Rongai | Mid | Brown | Purple | Pubescent | Forage variety |
| Black Rongai | Late | Black | Purple | | Dual purpose |
| CPI 81364 | Late | Brown | White | Pubescent | Grain variety |
| DL1002 | Late | Black | Purple | | Grain variety |
| Kikuyu Mkt | Late | Black | Purple | | Grain variety |
| Tx – 24 | Late | Cream | Purple | | Forage variety |
| Q6880B | Late | Black | Purple | | Dual purpose |
| Eldo KT Cream | Late | Brown | White | | Grain variety |
| Kikuyu X-Meru | Late | Black | Purple | | Grain variety |
| Eldo KT- BlackII | Mid | Black | Purple | | Grain variety |
| HA – 4 | Early | Cream | White | Pubescent | Dual purpose |

**Table 4. 6:**Comparison of agronomic traits and yield components for 18 dolichos genotypes for 2019 and 2020 cropping seasons

| Genotype | Stand count | Days to 50% flowering | Plant height | Peduncle length | Pod length | No. of peduncle | No. of pods plant$^{-1}$ | Yield | Biomass |
|---|---|---|---|---|---|---|---|---|---|
| | | | | cm | | | | t ha$^{-1}$ | |
| Machakos I | 33.188a-c | 90.43b | 86.43b | 9.72b-f | 4.11hi | 10.56c-e | 27.56ef | 0.19e | 0.62d |
| Machakos II | 32.5a-d | 97.813ab | 95.75ab | 8.89ef | 4.29b-h | 8.31e | 26.5f | 0.35c-e | 1.4ab |
| Machakos III | 34.625ab | 101.188a | 98.06ab | 8.38f | 4.18f-h | 11.06b-e | 33.25b-d | 0.47b-d | 0.68d |
| Machakos Kiboko | 34.938a | 97.188ab | 86.56b | 11.29bc | 4.36b-g | 10.93c-e | 33.43b-d | 0.38c-e | 0.72d |
| Eldo-KTL-Black | 32.625a-d | 101a | 99.5ab | 9.23d-f | 4.42b-d | 11.37b-d | 30.62d-f | 0.53b-d | 1.23a-c |
| CIAT 22759 | 36.938a | 100.063a | 99.5ab | 9.11d-f | 4.27b-h | 9.68c-e | 31.62c-e | 0.36c-d | 0.66d |
| Echo Cream | 28.43d | 100.625a | 87.56b | 8.96d-f | 4.68a | 11.56b-d | 36.62a-c | 0.65b | 1.5a |
| Brown Rongai | 35.625a | 100.375a | 92.06ab | 12.49ab | 4.41b-f | 16.25a | 31.37c-f | 0.97a | 1.02b-d |

**Table 4.6:** Continued….

| **Genotype** | **Stand count** | **Days to 50% flowering** | **Plant height** | **Peduncle length** | **Pod length** | **No. of peduncle** | **No. of pods plant$^{-1}$** | **Yield** | **Biomass** |
|---|---|---|---|---|---|---|---|---|---|
| | | | ——— | cm | ——— | | | **t ha$^{-1}$** | |
| Black Rongai | 34.25a-c | 101.188a | 117.18a | 10.38b-f | 4.21c-i | 10.62c-e | 27.18f | 0.51b-d | 0.82cd |
| CPI 81364 | 34a-c | 103.563a | 98.06ab | 9.62c-f | 4.46ab | 8.12e | 32.43b-d | 0.45b-d | 0.98b-d |
| DL1002 | 34.813a | 100.875a | 97.62ab | 11.28b-d | 4.41b-e | 10.12c-e | 38.93a | 0.46b-d | 0.74d |
| Kikuyu Market | 35.5a | 101.313a | 88.81b | 10.38b-f | 4.03i | 9.56c-e | 32.37c-e | 0.5b-d | 0.75cd |
| Tx-24 | 29.813b-d | 102.125a | 103.5ab | 11.7bc | 4.15g-i | 8.75ed | 34.37a-d | 0.3ed | 0.88cd |
| Q6880B | 33.813a-c | 99.125ab | 95.43ab | 8.4f | 4.18e-i | 11.06b-e | 30.12d-f | 0.43b-e | 0.88cd |
| Eldo-KT-Cream | 33.125a-c | 99.813a | 85.62b | 9.48c-f | 4.19d-i | 11.68b-d | 38.12ab | 0.34c-e | 0.89cd |
| Kikuyu × Meru | 35.563a | 102.25a | 78.62b | 8.75ef | 4.43bc | 14ab | 33.37b-d | 0.58bc | 0.84cd |
| Eldo-KT-Black II | 34.688a | 98.375ab | 90.25ab | 10.81b-e | 4.18e-i | 11.87bc | 34.93a-d | 0.31ed | 0.81cd |
| HA-4 | 29.56cd | 79.06a | 79.75b | 14.26a | 4.36b-g | 12.43bc | 30.81d-f | 0.19e | 0.63d |
| **MSD $_{(0.05)}$** | **4.54** | **8.71** | **28.25** | **0.12** | **0.23** | **3.02** | **5.14** | **0.25** | **0.45** |

*Means in a column followed by the same letters are not significantly different using Tukey's HSD test at $P \leq 0.05$.

## 4.2Dolichos genotypes support bean aphid parasitization by (*Aphidius colemani*) natural enemy

### 4.2.1 Analysis of variance for aphid abundance and number of mummies

Analysis of variance indicated that dolichos genotypes was significant for number of aphids (ANOVA: F $_{(3, 45)}$ = 2.19; *P=0.02*) also statistical difference for number of mummies (ANOVA: F $_{(3, 45)}$ =0.97; *P=0.51*). In addition, there was a significant interaction (ANOVA: F $_{(3, 45)}$ = 3.81; *P<0.0001*) of genotype and crop growth stage for both number of aphids and number of mummies. (Table 4.1).

**Table 4. 7:**Analysis of variance of 4 dolichos genotypes for number of aphid and mummies.

| **Source of variation** | **Df** | **Number of aphids** | **Number of mummies** |
|---|---|---|---|
| Replication | 3 | 7.51 | 7.09 |
| Genotype | 3 | 187327.47*** | 2836.64*** |
| Stage | 3 | 287406.39*** | 15868.51*** |
| Genotype ×Stage | 9 | 27865.19*** | 486.33*** |
| Error | 45 | 7.11 | 3.14 |
| CV | | 1.11 | 3.07 |
| $R^2$ | | 0.99 | 0.99 |

***, significant at $P \leq 0.001$.

### 4.2.2 Influence of dolichos genotypes on aphid populations and parasitization

The number of aphids and mummies recorded was influenced by the dolichos genotypes in the cage exclusion experiment (Figure 4.1). Of the 4 genotypes tested, Kikuyu Market (EU12) registered a greater aphid population (60.4) and mummies (39.7) than genotype Machakos 1 (EU1) which was the most tolerant, exhibiting 142 aphids and 42 mummies, respectively (Figure 4.1).

Aphid infestation was higher at the vegetative and flowering stage compared to the seedling stage and podding stages. Genotype Kikuyu market (EU12) had the highest number of aphids at (627.5) followed by genotype HA-4 which had (515) at vegetative stage. At the flowering stage genotype Brown Rongai showed low number of aphids (84.3) while genotype Machakos 1(EU 1) had the lowest number of mummies (36.5).

When the effect of crop growth stage on the numbers of aphids and mummies was evaluated, it was observed that aphid infestation significantly changed at each stage. Aphids population were significantly higher in the vegetative and flowering stages and significantly lower in the seedling and podding stage. The number of mummified aphids was also observed to follow the same pattern (Figure 3.4).

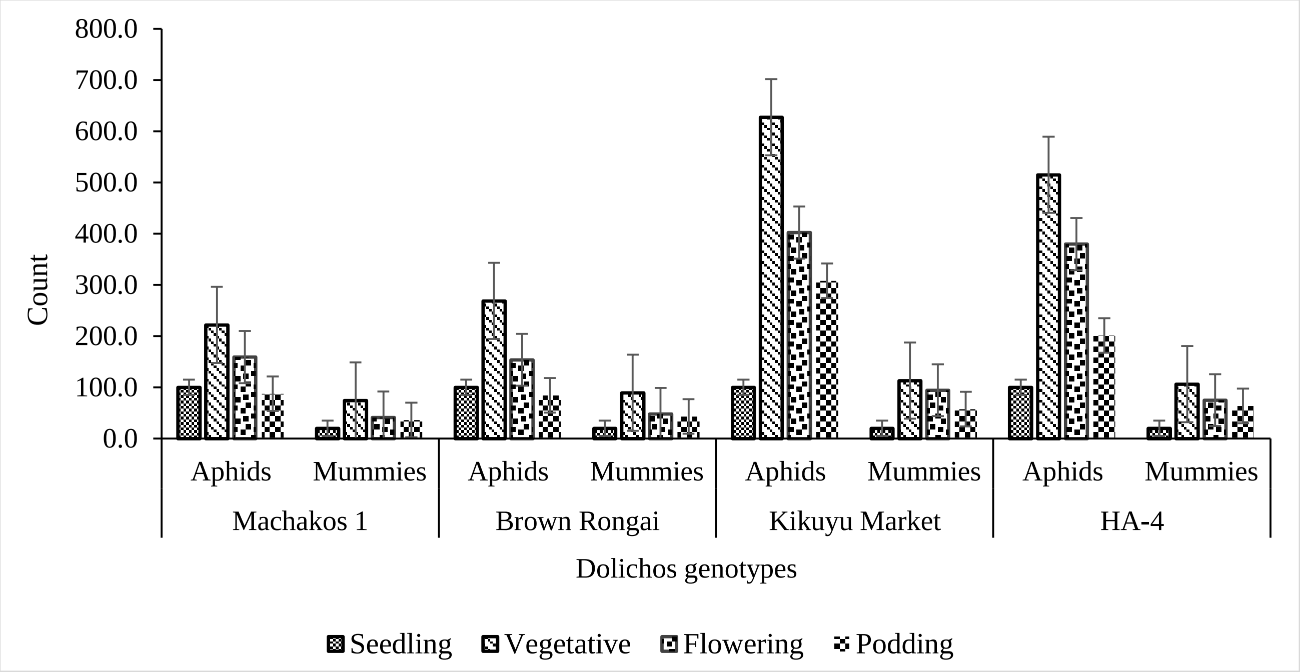

Figure 3. 4: Genotype and crop growth stage interaction effect on number of aphids and mummies (Mean±SE; n=4).

# CHAPTER FIVE

# DISCUSSION

## 5.1 Effect of dolichos genotypes and field margin vegetation on bean aphids' abundance, incidence, severity, and crop yield and yield components of dolichos

The findings of this study revealed significant variations among the dolichos bean genotypes evaluated for aphid infestation levels, natural enemies habitation and yield attributes. Aphid infestation was not linked directly to yield loss in this study, although this has been reported in lablab and other legume crops (El fakhuouri *et al*., 2021).Inclusion of field margin vegetation contributed towards bean aphid management as evidenced by enhanced natural enemy abundance and diversity which varied across the dolichos genotypes and cropping seasons. The variation in the aphid infestation during the 2019 and 2020 cropping seasons can be attributed to genetic differences among the dolichos genotypes and the prevailing environmental factors. Environmental factors such as temperature, rainfall and humidity have been widely reported to influence feeding, reproduction and development of aphids (Mondal *et al*., 2017). Aphid abundance, damage severity and incidence during the 2020 season was higher compared to 2019; this season also experienced slightly higher temperatures (mean of 23.08 ˚C) compared to 2019 which had (22.91˚ C), which can be attributed to the increased aphid populations. Similar findings on the positive correlation between aphid abundance and increasing temperatures have been reported (Karungi *et al*., 2000; Wosula *et al*., 2016).

Dolichos genotypes had slight differences in tolerance to aphid infestation, genotypes Eldo-KT-Cream and Machakos I had high aphid abundance compared to Brown Rongai that had lower abundance. Machakos1 genotype had low grain yield, this can be attributed to high aphid abundance level contrary to Brown Rongai which had high yield and low aphid abundance. These findings are consistent with Mwangi *et al*. (2008) who reported significant reduction of grain yield in common bean due to aphid infestation. Genotypes such Brown Rongai had highly hairy leaves and stems which may have inhibited aphid population build up. Studies by Prado *et al*. (2015) reported that morphological features such as trichomes and waxy layers which reduce insect colonization and feeding. These findings are similar to Chawe (2019) who reported positive correlation between pubescence and aphid population.

Field margin vegetation may play a key role in the conservation of natural enemies through the provision of floral resources such as nectar and pollen (Mkenda *et al*., 2019). Morphological and chemical defenses can also reduce biological control by reducing natural enemy colonization and foraging efficiency (Prado *et al*., 2015). Machakos I and Eldo-KT-

Cream genotypes supported high abundance of hover fly which are key parasitoids of aphids. However, natural enemies have been positively correlated with prey abundance (Östman, 2004). Therefore, in genotype Machakos-I high abundance of hover fly can be attributed to high aphid population which was recorded on the genotypes. Yield performance of dolichos genotypes cannot be solely linked to aphid infestation because of other factors such as rainfall and temperature have been reported to cause significant yield reduction. Average yields were reported in genotypes CPI 81364 and Q6880B with 0.98 and 0.88 t ha$^{-1}$, respectively, similarly, Grotelüschen *et al.* (2014) reported slightly higher yields of the two with 1.4 and 1.9 t ha$^{-1}$, respectively.

Dolichos genotypes showed great differences in plant morphology, ranging from plant height, peduncle length and pod length. Genotype Brown Rongai was the tallest with a high number of peduncles. This led to high production of forage hence high yield. Similar findings were recorded by Parmar *et al.* (2013) who studied the variability in traits of dolichos yield and yield traits and found that genotypes had a positive correlation of yield and yield traits such as peduncle number which was a positive contributor to high number of pods hence high yield. HA-4 and Echo Cream were also found to have high peduncle length and pod length. Echo-Cream as expected would have yielded highly but it recorded the highest biomass which is negatively correlated to yield. Additionally, genotype HA4 had both low yield and low biomass which could be attributed to the genetic makeup of the plant (López-Bellido *et al.*, 2005).

Morphological variation within dolichos genotypes have been attributed to prevailing environmental conditions (Mondal *et al.*, 2017). Temperature conditions, revealed a high variation in observed flowering times from days after planting (DAP), ranging from about 60 to 120 DAP for the different short-season lablab accessions. Genotype CPI-82364 and Q-6880B had a late maturity of 103 and 99 DAP respectively. Similarly, Nord *et al.* (2020) noted that regardless of conditions, accession Q-6880B was found to have delayed development, indicating no photoperiod sensitivity even at higher temperatures. Genotypes that matured early such as HA4 therefore can be said to have high photoperiod sensitivity. Selection of dolichos genotypes that are tolerant bean aphid over different cropping seasons could be a useful step towards utilizing host plant resistance in insect pest management programs. Smallholder farmers in dolichos major growing areas use synthetic insecticides with no information on the potential of natural pest regulation from the natural enemies. However, selection of tolerant genotypes should incorporate farmers' needs to satisfy their needs while increasing the demand of the released genotypes.

The results of this study showed that some level of bean aphids tolerance exist in the dolichos genotypes evaluated. Brown Rongai genotype had qualities such as lower aphid abundance, high peduncle number and high yield. Physiological factors were also a major contributor to aphid abundance and natural enemies. Highly hairy genotypes had lower abundance of aphid abundance. Further research on the specificity of field margin is key as it would lead to a more natural and cost-effective management of aphid in dolichos.

## 5.2 Effect of dolichos genotypes on bean aphid natural enemies (*Aphidius colemani*)

Dolichos genotypes Machakos 1(EU1), Brown Rongai (EU8), Kikuyu Market (EU12) and HA-4 (EU18) were evaluated for their relative tolerance against aphids infestation. All the evaluated genotypes were infested by aphids. However significant differences in colony abundance were recorded by Kamotho *et al*. (2015) in Kenya, and Meena *et al*.(2009). In India, Magalingam (2013) also reported the effect of genotype on differential aphid infestation previously. The variations in infestation rates by genotypes could be explained by genotypic differences in morphological factors like stem diameter. Physiological factors and molecular cues have been reported to drive aphids' host selection and feeding regimes (Smith *et al*., 2014). A characteristic and complex mixtures of plant anatomical and chemical compounds mediate aphid selection of acceptable feeding sites and access to plant sap, the source of aphid nutrients. Similarly, Machakos 1(EU1), and Brown Rongai (EU8) genotypes showed moderate resistance against *Aphis craccivora* (Ngure *et al*., 2021).

To determine the effect of growth stage on aphid infestation, four plant growth stages (seedling, vegetative, flowering and podding) were evaluated. Dolichos genotypes were observed to be susceptible to infestation at all the growth stages but significant differences in number of aphids were recorded. Aphid resistance is manifested by phenotypic variations depending on host plant and aphid species. Scientists who study insect behaviour have distinguished two mechanisms of resistance that affect insects: antixenosis (also known as non-preference), which tends to affect the insects' foraging patterns thereby deterring primary infestation of the crop, and antibiosis, which affects their biotic potential, e.g. growth, development, and reproduction (Dogimont *et al*., 2010). The results of the study concur with findings of other studies where it was observed that aphid infestation in dolichos was significantly affected by plant growth stage and that vegetative stages of the plant were highly susceptible to aphid colonization and colony establishment (Hassan *et al*., 2009; Kamotho *et al*., 2015; Ngure *et al*., 2021;Paikra*et al*., 2019).

The low infestation at seedling stage could be explained by the few numbers of days after planting (DAS). Previous studies have reported that aphid colonization in dolichos is directly correlated to number of days after planting (Hassan *et al*., 2009). That, and the fact that probably the aphids were yet to identify the host. Additionally, cultural practices at seedling stage particularly watering may also have reduced chances of colony establishment and proliferation. Aphid infestation was significantly highest in the vegetative and flowering stages due favorable physiological factors (Boit *et al*., 2018; Mondal, 2021; Sharma *et al*., 2017). This trend is similar to an observation that was recorded by Hassan *et al*.(2009).

The abundance of aphid reduced significantly in the podding stage, this could be attributed to a buildup of secondary metabolites such as, phenolic compounds, usually at high level at these stages and are reported to reduce aphid colony establishment and proliferation (Prado *et al*., 2015). In addition, some secondary metabolites are known to play a key role in chemical defense of plants to insect infestation. Ngenya (2012) reported a decreased number of Russian wheat aphid (*Diuraphis noxia*) in some wheat varieties due to presence of hydroxamic acid.

Evaluation of dolichos genotypes for resistance to aphids' infestation is vital in complementing the conventional laid strategies applied in Integrated Pest Management (IPM). All the four dolichos bean genotypes evaluated were susceptible to aphid colonization. However, aphid colonization and proliferation were significantly influenced by the dolichos bean genotype. It was higher during vegetative and flowering growth stages due to favourable physiological properties of the genotypes under study. Aphid abundance was low in seedling stage and podding stage due to fewer days after planting and buildup of plant defense mechanisms and unfavourable physiological characteristics. Based on the findings, regular assessment and evaluation of dolichos genotypes, cultivars and landraces against aphids is vital for natural pest regulation on dolichos crop in Kenya. Further research should be done involving screening of the varieties for resistance to aphid colonization using biochemical and molecular methods.

# CHAPTER SIX

# CONCLUSION AND RECOMMENDATIONS

## 6.1 Conclusions

From the findings of this study the following conclusions were made:

i. Dolichos genotypes tolerant to infestation by *Aphis fabae* has the potential to suppress bean aphids and achieve higher grain yield of dolichos.

ii. Field margin vegetation around a dolichos bean crop has the potential to support natural enemies of bean aphids which can contribute to natural pest regulation.

iii. The dolichos genotypes evaluated showed significant variation on growth yield and yield components planted either in presence or absence of field margin vegetation.

## 6.2 Recommendations

i. It is recommended that smallholder farmers adopt bean aphid tolerant/resistant dolichos genotypes with field margin vegetation for environmentally-friendly management of bean aphids and conservation of aphid natural enemies in dolichos production.

ii. Further studies on screening of dolichos genotypes and multipurpose flowering plants that can support natural enemies for conservation biological control is recommended.

iii. This study recommends further evaluation for the promising dolichos genotypes with increased growth vigour, increased yield and biomass to boost dolichos production.

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# APPENDICES

## Appendix A:Combined analysis of variance of 18 dolichos genotypes in the presence or absence of field margin vegetation for aphid abundance, damage severity and percent incidence in 2019 and 2020.

| Source of variation | df | Aphid abundance | Damage Severity | Percent incidence |
|---|---|---|---|---|
| Season | 1 | 459.17*** | 371.28*** | 1067653.78*** |
| Growth stage (Stage) | 3 | 43.04*** | 1.66*** | 84921.09*** |
| Season×Stage | 3 | 124.36*** | 0.95*** | 22765.02*** |
| Margin | 1 | 0.99** | 0.68* | 771.13 |
| Season×Margin | 1 | 2.16*** | 2.00*** | 518.93 |
| Stage×Margin | 3 | 0.30 | 0.21 | 142.44 |
| Season×Stage×Margin | 3 | 0.18 | 0.15 | 97.06 |
| Genotype | 17 | 0.53*** | 0.42*** | 3587.37*** |
| Season×Genotype | 17 | 0.21 | 0.12 | 1800.07*** |
| Stage×Genotype | 51 | 0.04 | 0.09 | 220.89 |
| Margin×Genotype | 17 | 0.59*** | 0.26 | 772.03*** |
| Season×Stage×Genotype | 51 | 0.07 | 0.08 | 219.04 |
| Season×Stage×Margin×Genotype | 119 | 0.10 | 0.09 | 214.11 |
| Replication | 3 | 3.84*** | 2.03*** | 9075.02*** |
| Error | 861 | 0.15 | 0.17 | 314.40 |
| **CV (%)** | | **20.32** | **25.28** | **30.10** |
| **$R^2$** | | **0.89** | **0.74** | **0.85** |

*, **, *** significance at P≤0.05, P≤0.01 and P≤0.001, respectively.

**Appendix B:Analysis of variance for species richness, abundance and diversity index on dolichos genotypes in 2019 and 2020 cropping seasons.**

| Source of variation | df | Richness | Abundance | Diversity |
|---|---|---|---|---|
| Season | 1 | 5.28*** | 206.31*** | 2.20*** |
| Genotype | 17 | 0.59* | 15.13 | 0.00 |
| Season×Genotype | 17 | 1.03*** | 11.64 | 0.00 |
| Margin vegetation | 1 | 0.22 | 38.86 | 0.00 |
| Season×Margin | 1 | 1.38* | 0.80 | 0.01* |
| Trapping area | 1 | 45.92*** | 900.43*** | 0.04*** |
| Season× Trapping area | 1 | 14.67*** | 5.07 | 0.00 |
| Genotype× Trapping area | 17 | 0.59* | 9.83 | 0.00 |
| Season×Genotype×Trapping area | 17 | 0.73* | 16.17 | 0.00* |
| Error | 34 | 0.27 | 15.64 | 0.00 |
| CV | | 13.22 | 32.50 | 11.75 |
| $R^2$ | | 0.93 | 0.84 | 0.97 |

*, **, *** significance at P≤0.05, P≤0.01 and P≤0.001, respectively.

**Appendix C**: **Combined analysis of variance of 18 dolichos genotypes in the presence or absence of field margin vegetation for yield components in 2019 and 2020.**

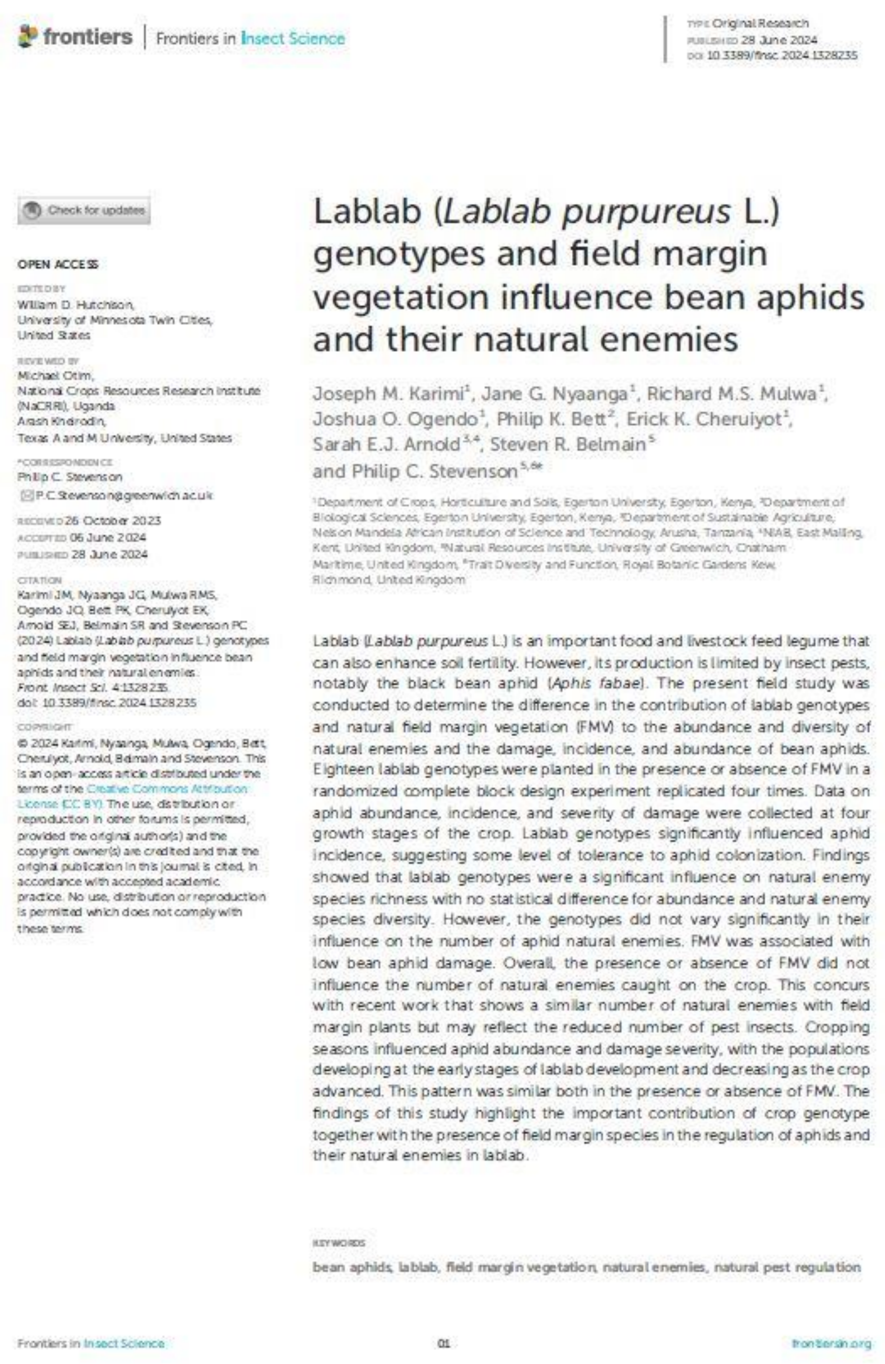
| Source of variation | df | Stand count | Days to 50% flowering | Plant height (cm) | Peduncle length(cm) | No. of peduncle | Pod Length(cm) | No. of pods/plant | Yield (t/ha) | Biomass (t/ha) |
|---|---|---|---|---|---|---|---|---|---|---|
| Season | 1 | 1870.68*** | 9.38 | 348.92 | 0.00 | 0.00 | 0.00 | 1237.53*** | 0.03*** | 0.06*** |
| Margin | 1 | 64.22* | 5.01 | 36.83 | 0.07 | 0.01 | 0.04 | 31.33 | 0.00 | 0.00 |
| Season×Margin | 1 | 55.12* | 6.72 | 92.25 | 0.07 | 0.01 | 0.04 | 31.33 | 0.00 | 0.00 |
| Genotype (G) | 17 | 83.42*** | 512.05*** | 1355.64*** | 39.43*** | 0.08*** | 0.39*** | 192.30*** | 0.01*** | 0.01*** |
| Season×Genotype | 17 | 32.02** | 153.64*** | 519.31 | 35.90*** | 0.07*** | 0.28*** | 153.53*** | 0.00*** | 0.00*** |
| Margin×Genotype | 17 | 12.18 | 26.63 | 218.73 | 6.01* | 0.01 | 0.04 | 12.03 | 0.00 | 0.00 |
| Season×Margin×G | 17 | 14.60 | 27.02 | 418.17 | 6.01* | 0.01 | 0.04 | 12.03 | 0.00 | 0.00 |
| Replication | 3 | 573.81*** | 48.33 | 438.89 | 0.00 | 0.00 | 0.00 | 82.79** | 0.01*** | 0.00** |
| Error | 213 | 13.22 | 48.65 | 511.69 | 3.56 | 0.01 | 0.04 | 16.95 | 0.00 | 0.00 |
| **CV(%)** | | 10.83 | 7.07 | 24.22 | 18.54 | 8.76 | 4.37 | 12.69 | 9.03 | 11.05 |
| **$R^2$** | | 0.68 | 0.54 | 0.29 | 0.66 | 0.64 | 0.63 | 0.68 | 0.61 | 0.53 |

*, **, *** significance at P≤0.05, P≤0.01 and P≤0.001, respectively.

## Appendix D:Author's Own Publications

*a) Publications in refereed scientific journals*

**Karimi, J. M.,** Nyaanga, J. G., Mulwa, R. M., Ogendo, J. O., Bett, P. K., Cheruiyot, E. K., ... & Stevenson, P. C. (2024). Lablab *(Lablab purpureus* L.) genotypes and field margin vegetation influence bean aphids and their natural enemies. *Frontiers in Insect Science*, *4*(6), 1328235.



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Karimi JM, Nyaanga JG, Mulwa RMS, Ogendo JO, Bett PK, Cheruiyot EK, Arnold SEJ, Belmain SR and Stevenson PC (2024) Lablab (*Lablab purpureus* L.) genotypes and field margin vegetation influence bean aphids and their natural enemies.
*Front. Insect Sci.* 4:1328235.
doi: 10.3389/finsc.2024.1328235

### Lablab (*Lablab purpureus* L.) genotypes and field margin vegetation influence bean aphids and their natural enemies

Joseph M. Karimi[1], Jane G. Nyaanga[1], Richard M.S. Mulwa[1], Joshua O. Ogendo[1], Philip K. Bett[2], Erick K. Cheruiyot[1], Sarah E.J. Arnold[3,4], Steven R. Belmain[5] and Philip C. Stevenson[5,6*]

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Lablab (*Lablab purpureus* L.) is an important food and livestock feed legume that can also enhance soil fertility. However, its production is limited by insect pests, notably the black bean aphid (*Aphis fabae*). The present field study was conducted to determine the difference in the contribution of lablab genotypes and natural field margin vegetation (FMV) to the abundance and diversity of natural enemies and the damage, incidence, and abundance of bean aphids. Eighteen lablab genotypes were planted in the presence or absence of FMV in a randomized complete block design experiment replicated four times. Data on aphid abundance, incidence, and severity of damage were collected at four growth stages of the crop. Lablab genotypes significantly influenced aphid incidence, suggesting some level of tolerance to aphid colonization. Findings showed that lablab genotypes were a significant influence on natural enemy species richness with no statistical difference for abundance and natural enemy species diversity. However, the genotypes did not vary significantly in their influence on the number of aphid natural enemies. FMV was associated with low bean aphid damage. Overall, the presence or absence of FMV did not influence the number of natural enemies caught on the crop. This concurs with recent work that shows a similar number of natural enemies with field margin plants but may reflect the reduced number of pest insects. Cropping seasons influenced aphid abundance and damage severity, with the populations developing at the early stages of lablab development and decreasing as the crop advanced. This pattern was similar both in the presence or absence of FMV. The findings of this study highlight the important contribution of crop genotype together with the presence of field margin species in the regulation of aphids and their natural enemies in lablab.

KEYWORDS
bean aphids, lablab, field margin vegetation, natural enemies, natural pest regulation

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*b) Publications in refereed conference proceedings*

**Karimi, J. M.,** Mulwa, R.S., Nyaanga J.G., Ogendo, J.O., Cheruiyot, E., & Bett, P.K (2020). Effect of Dolichos*(Lablab purpureus L.)*genotypes and field margin species on bean aphids' population and their natural enemies. *Book of abstracts*, Proc. 13th Biennial Egerton University International Virtual Conference: Innovation, Research and Transformation for Sustainable Development, 24th – 26th November 2020. pp.67

# Effect of Dolichos (*Lablab purpureus* L.) Genotypes and Field Margin Species on Bean Aphids' Population and their Natural Enemies

***J. M. Karimi*[1]*, R. M.S. Mulwa*[1]*, J. G. Nyaanga*[1]*, J. O. Ogendo*[1]**,***
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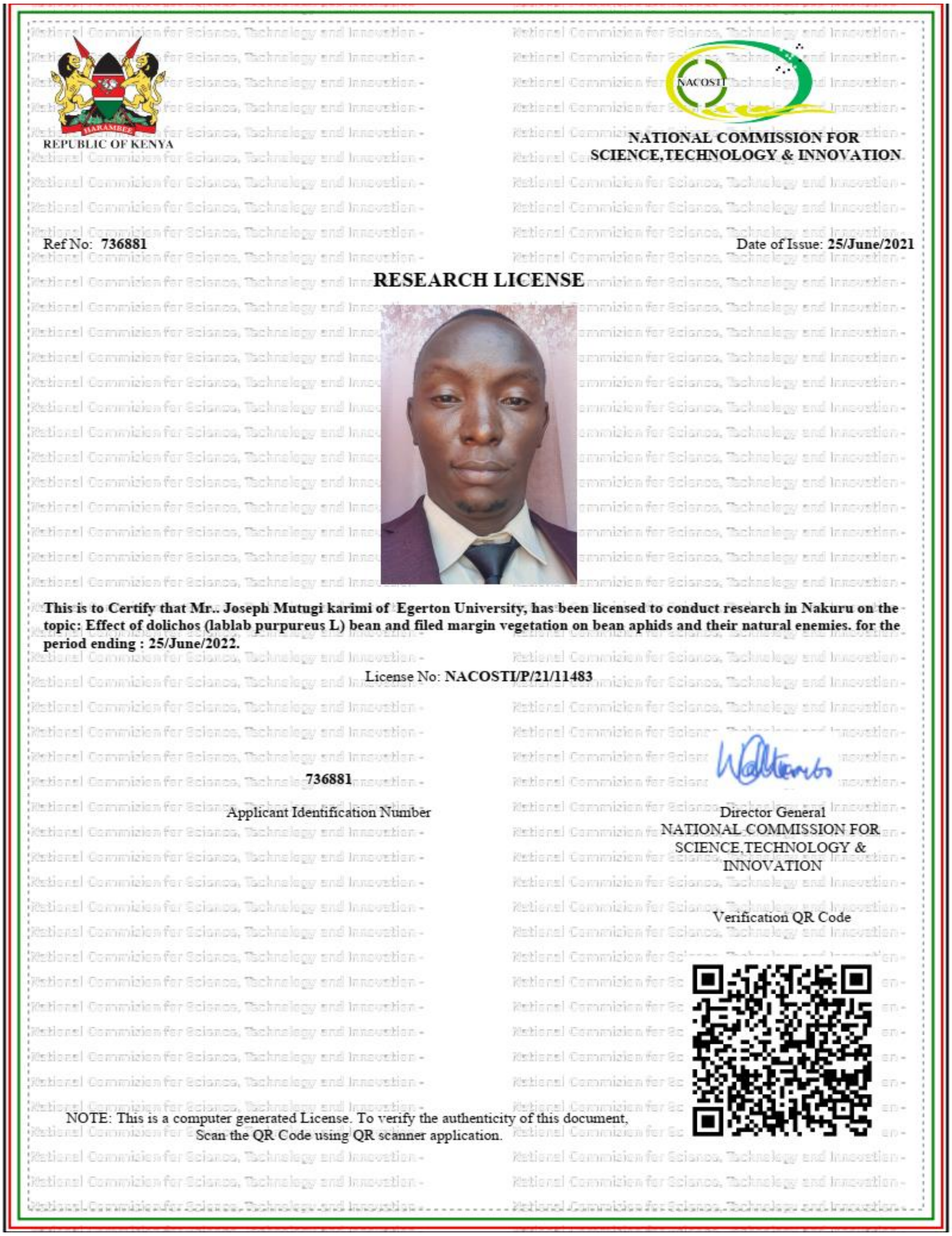
Plant genotypes vary in influencing predatory activity and suppression of population development of aphids. Host plant resistance plays a key role in managing pests and conserving of natural enemies in an agroecosystem. Field evaluations were conducted to determine the effect of various genotypes on abundance of aphids and their natural enemies in Dolichos bean (*Lablab purpureus* L.). Eighteen (18) genotypes were planted at Egerton University, with and without the field margin vegetation, in a randomized complete block design (RCBD) replicated four times. Data on abundance, severity and incidence of aphids were collected at five different crop growth stages: seedling, early vegetative, late vegetative, flowering and podding. Population of natural enemies was monitored and trapping was done by use of pan traps and sticky traps. Data on counts were transformed using log transformation $\log_{10}(x+2)$ before being subjected to analysis of variance using **PROC GLM** in SAS software and treatment means separated using **Tukey's** Honest Significance Difference test ***($P \leq 0.05$)***. Results showed that aphid incidence was significantly different at seedling and late vegetative stages across the genotypes. Genotypes EUD12 and EUD1 had the highest aphid incidences of 36.25% and 35.00%, respectively, at seedling stage as well as 10.0% and 3.75%, respectively, at late vegetative stage. Ten families of natural enemies were identified with Tachnidae having the highest proportion (52.97%) followed by Braconidae (22.68%), Coccinellidae (10.32%) and Syrphidae (4.48%) in decreasing order. Population of natural enemies was significantly different within the crops (81.7%) and along the field margin vegetation (18.3%). Specifically, 88% tachnids, 81% braconids, 62% coccinellids and 85% syriphids were found within the crops as compared to the field margins (12-18%). These findings show that genotype and field margin vegetation directly influence a sustainable natural pest regulation system in Dolichos bean.

**Keywords***:* aphids, Dolichos, genotypes, field margin vegetation, natural enemies.

## Appendix E:Research Permit



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